Membrane Module Instructions

Introduction

This module is going to explore Brownian motion, diffusion, and the hydrophobic effect. The topics we are going to investigate form the foundations for an enormous number of biological processes. We will explore these topics through simulations of water and phospholipids. Phospholipids form the membranes that a cell uses to separate its surroundings from its interior. We will show you computer simulations of these molecules, which can be thought of as realistic movies that demonstrate their motion as it would occur in nature based on our understanding of physics and the interactions between the molecules.

Procedure

The entirety of today's lesson will be contained in a plugin for VMD we have written and named VMD Lite. The VMD Lite lesson will guide you through all of the visualizations we have prepared.

1. Open VMD Lite by typing vmdlite into the prompt on a TkConsole window within VMD, and pressing Enter. ← are we just distributing a version that auto loads VMD Lite? The tkcon is unnecessary if we can help it.
2. Click "Lesson" on the welcome window that appears
3. Select the lesson named "Phospholipids" on the right side, then click "Load Phospholipids"

Slide 1: Brownian Motion

4. Immediately after loading the lesson you should see a number of spheres inside a cubic volume in the Display window. These spheres are representations of water molecules. Try moving the view around by clicking and dragging in the screen. You can also zoom in by using the scroll wheel on the mouse.
5. Drag the bar, or press "Play" to watch the simulation of the water, and notice that the molecules move very quickly. **How fast do you think water moves?**
   a. Let's calculate the speed of the pink water molecule using its change in position between two frames.
   b. Pick any frame and write down the coordinate value. You should see it above the "Exit" button in the Module window.

c. Click the "Next frame" and write down the coordinates again.
d. Use the distance formula (below) to calculate the distance that the water molecule traveled. Note that the coordinates are given in Å (10^{-10} m).

$$\left(\sqrt{(x_1 - x_2)^2 + (y_1 - y_2)^2 + (z_1 - z_2)^2}\right)^2$$

e. The time between frames is 20 ps (10^{-12} s). Use this to calculate the speed of the molecule. (speed=distance/time) How does your calculated answer compare with your guess from before?
f. The cruising speed of a Boeing 747 is 245 m/s. How fast is your water molecule moving in relation to this?

The type of movement you just witnessed is called **Brownian motion**. This random motion of molecules arises from their **kinetic energy**, which is measured by **temperature**. Because this energy is very large at the molecular scale, it creates an incredibly chaotic environment with molecules constantly bombarding one another at several times the speed of a commercial airliner.

6. Click "Next" (slide 2) to proceed to the next section.

Slides 2-7: Hydrophobic Effect

In VMD Lite, you should now see a graphical representation of a phospholipid bilayer, also known as a membrane. As mentioned above, this type of structure encloses the entirety of the cell as well as various compartments inside. Considering such a highly ordered structure is assembled in the chaotic environment we explored in the last section, ***what fraction of a cell's energy do you think the cell expends to build it?***

7. Click "Next" (slide 3) to view a zoomed in representation of an individual phospholipid molecule. Click "Next" (slides 4-5) twice more to observe and read about the head and tail sections of the lipid, which are polar and non-polar, respectively.
8. Click "Next" (slide 6) once more.

Here we have prepared a system composed of randomly oriented phospholipid molecules and water molecules (note that the system uses periodic boundary conditions to mimic the conditions of a much larger environment); we are going to simulate their dynamics. ***Considering the chaotic environment we observed in the water box, what do you think will happen?***

9. Take some time to consider the question above, and discuss your prediction with your lab partner.

10. Click the play button to watch the simulation. ***What do you observe? Does the result match your earlier prediction?***

We want to be extremely clear on the following point: **we did not put any additional energy into this system**. The sort of spontaneous self-assembly you saw here happens in nature all the time due to nothing but the physical interactions between the molecules. You might find it interesting that what you are seeing here occurred in 100ns, which is about 1/1000 of the time it takes to blink your eyes.

11. If we didn't put any energy into moving the phospholipids around, ***how do you think they got to where they are and formed the structure you see?*** Discuss your thoughts with your lab partner.

12. Think about the possible important interactions that might exists among the different molecules. ***Write down a model that explains the behavior you observe.***

13. Click "Next" (slide 7) to advance the lesson.

14. You should now see several buttons to select simulations we have prepared in which we have removed one or two elements of the membrane system you just observed, such as water, lipid tails, or lipid headgroups. Before playing any of the simulations, ***make a prediction as to what will happen for each using your earlier model, and write it down***.

15. Click to select and play each of the simulations. Before continuing onto each subsequent simulation, discuss the following questions: ***What are the similarities and differences of each with respect to the initial membrane simulation? Are the predictions you made correct?***

16. After watching the missing-element simulations, ***develop a new model which explains what you observe in these simulations, as well as in the original membrane simulation.***

Concluding remarks

A few of the things we hope you learned in this module are

- Molecules in the cell move around really fast, making the environment chaotic.
- This fast motion helps molecules that want to interact find each other quickly.
- Polar and non-polar interactions produce the spontaneous assembly of membranes, as well as other structures in biology; this is known as the ***hydrophobic effect.***
- Non-polar (hydrophobic) regions do not aggregate because they have strong interactions with each other – if they did, the simulation of lipid molecules in vacuum would not have had the tails pointing out. In short, the hydrophobic effect has nothing to do with the fact that tails are attracted to tails. Instead, it arises because water molecules would rather interact with each other than the tails, and as a consequence effectively "push" all the tails together to the inside of the membrane. But since the lipid also has a polar part that does want to interact with water (the headgroup), it is placed on the water-facing of the membrane. (The subtle nature of the hydrophobic effect is actually commonly misunderstood in the life science research community as a whole, so if this concept is a little surprising to you, don't worry.)