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Effect of temperature changes on the cell membrane with molecular dynamics simulation

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ABSTRACT

The cell membrane is an important component of the cell and has a significant role in many cellular processes such as cell proliferation, adhesion, migration and signal detection. Therefore, studding the cell membrane and its characteristics has a crucial role in a thorough understanding of the cell functions. On the other hand, studding the interactions in biological systems such as membrane at the molecular levels is extremely important and since the empirical methods and their results cannot provide accurate and sufficient information in molecular scale, molecular simulations have recently attracted a lot of attention among biologists. Molecular simulation presents quantitative and qualitative information about the interaction of membrane molecules during environment changes, as well as provides a test bed for evaluating the available assumptions about cell membrane. In this research, cell membrane properties such as density, chain order and permeability were studied by applying the changes in temperature and evaluating the thermal motion of the lipid molecules using the molecular dynamics. The results indicate the importance of temperature variations in the function of the membrane, particularly in selective permeability. It also may cause cellular damage when the temperature is out of especial range.

Keywords: *phospholipid, cell membranes, molecular dynamics simulation, GROMACS, temperature changes.*

1. INTRODUCTION

The biological membrane is one of the important components of the cells, which is the first part placed against external factors. The interaction of membrane components with these factors can determine the behavior of the cell [1-3]. Accordingly, many cell functions are associated with the membrane, such as migration, cell proliferation, adhesion, and signal reception [4].

Regardless of the membrane type, all cell membranes composed of lipids and proteins and have a common structure. The lipid part contains millions of lipid molecules that form two adjacent sheets called a lipid bilayer [5-7]. The lipid bilayer is the basis of membrane structure and it can act as a permeable barrier for most water-soluble molecules. On the other hand, protein molecules mediate many membrane functions and caused difference in membranes [8, 9].

In tissue engineering and biology, understanding of the membrane especially in molecular scale is incredibly important. The cell function and behavior become different in response to alteration in cell environment. These changes in cell behavior are caused by varying the performance of its components like the cell membrane. Alteration in the performance of membrane as the protective part is extremely important [10, 11]. Understanding

how cells behave during environmental condition changes, such as temperature and pressure, at the molecular scale, is necessary for implements of therapeutic and biological purposes [12-15]. In spite of various experiments designed to study the behavior of the cell membrane, accurate information about the molecular scale has not been available yet. Therefore, computer simulation techniques have recently attracted a lot of attention from researchers [8, 9].

Molecular simulation is a useful tool for studying and understanding the behavior of biological systems. The quantitative prediction of biophysical and biochemical processes partly becomes possible using molecular simulation [16-20]. Because of the widespread use of molecular dynamics methods and development of computers and computational techniques, special software have been designed for molecular dynamics simulation [21].

In the present work, molecular dynamics simulation was performed to investigate the behavior of the membrane phospholipid bilayer during temperature changes, using the GROMACS software. Since Palmitoyloleoylphosphatidylcholine (POPC) is the most abundant lipid in animal cell membranes [22], lipid bilayer containing these molecules was considered as the simulation system.

2. SIMULATION METHOD SECTION

In every simulation step, the system consists of a lipid bilayer of POPC molecules in water. CHARMM27 and SPC force field were used to create membrane structure with VMD software. The created lipid bilayer and the structure of phospholipid molecules (POPC) are shown in Figure 1.

For energy minimization step, Steepest Descent method was used. Leap Frog algorithm was the chosen method for NVT and NPT equilibration step, as well as the implementation of

molecular dynamics. The system was investigated under two approaches of decrease and increase of temperature via a step-by-step procedure. In this simulation, the desired temperature changes were applied to the system with coding in Linux environment. In the first approach, increasing the temperature of the POPC lipid bilayer which was placed in a solution of water molecules was studied. Initial temperature of system was considered above the transition temperature of POPC molecules and at any md run, the

temperature was increased 10°C. In the second approach, reducing the temperature on the lipid bilayer was studied. In the second approach, reducing the temperature on the lipid bilayer was

studied. So that the temperature of the 300 K is started and has 10°C decrease in temperature at every molecular dynamics run.
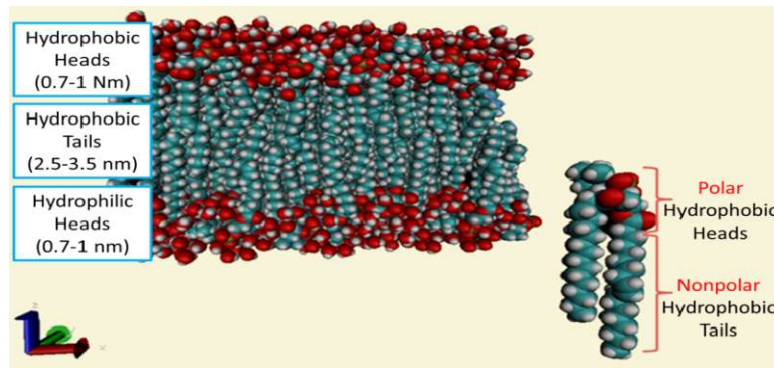

Figure 1. The lipid bilayer and the structure of the phospholipid molecules (POPC).

3. RESULTS SECTION

By evaluating the effect of temperature on the lipid bilayer systems, the most important physical properties of the membrane (density, diffusion coefficient, chain order parameter and the number of hydrogen bonds formed in the system) were investigated and analyzed using two different simulations which are mentioned in the previous section. Simulation results will be discussed later in this report.
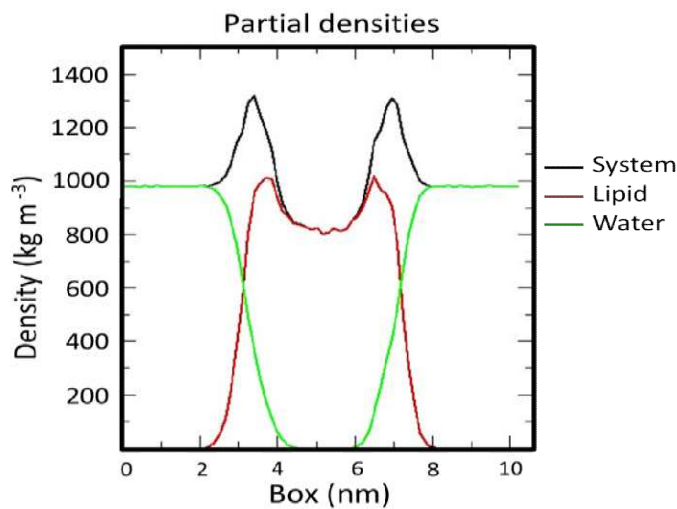

Figure 2. Distribution of the mass density of the simulated system, water molecules and lipid molecules (POPC).

As the first property, density curve is shown in Figure 2 for POPC membranes in the ground state. The outer surface of the lipid bilayer has the highest density of mass in the system and is located in a distance of 1.5 to 2 Nano meter from the center of the bilayer, which includes the region of the lipid head and water. Density of system from its maximum amount in the region of the lipids, by a factor of about 2, in the center of the bilayer has decreased which is in accordance with experimental results and similar results have already been reported in the literature [23, 24]. Aqueous phase refers to a region with density around 3970 kg/m^3 and 3nm distance from the lipid center. As shown in Figure 2

water molecules penetrate the membrane. This penetration is due to the polar heads of the lipid molecule which exist in this area.
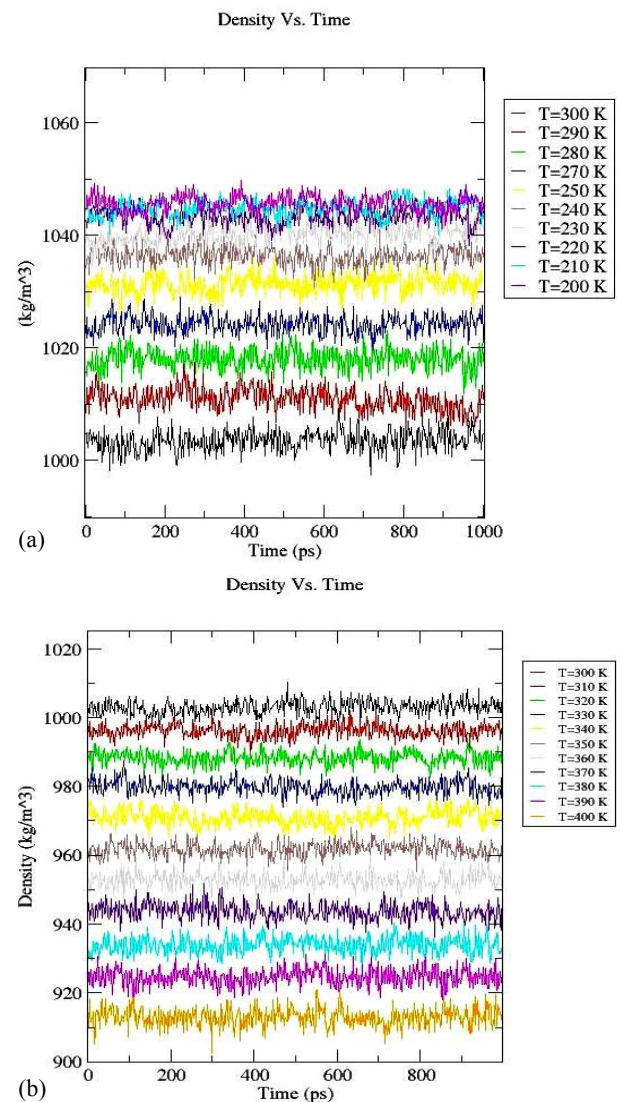

Figure 3. The mass density for POPC lipid bilayer membrane with (a) reducing and (b) increasing the temperature for 10 ° step case.

Only about 2 nm of the membrane is protected against water atoms. It is due to the hydrocarbon tail of lipids which is hydrophobic. According to the diagram, the thickness of the lipid bilayer is about 5.5nm.

The second investigated feature in this simulation is the density variations of the system during the temperature changes. During the simulation, the system pressure was constant (1 bar). Since the density of the fluids is a function of temperature and pressure, the observed density variations is related to temperature changes. Mass of the system is constant hence process of density variations is related to the volume of the system ($\rho = \frac{M}{V}$).

As shown in Figure 3(a), increasing the temperature leads to a decrease in the density of the system. Because of temperature rising, volume increases. (In order to observe changes, temperature range is considered large.)

(a)

(b)
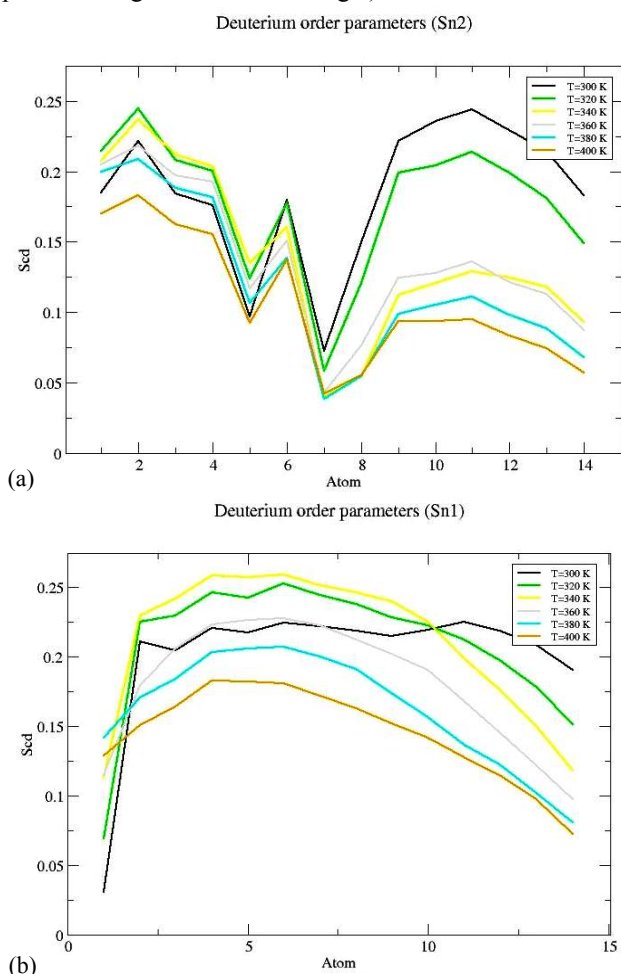
Figure 4. The chain order parameter (a) Sn2 and (b) Sn1 under increasing temperature for 10 ° step case.

In figure 3(b), by decreasing temperature, we see an increase in density but at temperatures less than 270 K, density decreases. This difference is due to change in density and different behavior of the water after 277 K (4°C). (After 4°C water density decreases with decreasing temperature.). Because in the system, lipid molecules are located besides the water molecules, the results are behavior resultant of the two fluids.

Order parameter is third properties studied in this paper. Average orientation of the lipid bilayer hydrocarbon chains versus the bilayer normal vector is considered as order parameter.

In the Figure 4, order parameter of two lipid chains can be observed in different temperatures. According to the charts, as the temperature becomes higher, irregularities increase and curves are shifted towards lower values. By decreasing temperature, broken of curve becomes greater and this means that direction of bonds which are located next to each other is different. This helps that the chain in terms of space adoption are located in a zigzag mode and the entire chain is placed in the normal direction of the membrane and regularity of the system has increase.

Another investigated parameter is the number of hydrogen bonds which are formed in the system. Hydrogen bonds between water molecules occur with each other and with the membrane lipids. By increasing the temperature, the movement of the molecules becomes greater and the number of hydrogen bonds reduces, because the molecules do not have sufficient time to form hydrogen bonds (Figure 5(b)).

As shown in Figure 5(a), decreasing in temperature is caused increasing the number of hydrogen bonds. Decreasing the temperature is caused decreasing the rate of molecules movement, so the molecules have more opportunity to find the appropriate location of each other and form a hydrogen bond. After the temperature of 4°C, the water density decreases again and the distance between the molecules becomes greater thus the number of bonds reduce. For this reason, at temperatures less than 288°K, too little difference between the figures is observed and effect of the presence water in the system can be observed too.

(a)

(b)
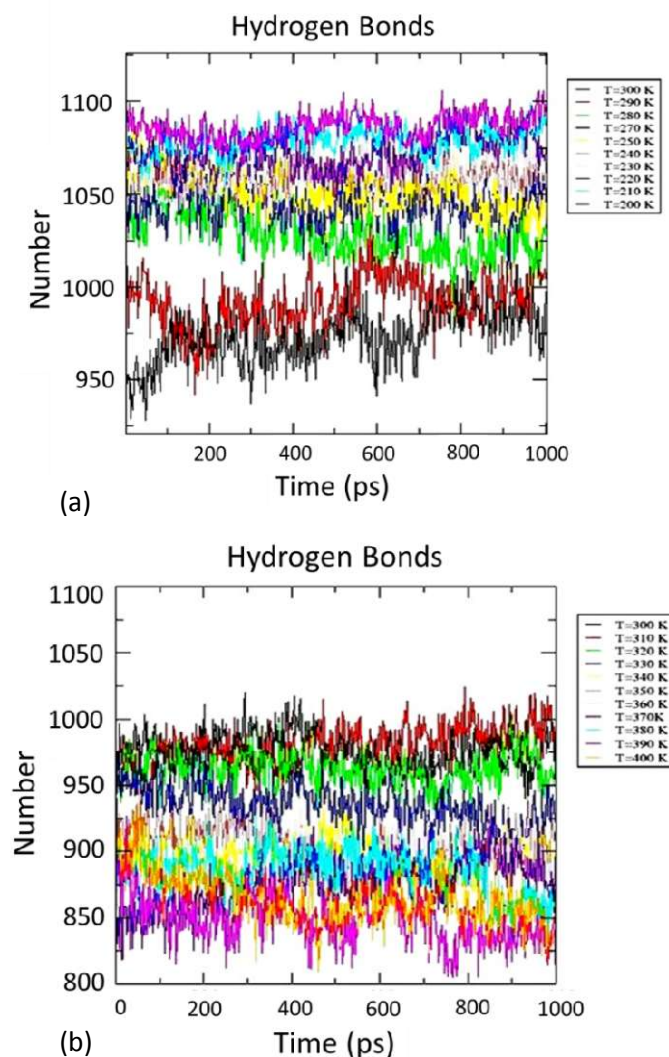
Figure 5. Changes in the number of hydrogen bonds in the system under the conditions (a) reduction and (b) an increase in temperature for 10 ° step case.

(a)

(b)
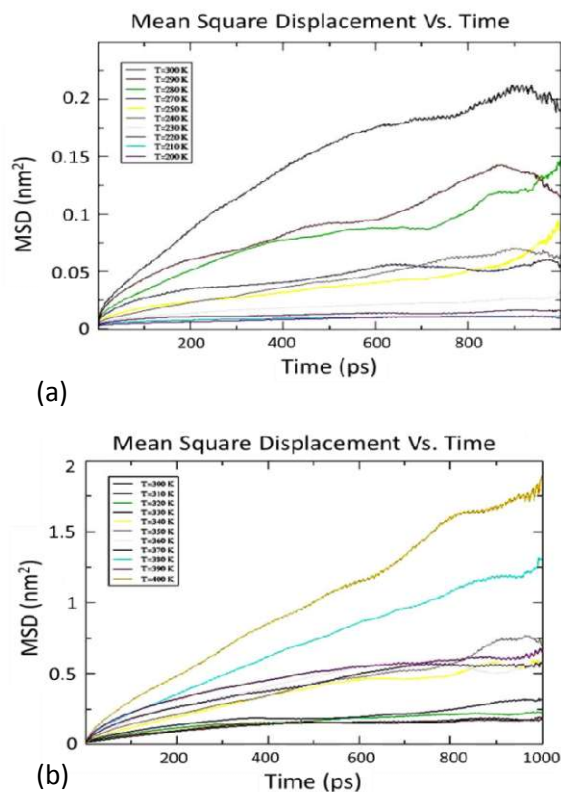
Figure 6. Changes in mean square displacements for lipid molecules under the conditions (a) reduction and (b) an increase in temperature for 10 ° step case.

The last feature is the study of changes in diffusion coefficients during temperature changes. Details of the lipid bilayer dynamic play important role in the passive transfer of small molecules in the cell membrane. In the simulations, penetration of lipid bilayer molecules is calculated by using the slope of the mean square displacement (MSD) versus time.

In accordance with the Figure 6(b) is observed that MSD increases with increasing the temperature, which indicates an increase in the diffusion coefficient. In fact temperature is caused increasing energy of the molecules and thus increasing their mobility. Ultimately the more mobility of molecules causes higher diffusion. On the other hand, as previously explained an increase in temperature leads to a decrease in the density of the fluid which in turn affects increasing the amount of particles diffusion because it reduces the inhibitions of space available in the way of the particle. Also this point should be noted that as increasing temperature increases the mobility of the particles increases the mobility other particulates in the environment as well. Resulting in the high mobility of the environment particles is caused increasing the probability of particle collisions with them, these collisions are according to the direction of collision, may have positive or negative effect on particle diffusion.

These results of the simulation are consistent with the laws of physics and thermodynamic which shows that the simulation process is correct.

4. CONCLUSIONS

The purpose of this study was to investigate the lipid bilayer membrane characteristics and physical properties under various temperatures using molecular dynamics simulations. At physiological temperature, the cells act properly. When the temperature rises, Fatty acid tails of the cell membrane have more mobility and the phospholipid bilayer becomes more fluid. These end up change in the permeability of the cell so some unallowed molecules enter the cell and cause some damages.

Also reducing the temperature has influence on the cells and their membranes. At lower temperature, fatty acid tails of the phospholipids become more rigid and tough and this affects the permeability and cells ability to live.

By decreasing the temperature, the number of hydrogen bonds in the membrane increases and the membrane becomes more rigid and less flexible. These factors lead to a decrease in membrane fluidity and an increase in the order of hydrocarbon chains. So chains will have more regular arrangement. When fluidity of cell decreases, movement and growth of cells could be difficult. Another effect of temperature fall on the membrane was altering the system permeability. At lower temperatures, membrane permeability decreases. This change prevents from entrance of the vital molecules into cells.

The survey results can be used in drug delivery applications by using thermotherapy and also is helpful for studying the process of cell freezing. On the other hand, we can use this information to identify cancer cells. Environment of the cancer cells in comparison with the natural cells has higher temperature. Consequently the results about high temperatures described in this paper can be used to identify cancer cells. Also results of this study can be used to transfer genes, drugs, nanoparticles and etc. into the cells. The thermal motion of lipids changes the orientation of the lipid molecules in the membrane and suggests the initial defects in membrane. These defects are suitable places for the penetration of molecules into cells and accelerate the transfer of the desired component.

5. REFERENCES

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6. ACKNOWLEDGEMENTS