In Silico Design and Molecular Dynamics Simulation of Glycophorin A Transmembrane Dimer, in Pure DPPC and Mixed DPPC-DMPC Phospholipid Bilayers

Delara Mohammad-Aghaie[*], *Fatemeh Razavi*

Department of Chemistry, Shiraz University of Technology, Shiraz, 71555-313, IRAN

Abstract

Molecular Dynamics (MD) simulations play a key role in realizing the structure/function relationships of membrane proteins and their interactions with the phospholipid bilayer membranes. This study aims to simulate the pure Dipalmitoyl Phosphatidylcholine (DPPC) bilayer and specify how this system responds to 1) the insertion of Dimyristoyl Phosphatidylcholine (DMPC) molecules, and 2) the simultaneous presence of DMPC molecules and Glycophorin A transmembrane. Different structural properties of the lipid and protein molecules are employed to address these questions.

Keywords: Glycophorin A, Membrane Protein, Molecular Dynamics Simulation, Phospholipid Bilayer

I. Introduction

In recent years, molecular dynamics simulations have become increasingly valuable for understanding the membrane proteins function, as well as their interactions with, and dynamic behavior in bilayer membranes. [1]

Glycophorin A (GpA), is the major-intrinsic sialoglycoprotein of human erythrocyte membranes. MacKenzie et. al. [2] have determined the dimeric structure of GpA trans membrane (GpAtm), a peptide corresponding to residues 62 to 101 of human GpA, by nuclear magnetic resonance (NMR) method.

In this study, we are going to perform molecular dynamics simulations on the phospholipd bilayer membranes, with and without integral protein, which is GpAtm here. Lipid bilayers are home to membrane proteins, therefore their accurate modeling is very crucial in computational biology and chemistry. Membuilder [3] is a web server which prepares the initial configuration of a membrane model, composed of different phospholipid molecules, based on both united-atom and all-atom force fields. This survey considers MD simulations of three distinct systems: Pure DPPC bilayer with 128 molecules, Mixed phospholipid bilayer (DPPC 60% - DMPC 40%) and finally the mentioned mixed bilayer, while GpAtm is inserted in it, as an integral protein. This way we can follow how the presence of DMPC molecules affect the structure of pure DPPC bilayer and later, how trans membrane part of Glycophorin A interacts with the mixed bilayer, and the mutual effects of lipid and protein molecules on each other. Root Mean Square Deviation (RMSD), Radial Distribution Function (RDF), Deuterium Order parameter (DOP), Area Per Lipid (APL) and Bilayer Thickness are among the structural properties, which are analyzed here.

II. Methodology

The initial structures of both pure DPPC and mixed (DPPC 60%-DMPC 40%) phospholipid bilayers were generated by employing the Membuilder server. Resulting structures were solvated in tip3p water molecules and neutralized with adequate number of sodium and chloride ions. Later the tip3p water molecules were replaced with the more common SPC/E ones. Starting structure of GpAtm was taken from the Orientations of Proteins in membranes (OPM) database, with the pdb code of 1AFO. At first, two MD simulations were performed on pure and mixed bilayers, each for 10 ns, while Berger forcefield was used for lipid molecules. Later the g_membed program was used to insert the GpAtm into pure and mixed studied bilayers. This computational tool can insert a membrane protein into an equilibrated lipid bilayer, with minimal perturbation. The pure and mixed bilayers

containing GpAtm, were simulated each for 100 ns, while the all-atom gromos53a6 forcefield, including Berger lipid parameters, was used for the simulation of the lipid-peptide system.

In all simulations, Gromacs 4.5.4 package was employed and the temperature was maintained at 323K (above the gel-to-fluid phase transition temperature of the bilayer system), using the Nose-Hoover thermostat. RMSD of DPPC, DMPC, and GpAtm molecules, along with the Box-x length were used to confirm the equilibration of simulated systems. Here we just present some results of the 100 ns simulation of mixed bilayer containing GpAtm, in comparison with the ones from pure DPPC and mixed bilayer systems simulated for 10 ns.

III. Results and Discussion

Fig. 1, represents DOP graphs for pure DPPC bilayer and mixed bilayer with and without GpAtm. After replacing 40% of DPPC molecules, with DMPC, clear decrease in DOP is observed. DMPC molecules have shorter hydrocarbon length in comparison to DPPC ones, resulting in the decrease of order in DPPC molecules alignment. After insertion of GpAtm in the next step, interaction of lipid molecules with protein, gives rise to the more ordered hydrocarbon chains of DPPC molecules, although they are less ordered yet, comparing to the pure DPPC bilayer. Inspection of the RDF graphs for the three mentioned systems, shows the same decrease and increase in the long-range order of DPPC molecules, after adding DMPC molecules and GpAtm, respectively. Fig.2 shows the membrane thickness contour plots for the mixed DPPC-DMPC bilayer including GpAtm, at the first (0ns) and last (100 ns) MD configurations. As the system evolves with time, the bilayer thickness decreases around the box-center, where the GpAtm has been inserted. It is an indication of the negative hydrophobic mismatch (where the hydrophobic length of the peptide is shorter than the hydrophobic thickness of the lipid bilayer). Based on energy requirements, in order to maximize favorable interactions between the hydrophobic length of the integral protein with the hydrophobic bilayer thickness, DPPC molecules shorten their chain lengths, which results in less bilayer thickness.

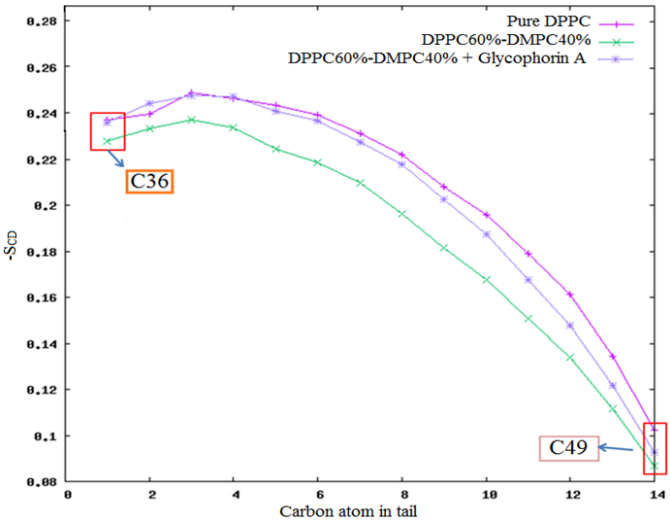

Fig.1 : DOP graphs for three studied systems

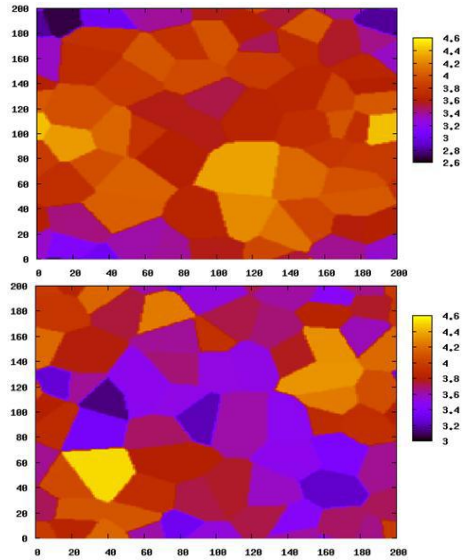

Fig. 2: Bilayer thickness for mixed bilayer system containing GpAtm at first (UP) and last (100ns) MD configurations

References

[1] J. Gumbart, Y. Wang, A. Aksimentiev, E. Tajkhorshid, K. Schulten, "Molecular Dynamics Simulations of Proteins in Lipid Bilayers", *Curr. Opin. Struc. Biol.* vol. 15, pp. 423-431, Aug. 2005.

[2] K. R. MacKenzie, J. H. Prestegard, D. M. Engelman, "A Transmembrane Helix Dimer: Structure and Implications" *Science*, vol, 276, pp. 131-133, Apr. 1997.

[3] M. M. Ghahremanpour, S. S. Arab, S. Biook Aghazadeh, J. zhang, D. van der Spoel, "Membuilder: a web-based graphical interface to build heterogeneously Mixed Membrane Bilayers", *Bioinformatics*, vol. 30, pp. 439-441, 2014.