Study the Interaction of Human Beta Defensin Type 3 with Lipid Membrane

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Introduction

Antimicrobial peptides (AMPs) have been considered to be the key elements in innate immune system of many organisms. In vertebrates, defensins are the most extensively studied AMPs. Based on sequence homology and paring of six cysteine residues, defensins are classified into three categories –α, β, and γ defensins. Only α and β defensins are endogenously found in human. β defensins are found in epithelial cells and Human β-defensins (hBDs) are of particular interest due to their potent antimicrobial activity [1]. Three hBDs (hBD-1, hBD-2, and hBD-3) are being isolated from their natural sources so far but new hBDs are continuously being identified in conjunction with the development of bioinformatics. Compared to hBD-1 and hBD-2, hBD-3 has a charge density of +11, and possesses a broad spectrum of potent bactericidal activities in a salt insensitive manner against both gram positive and negative bacteria, including many drug resistant strains [2]. So, in an era where the microorganisms have been developing resistance against classical antibiotics, understanding the function of AMPs like hBD-3 can be of great interest in medical fields. Its mode of action involves electrostatic interaction with the negatively charged membrane which leads to permeability changes or pore formation in bacteria, fungi membrane and virus envelope [3].
hBD-3 usually functions in a dimer form. In this project, hBD-3 dimer interaction with different lipid membranes are investigated.

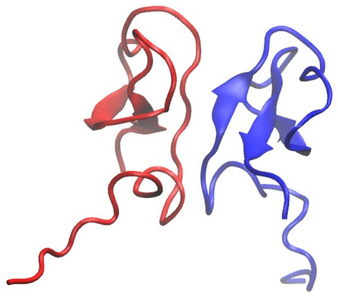

Figure 1. hBD-3 dimer structure with one unit shown in red, the other in blue.

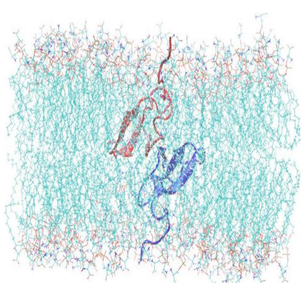

Figure 2. Side view of the hBD-3 dimer in lipid bilayer.

Simulation Details

In this study, the interaction of hBD3 dimer with two negatively charged phospholipids –POPS(1-Palmitoyl-2-oleoyl-sn-glycero-3 –phosphatidylserine) and POPG (1 -Palmitoyl-2-oleoyl-sn-glycero-3-phosphatidylglycerol) are investigated using all-atom molecular dynamic simulation to understand the antimicrobial mechanism of hBD-3. The dimer structure of hBD-3 has been predicted using GRAMM-X protein-protein docking software. Using CHARMM-GUI online software, the all atom molecular dynamic systems were set up by inserting hBD-3 dimer in homogeneous lipid bilayer consisting of 100 lipids in each leaflet of the lipid bilayer. In total, 8 systems were set up. The systems were set up using charmm36 force field at 313.15 K and 1 atm and simulation was performed using NAMD program.

Table1: Overview of Simulation Systems

	Disulfide Bond	Protonation	Type of Lipid	Concentration of NaCl (M)
Charmm-gui1	Yes	No	POPS	0.52
Charmm-gui2	Yes	Yes	POPS	0.52
Charmm-gui3	Yes	No	POPG	0.50
Charmm-gui4	Yes	Yes	POPG	0.50
Charmm-gui5	Yes	No	POPS	1.04
Charmm-gui6	Yes	No	POPG	1.00
Charmm-gui7	No	No	POPS	0.52
Charmm-gui8	No	No	POPG	0.50

Results And Analysis

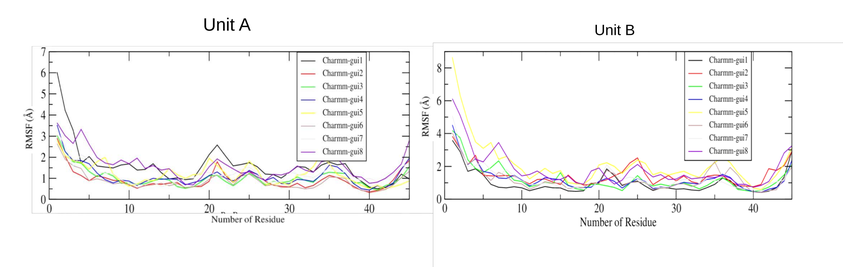

Figure 3. Change in RMSF of two monomers of hBD-3 dimer inside lipid bilayer.

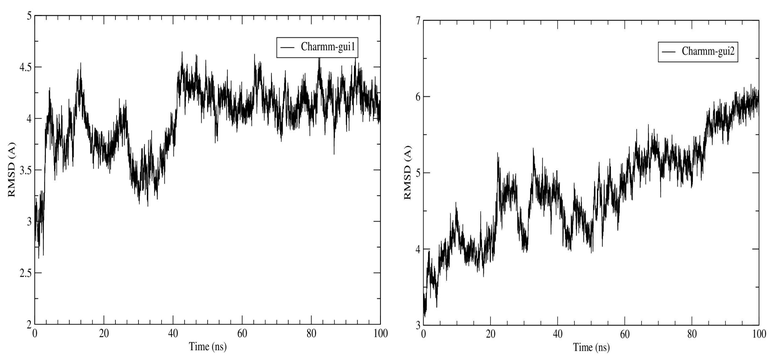

Figure 4. RMSD of hBD-3 dimer during 100 ns simulations.

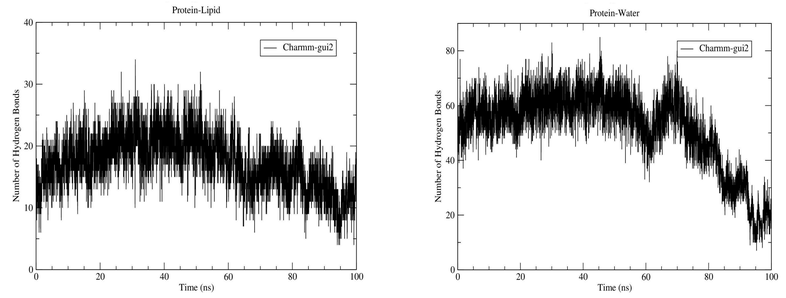

Figure 5. Change in Number of Hydrogen Bonds during the simulation.

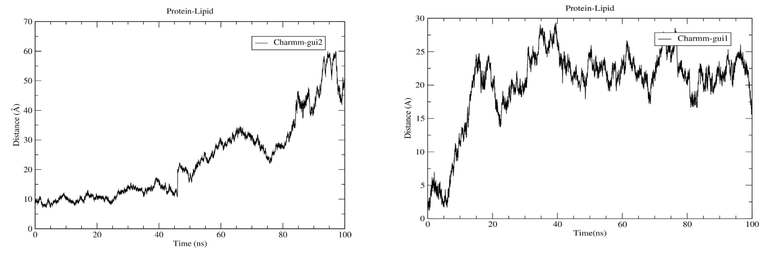

Figure 6. Center of Mass Distance Change during the simulation.

Conclusion

- hBD-3 dimer showed a bit instability in POPS at protonated state other than that it was quite stable in both POPG and POPS in all conditions.
- Protonation and NaCl concentration does not influence notably the structure and dynamics of hBD-3 but hBD-3 structure is found to be more flexible in absence of disulfide bond.

Future Plan

- Will investigate lipid-hBD-3 interaction and PMF(potential Mean Force) during insertion of two hBD-3 dimer using Umbrella-Sampling method.

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References

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