Chapter 11
Summary and Future Prospects

Summary and Future Prospects:

11.1. Overview of results:

The main theme of this thesis is to study the computational approach to examine the alterations in the conformational dynamics of various α-Syn domains and mutants in the membrane-bound state. We have investigated the membrane bound α-Syn in presence of inhibitors and also, the effect of PTM on the protein-membrane interactions.

In the first part of the thesis, we have involved the study of WT α-Syn at atomistic level resolution. The conformational stability of membrane bound α-Syn was studied to understand the α-Syn-membrane interactions that may be useful for developing a new therapeutic approach for treating PD and other neurodegenerative disorders. The amylodogenic NAC region of α-Syn was observed to expose itself from the lipid bilayer surface, which plays a role in the interconversion between the "extended" and "broken-helix" states and consequently leads to the formation of conformational intermediates that are prone to aggregation.

We have characterized the co-solute properties of crowded intracellular environment along with the excluded volume effect to understand the α-Syn dynamics in cell. We have also demonstrated the isolation of the most probable conformer of α-Syn from structural MD analysis based on some critical aspects that emphasizes on its nature of druggabilty as a potential drug target.

In the second part of the thesis, we have demonstrated the key structural features of A30G α-Syn in both membrane-associated and free monomer states using computational approach. Our findings suggested that the structure of A30G α-Syn in solution as a free monomer was noticed to be mostly unfolded, but it did show a preference for helical conformation, which may be important in the aggregation of α-Syn into fibrils.

Then we have compared the conformational dynamics of α-Syn mutants (A30P, A53E, A53T, E46K, G51D and H50Q) and its subsequent aggregation propensity in its membrane bound state. From the MD trajectory analysis, it was evident that membrane bound H50Q α-Syn showed highest flexible region in NAC region that infer diverse effect on aggregation propensity.

Within the third part of the thesis, we have put forward the potential binding position of these two drugs, NPT100-18A and NPT200-11, on α-Syn and the impact of these two drugs on the α-Syn and lipid membrane interactions at an atomistic level. Our findings showed α-Syn in the

presence of peptidomimetic and aminosterol inhibitors (Squalamine and Trodusquemine), adopts well-defined α-helical structures during its interaction with lipid membranes, which inhibits the transition of the α-helical secondary structure into a coiled structure.

We have also studied the conformational characteristics of the truncated CTD α-Syn (1-99 and 1-108) affects its interaction with the membrane and subsequently has an impact on the aggregation. From our findings, the truncated CTD can be suggested to modulate the α-Syn aggregation by interfering with the binding of the α-Syn protein to the membrane and providing support for the pathogenic function of CTD truncation in PD development

Similarly, the MD analysis was performed that gave an insight into the role of phosphorylation at Tyrosine 39 (pY39), Serine 87 (pS87) and Serine129 (pS129). The conformational snapshots of pY39 α-Syn obtained showed a high degree of fluctuations in the N-terminal region that disrupts the helix-2 binding region. In the case of pS87 α-Syn, due to the phosphorylation the C-terminal is flanked out of the membrane that aids in decreased binding affinity towards the membrane bilayer. PTM such as pS129 α-Syn is capable of stabilizing the propensity of the protein to adopt an α-helical rich conformation due to a decrease in the hydrophobicity in binding sites.

11.2. Future Prospects:

Future research in this field will likely focus on improving simulation methodologies, exploring lipid diversity, investigating disease-associated mutations, facilitating drug discovery efforts, and validating computational predictions with experimental data. Integrating MD simulations with experimental techniques, such as nuclear magnetic resonance (NMR) spectroscopy, X-ray crystallography, and single-molecule imaging, can validate computational predictions and provide complementary insights into α-Syn membrane interactions. By comparing simulation results with experimental data, researchers can refine simulation models and improve their accuracy in capturing the complex dynamics of α-Syn membrane interactions. By characterizing the lipid composition of cellular membranes and studying how alterations in lipid metabolism impact α-Syn -membrane interactions, researchers can gain valuable insights into the role of lipids in modulating α-Syn function and aggregation.