AMYLOID β PEPTIDE AND C99 INTERACTION WITH LIPID BILAYER PROVIDES INSIGHT INTO NUCLEATION ON THE MEMBRANE

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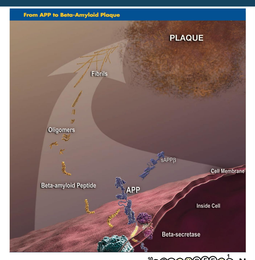
Introduction

Alzheimer's Disease (AD), a neurodegenerative disorder, results in aggregation of Amyloid Beta (Aβ) peptides. These peptides are thought to be the causal factor for neuronal degeneration, but the mechanism of remains unknown. The most significant feature of the endpoint of aggregation is the fibril and plaque formation.

Aβ arises from a specific cut of the Amyloid precursor protein (APP). C99 peptide part of APP is the intermediary that contains Aβ40. What remain unknown are the determinants that dictate which product arises and the method and locations of aggregation that lead to amyloid fiber production.

the interaction of these peptides with cell membranes is probably one of the key pathological events responsible for neuron cell death in AD. However, the molecular mechanism underlying the interaction is not known.

We explore peptide-lipid interactions with C99 and Aβ. We seek to learn how the membrane modulates the conformation and dynamics of Aβ-related peptides leading to beta-sheet formation on the pathway to aggregation and formation of the fibrils.

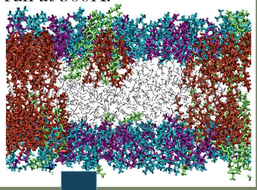


Materials and Method

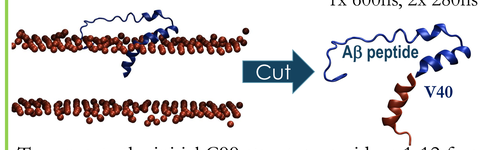
General Parameters

Simulations were run by NAMD 2.12 with TIP3P and CHARMM36m force field. All systems used the HMMM lipid solvent model and were run at 300K.

POPC: 38
Sphingomyelin: 6
Cholesterol: 29
PpE: 27

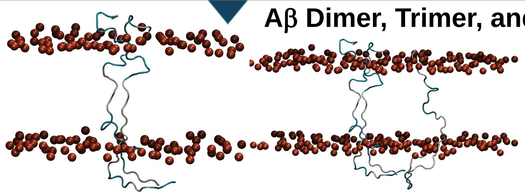


300ns, cut at 220ns — **$C99_{1-55}$** — 3x replicas 1x 600ns, 2x 280ns

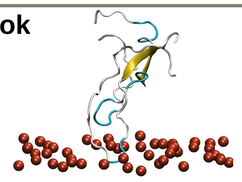


To generate the initial C99 structure, residues 1-12 from an Aβ40 water SDS micelle (Mitayashi 2009) and residues 13-55 from C99 in LMPG micelle (Sanders 2012) were used. Lipid composition is full DMPC. After 220ns, the peptide was cleaved at residue 40 and multiple replicas were run.

Aβ Dimer, Trimer, and Hook



Dimer and trimer structures are different unfolded Aβ(40) structures placed within 5A of each other inside the membrane.

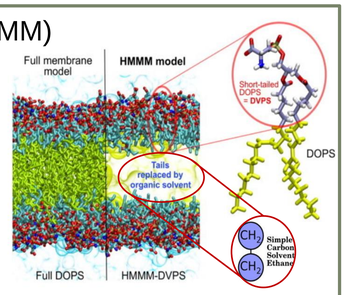


The hook initial structure is a dimer taken from a 5μs simulation of unfolded Aβ40 peptides near a (POPC , POPE, Cholesterol) membrane.

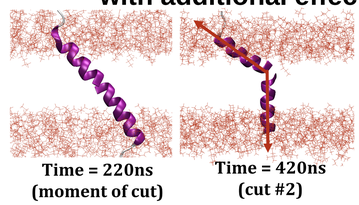
Highly Mobile Membrane-Mimetic (HMMM)

To accelerate the membrane contribution to the conformational changes of Aβ without the loss of atomistic detail, we used the novel high mobility mimetic membrane (HMMM) models.

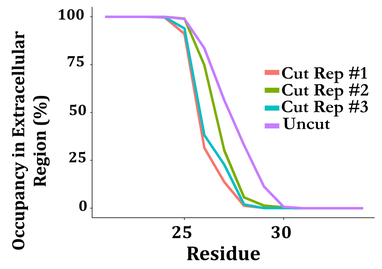
These membrane models increase lateral lipid movement by an order of magnitude while maintaining atomistic detail by modeling the lipid tails nearest the membrane center as a fluid organic solvent while maintaining the atomic description of the lipid head-groups.



The cut results in large-scale conformational shift of the TM-helix with additional effects for the extracellular region

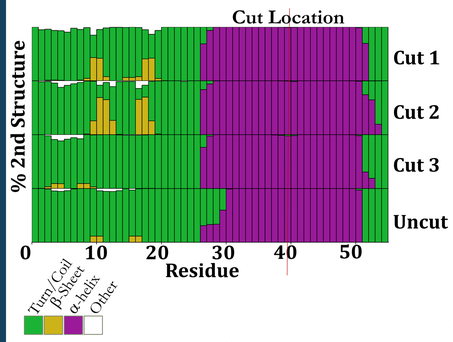


The angle between the parts of the $C99_{1-55}$ transmembrane domain above and below the cut is stable for ~150 ns and then shows greater fluctuations after the cut.

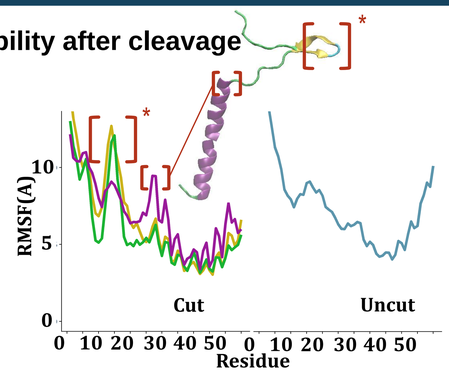


We define occupancy in the extracellular region as the percentage of the time a residue's center of mass is above that of the Phosphorus atoms in lipid headgroups. We find that after cleavage, a few residues (V24, G25, S26, N27, K28, G29) spend less time in the extracellular region. **In other words, Aβ40 interacts more strongly with the headgroups following cleavage than does $C99_{1-55}$.**

$C99_{1-55}$ has more β-content and flexibility after cleavage

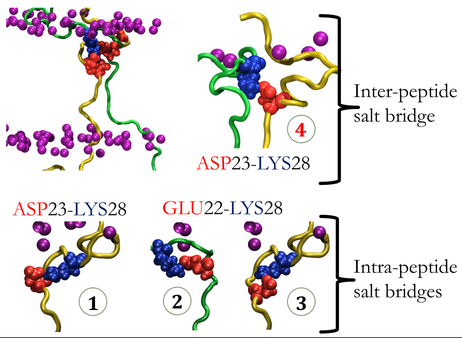


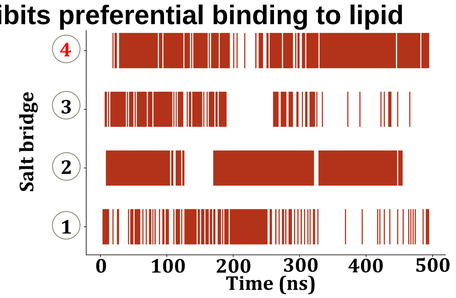
The higher expression of β-content in the extracellular domain of the cut region (Aβ(1-40)) both imply a mechanism initial nucleation.



The root-mean-squared fluctuation (RMSF) of the three cut simulations contain regions of higher variation in structure as compared to the uncut, and these regions occur far away from residue 40, the cut location.

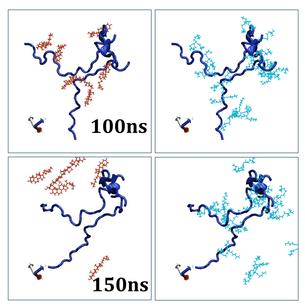
Unstructured Aβ40 dimer in the membrane is stable, with the Asp23-Lys28 salt bridge forming and exhibits preferential binding to lipid





We consider a salt bridge stable if the hydrogen-nitrogen distance is < 3.5A.

We observe that Aβ40 interacts preferentially with POPC and lacks lengthy interactions with Cholesterol. We show lipids within 5A of the protein at 100ns and those same lipids at 150ns. In the image, while all Cholesterol lose strong contact with Aβ, many of the POPC remain in place.



Conclusions and Future Work

Dimer of C55, continue C55 with more replicas, Meredith experimental work, start hook from higher up, different C55 orientation, more membrane composition

The Aβ40 hook forms stronger interaction with the current membrane as compared with a POPC:POPE:CHOL membrane

In contrast to the upright profile of the hook in its original environment, the hook in this simulation creates more contacts with the membrane. As well, while the original conformation yielded a single membrane hook, we observe two with the new membrane composition.

Starting Conformation (after 5μs in POPC:POPE:CHOL)

Conformation after 600ns in
POPC: 38 Sphingomyelin: 6
Cholesterol: 29 PpE: 27