MOLECULAR DYNAMICS SIMULATIONS TO DETERMINE THE NEUROPROTECTIVE MECHANISMS OF CURCUMIN

W. Stone and C. Vander Zanden

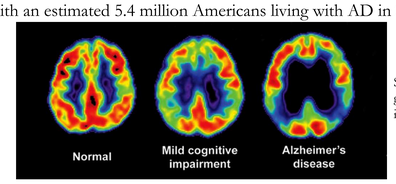
University of Colorado Colorado Springs – Department of Chemistry and Biochemistry

Abstract

Since 1992, Amyloid Beta (Aβ) protein has been investigated as the causative agent in Alzheimer's Disease due to its neurotoxic effects on cell membranes. Curcumin is a polyphenol found in turmeric and has been demonstrated to have neuroprotective effects against Aβ. In order to investigate the chemical mechanisms of this protection, atomistic molecular dynamics (MD) simulations were designed to model Aβ interactions with a model lipid membrane. In a parallel system, curcumin was embedded into the lipid membrane and simulations were performed to determine how the polyphenol alters Aβ interactions with the membrane. Visual inspection of coordinate files and RMSD calculations revealed that the curcumin alters the trajectory of the protein and bilayer thickness calculations showed that the membrane is thicker in the presence of curcumin.

Introduction

- Alzheimer's Disease (AD) is the most common neurodegenerative disease, with an estimated 5.4 million Americans living with AD in 2016[1]

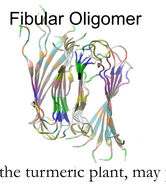


Series of PET scans showing the decline in glucose metabolism associated with cognitive impairment**

- Amyloid Beta (Aβ) is suspected as a causative agent in AD due to its harmful effects on the cell membrane of nerves. Comes in 3 forms:

Monomer

Fibular Oligomer



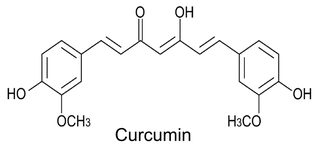
Non-Fibular Oligomer

?

(The structure of the non-fibular oligomer has yet to be fully determined)

- Curcumin, a polyphenol found in the turmeric plant, may protect the membrane from the neurotoxic effects of Aβ[2]

Turmeric

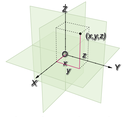


Curcumin

Methodology

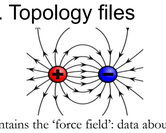
Molecular dynamics were conducted using GROMACS, a software for simulating biologic systems at an atomistic level using equations from classical mechanics. Building a simulation requires several steps:

1. Coordinate Files



contains data about where the atoms are relative to each other*

1. Topology files



contains the 'force field': data about what forces atoms will put on each other*

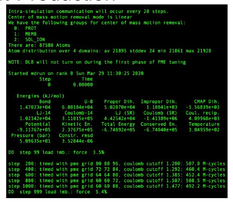
2. Energy Minimization

A short simulation (<100ps) with special parameters to allow molecules in the system to find an energy minimum

3. Equilibration

Another short simulation (<100ps) to allow the temperature and pressure of the system stabilize*

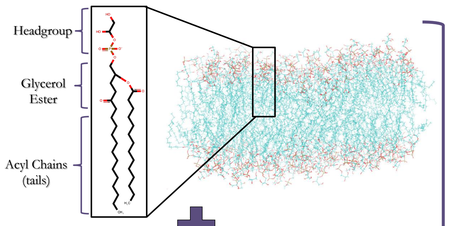
4. Production



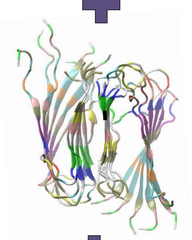
A full length (100ns) simulation using the parameters expected in a real-life system. Simulations were run on Bridges Pittsburgh Supercomputer[3]

Experimental Design

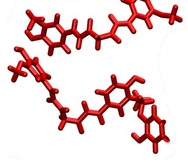
DMPG lipid bilayer:
DMPG is two-tailed phospholipid that makes up part of the cell membrane. In our experiments, the entire membrane is made from DMPG



Aβ:
The structure of proteins is discovered through nuclear magnetic resonance (NMR) spectroscopy. These structures are then stored in the protein data bank (PDB)



Curcumin (CUR):
Structures of small molecules can be found on PubChem. The force field for curcumin can then be generated using CGenFF, an online server maintained by the ParamChem project.



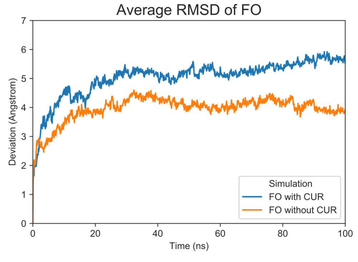
Generated in CHARMM-GUI, a web-based platform for building biomolecular systems. This provides both the coordinates and force fields for these molecules.

4 Different Systems Built:
Each simulation was run in triplicate for 100ns each

- DMPG + Aβ FO + CUR
- DMPG + Aβ FO
- DMPG + CUR
- DMPG

Results: Protein Analysis

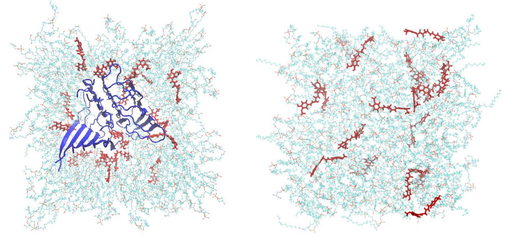
- How a protein interacts with its environment is determined by its shape. In a 100ns simulation, vibrational motion, rotational motion, and changes in the helical folds of the protein can be observed.
- Root Mean Square Deviation (RMSD) is a way to show how the structure of the protein changes during simulation by comparing the location of the coordinates to where they were on the first frame:



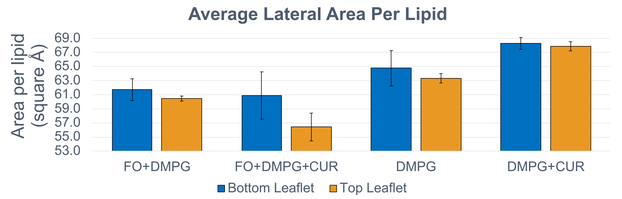
The FO deviates more from its original position when in the presence of curcumin. This could indicate that the curcumin is altering the structure of the protein.

- Using Visual Molecular Dynamics (VMD) software, visual representations of the coordinates can be created:

On the left image, the curcumin can be seen aggregating around the FO. In contrast, the right image shows how the curcumin distributes randomly when the FO is not present

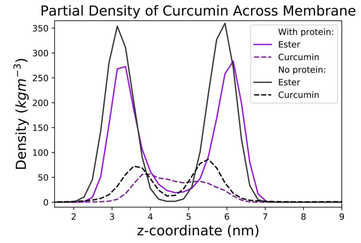


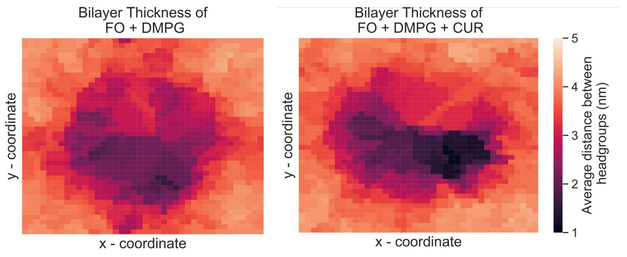
Results: Membrane Analysis



This tells us how much space each lipid has along the x-y plane. The presence of protein 'crowds' the system, reducing the space each lipid headgroup has

The partial density shows how different parts of a system are distributed perpendicular to the membrane. The curcumin tends to aggregate around the ester groups of the lipids when protein is not present, and it aggregates closer to the center of the membrane when the protein is present





The average thickness for the entire membrane is the same for both systems (3.3 nm). However, in the presence of curcumin the protein became more 'compact', as the mode for FO + DMPG + CUR is 28 squares with a thickness of 3.2nm, while the mode of the FO + DMPG system is 28 squares with a thickness of 2.9nm

Conclusion & Future Directions

Preliminary data shows the presence of the curcumin does stabilize the membrane by maintaining membrane thickness around the periphery of the proteins. There is also evidence of curcumin altering the structure of the protein due to the partial density of curcumin and the RMSD of the protein. However, before a conclusion can be drawn, more analysis needs to be done, including:

- Identifying protein-lipid interactions (ID residues + classify as hydrophobic/hydrophilic)
- Identifying protein-polyphenol interactions (ID residues + classify as hydrophobic/hydrophilic)
- Analyze data from Aβ Monomer simulations for comparison

Once these analysis are complete and a conclusion is formed, future simulations can be designed.

Acknowledgements & References

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[2] Tang, M., Taghibiglou, C., & Liu, J. (2017). The Mechanisms of Action of Curcumin in Alzheimer's Disease. Journal of Alzheimer's Disease, 58(4), 1003–1016. https://doi.org/10.3233/JAD-170188

[3] John Towns, Timothy Cockerill, Maytal Dahan, Ian Foster, Kelly Gaither, Andrew Grimshaw, Victor Hazlewood, Scott Lathrop, Dave Lifka, Gregory D. Peterson, Ralph Roskies, J. Ray Scott, Nancy Wilkins-Diehr, "XSEDE: Accelerating Scientific Discovery", Computing in Science & Engineering, vol.16, no. 5, pp. 62-74, Sept.-Oct. 2014, doi:10.1109/MCSE.2014.80

**Photo source: https://www.berkeley.edu/news/media/releases/2009/07/14_alzheimers.shtml)