

## BITS :: Call for Abstracts 2024 - Oral communication

<i>Type</i>	Oral communication
<i>Session</i>	Proteins
<i>Title</i>	Transient interactions between the fuzzy coat and the cross- $\beta$ core of brain-derived A $\beta$ 42 filaments
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### *Motivation*

Alzheimer's disease (AD) leads to cognitive impairment primarily via synaptic loss and is responsible for an estimated two-thirds of dementia cases worldwide. One of its main hallmarks is the presence of extracellular senile plaques formed by the aggregation of the amyloid  $\beta$  (A $\beta$ ) peptide into amyloid fibrils (1), which are filamentous structures characterized by a highly-ordered cross- $\beta$  core. Recently, cryo-electron microscopy (cryo-EM) studies have revealed the structures of A $\beta$ 42 fibrils isolated from brain tissue of 10 Alzheimer's disease patients (2). These studies identify two distinct forms, type I and type II filaments. The structural models derived from both filaments lack the N-terminal residues, which are expected to be disordered and collectively form a fuzzy coat surrounding the cross- $\beta$  core of the filaments. Since the high conformational variability of this region, our understanding of the impact of the fuzzy coat on the cellular interactions of amyloid filaments remains elusive. To address this problem, we exploit the metadynamic electron microscopy metainference (MEMMI) method (3) to obtain the structural ensembles of A $\beta$ 42 in the type I and type II filaments. The analysis of these structural ensembles provides insights into the role of the fuzzy coat in modulating the properties and interactions of amyloid fibrils.

### *Methods*

MEMMI is a Bayesian method that incorporates cryo-EM electron density maps with metadynamic enhanced sampling molecular dynamics. By doing so, it enables a rapid and accurate description of the statistical structural ensemble of the protein that maximally conforms to the experimental data with atomistic resolution, while simultaneously inferring various sources of errors (e.g., data error, forward model error, limited number of replicas able to model the heterogeneity of the system). The initial filament structures are built starting from the PDB databank deposited type-I (PDB: 7q4b) and type-II (PDB: 7q4m) A $\beta$  filament structures, which only contain the cross- $\beta$  core residues. The full-length structure for both the filaments, comprehensive of the N-terminal residues, is modelled using RosettaFold (4). The cryo-EM density maps used as data in MEMMI are EMD-13800 and EMD-13809. For each full-length polymorph structure (type I and type II), the systems are solvated, and the net charge is neutralized by adding the correct number of counter ions. We use the AMBER99SB-ILDN force field and TIP3P water models. Energy minimization, equilibration, and production are carried out following the same protocol and parameters used previously (3), resulting in 8 replicas with an aggregate run time of 1  $\mu$ s, using PLUMED (5) (PLUMED.2.6.0-dev) and GROMACS (6) (GROMACS-2020.5). The cryo-EM restraint is updated every 2 steps, using neighbour lists to compute the overlaps between the model and the experimental voxel data expressed as a data Gaussian Mixture model (GMM). The biasing scheme is PBMetaD (7) with the well-tempered (8) and multiple-walkers protocols (9). As a post-processing step, the initial 25 ns of each replica are excluded as equilibration, followed by the generation of the final structural ensemble by resampling the generated configurations based on the converged unbiasing weights. The conformational ensembles obtained for both the fibril polymorphs are then analysed using Mdtraj (10) and Mdanalysis (11) toolkits.

### *Results*

By employing the MEMMI approach to process cryo-EM electron density maps, we determined the conformational ensembles of the two brain-derived amyloid filaments forms of A $\beta$ 42. The analysis of these ensembles sheds light on the highly dynamic fuzzy coats of these fibrils, illustrating their interactions with the cross- $\beta$  cores of the filaments, thereby revealing their influence on fibril behaviour. Type I filaments exhibit lower solubility compared to type II filaments, with the fuzzy coat contributing to increased solubility in both types. Additionally, the examination of density maps from cryo-EM and MEMMI structural ensembles reveals a slowing down of water and sodium ion diffusion near the filament surface, providing insights into the hydration dynamics of amyloid fibrils. This study demonstrates the effectiveness of the MEMMI approach in analysing cryo-EM maps to characterize the properties of amyloid fibrils.

### *Info*

#### Acknowledgements:

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*Availability* -

**Dissemination Material**

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*Summary*

This research deepens our understanding of AD pathology, highlighting MEMMI's effectiveness in analyzing cryo-EM maps to characterize complex structural properties.

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