Disease (CMTD), the most common disorder of the PNS. Disease mutations induce misassembly of PMP22, resulting both in loss of its function and toxic accumulation of misfolded PMP22 in the cell. Here we present a structural comparison of the wild type and the L16P disease-linked mutant form of human PMP22 to obtain insight into the molecular basis of CMTD. Human PMP22 was expressed in *Escherichia coli*, and purified in the detergent tetradecylphosphocholine. The purified protein provided moderately well dispersed ^{1}H-^{15}N TROSY spectra. NMR resonance assignments for the wild type protein revealed that the 72 observed backbone amide peaks out of 157 expected originate exclusively from the N-terminal STREP tag, transmembrane region 1 (TM1), extracellular loop 1 (ECL1), and the extreme C-terminus. Chemical shift index analysis suggested the residues from TM1 (Met1 to Ile29) form an α-helix, while no secondary structure was predicted for ECL1 (Val30 to Pro58). The L16P mutant was analyzed in a similar manner. A significant finding for the mutant was that the resonances from Ile8 to Val17 located at the middle of TM1 were not observed due to line broadening. Moreover, chemical shift perturbations were observed for residues from Leu18 to Ile24 which are located at the C-terminal end of TM1. These observations suggest that the L16P mutation induced a global conformational change in TM1 that results in its recognition as being folding-defective by components of membrane protein folding quality control system of the endoplasmic reticulum.

Protein Aggregates III

3375-Pos

Possible Pathway between Alpha Helical and Beta Helical Structures of the C-terminal in the Mammalian Prion Protein

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The normal form of the prion protein(PrP^{C}) has mostly alpha-helical (AH) secondary structure in the C-terminal region (residues 166-230), while the infectious form (PrP^{Sc}) has been proposed to have a left-handed beta helical (LHBH) structure(1). The mechanism of conformational change from PrP^{C} to PrP^{Sc} is unknown, but recent electron microscope data(2) and computer modeling(3) of in vitro grown prion fibrils suggest a possible LHBH structure in the C-terminal region. We use high temperature (500K) AMBER molecular dynamics over 10 ns runtimes to study the unfolding transitions commencing from both LHBH and AH C-terminal starting structures. Using stability, contact map, and energetic analyses we find that both structures unfold to very similar AH-like conformations and discuss the potential implications of this result for normal prion cellular function and for prion disease.

References

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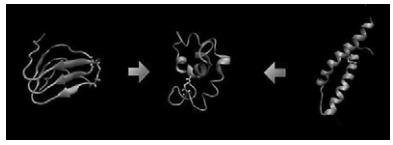
3) Kunes, K. C., et al. (2008) *Prion* **2,** 81-90.

The image below compares computational models of the initial LHBH structure(left), the initial AH structure (right), and the unfolded AH-like structure(middle).



3376-Pos

Molecular Mechanism of Inhibition of Amyloid Formation by Inositol

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Alzheimer's disease (AD) is a severe neurodegenerative disease with no cure. Currently, one method of targeting the underlying disease is to prevent or reverse the amyloid formation of Abeta1-42, a key pathological hallmark of AD. Scyllo-inositol is a promising small-molecule therapeutic that is found to exhibit stereochemistry dependent inhibition of formation of Abeta fibrils in vitro and is currently in phase II of clinical trials. However, the mechanism of action of scyllo-inositol at the molecular level is not known. We perform extensive atomistic molecular dynamics simulations of scyllo-inositol and its inactive stereoisomer, chiro-inositol, to systematically compare and characterize both the binding mode and the effect of inositol on the structure, morphology and aggregation equilibrium of the amyloidogenic fragment of Abeta42, KLVFFAE (Abeta16-22).

3377-Pos

Side Chain Interactions can Impede Amyloid Fibril Growth:Replica Exchange Simulations of Abeta Peptide Mutant

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Aggregation of Aβ peptides is related to the onset of Alzheimer's disease, but the molecular mechanism of the Aβ fibril formation is still poorly understood. Recently, we have studied the thermodynamics and free energy landscape of Aβ fibril growth using a hexamer system of $A\beta_{10\text{-}40}$ peptides by replica exchange molecular dynamics simulations and atomistic implicit solvent model. The system consisted of four peptides forming a fibril fragment and two incoming peptides binding to the fibril edge. We have demonstrated that deposition of the peptides onto the fibril follows the ''dock-lock'' mechanism. In the docking stage, disordered peptides dock to the fibril without their incorporation into the ordered fibril structure. In the locking stage, the incoming peptides form parallel β-sheets with the fibril. In this presentation, we report the effect of D23Y mutation in $A\beta_{10\text{-}40}$ peptides focusing on the changes in the deposition free energy landscape and in the interactions between incoming peptides and the fibril. We found that although D23Y mutation has a weak impact on the docking stage, it induces strong stabilizing effect on the locking stage of fibril growth. We explain these findings by elimination of off-registry side-chain interactions formed by Asp23 in the wild-type Aβ sequence. We conclude that strong off-registry side chain interactions have a capacity to impede fibril growth.

3378-Pos

Elucidating the Association and Dissociation Mechanism of β-Amyloid Protein by Targeted Molecular Dynamics Simulations

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The amyloid-β (Aβ) proteins are responsible for amyloid plaques in Alzheimer's disease and have been the most widely studied subject in the process of fibril growth. Although much progress has been made to elucidate amyloid fibril properties at a molecular level, the full identification and characterization of all the conformational states and oligomeric structures in the aggregation process and all the conformational changes that link between those different states are still needed to be revealed. Here, we present the results of targeted molecular dynamics (TMD) simulations with explicit water to investigate the structural and mechanistic aspects of the association and the dissociation of the Aβ42 dimer. We will discuss the reversibility and the driving forces of the Aβ42 dimerization process with several order parameters along the protein aggregation pathway.

3379-Pos

Charactering Amyloid-Beta Protein Misfolding from Molecular Dynamics Simulation with Explicit Water

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Formation of amyloid-beta (Aß) protein aggregates is the primary cause of amyloid diseases including Alzheimer's disease (AD). Here, we present the state-of-the-art atomic-level characterization of the misfolded state of Aß42 and early misfolding events from helical structure to form aggregation-prone structure in water by using all-atom molecular dynamics (MD) simulations in explicit water environment. Our simulations reveal one of the most important yet unsolved structural mysteries in early misfolding steps that the aggregation-prone structure (APS) of Aß42 is characterized by the non-helical backbone H-bond formation between K16L17 and V39V40I41 associated with the expansion of the hydrophobic exposure. Characterizing the nature of the misfolded state (APS) of Aß42, we provide new insight into the experimentally observed different aggregation propensities of Aß42 compared to those of Aß40. Based on the structural features of APS, we also speculated the hypothetical aggregation mechanism from APS of Aß42 to form fibril accounting three mandatory steps. The structural and mechanistic observations based on these simulations agree with the recent NMR experiments and provide the driving force and structural origin for the Aß42 aggregation process to cause AD.

3380-Pos

Fiber Formation of Silk-Like Proteins

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Beta-sheet forming proteins can fold and assemble into long fibers that play a structural role. While this phenomenon is most famous for its role in amyloidogenic diseases, such fibers also have high potential as biomaterials. In both cases, it is crucial to understand the entire formation process, but the fact that folding and assembly are often intertwined makes this very difficult.
The natural silk fibroin consists of several ''crystalline'' domains with a highly repetitive amino acid sequence, linked through hydrophilic, amorphous spacer sequences. Here, we focus on an artificial silk protein with a (EGAGAGA)x repeat for the crystalline domain (E is glutamate, G is glycine, A is alanine; x denotes the number of repeats) with hydrophilic flanking sequences. Experiments have shown that upon a change in pH, the EGAGAGA repeat will fold and