solutions of mTFP-PNA monomer units resulted in template directed induced assembly. Assembly was confirmed by SDS-PAGE, mass spectrometry, and size exclusion HPLC.
Fluorescence anisotropy was monitored over the course of a mTFP-PNA:DNA titration. Template coding for dimer formation was studied. The anisotropy showed a decreasing trend related to homo-FRET in the assembled forms. A maximal reduction (41%) was observed at a DNA to mTFP-PNA ratio of 1:2. Anisotropy, then increased steadily up to a 1:1 ratio.
This study demonstrates an inducibly assembled homo-FRET system using expressed protein ligation which may be extended to study oligomerization and cluster formation in living systems.

2025-Pos Board B44
Effect of Lipid Bilayers on Prion Peptide Aggregation: Insights from Coarse-Grained Molecular Simulations
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Prion diseases are neurodegenerative diseases associated with a conformational change of the normal cellular form of the prion protein (PrP^{C}) to an abnormal aggregated form (PrP^{Sc}). Recent research suggests that oligomeric rather than plague forms of prion protein (PrP) are the main toxic species, but it is not clear how they lead to disease development. However, the interactions of PrP with membranes have been reported to affect the behavior of PrP, and have been implicated in the toxicity of oligomers. To gain insight into the molecular basis of this effect, we use coarse-grained molecular simulations to study the ability of several amyloidogenic PrP and yeast prion protein fragments to interact with zwitterionic and anionic model membranes. Monomeric and oligomeric forms of PrP are studied in water and in the presence of phosphatidylcholine (POPC) and phosphatidylserine (POPS) bilayers. The conformation of PrP and peptide-lipid interactions are characterized, and the influence of peptide binding to different lipid bilayers on the aggregation process is analyzed.

2026-Pos Board B45
Pores Versus Fibrils: Calcium Ions Regulate Different IAPP-Mediated Membrane Damage Mechanisms
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The disruption of plasma membrane integrity by amyloidogenic proteins is linked to the pathogenesis of a number of common, and frequently deadly, degenerative diseases. using hIAPP (an amyloidogenic peptide associated with beta-cell death in type II diabetes) as an example, we demonstrate that Ca^{2+} ions modulate the membrane interaction of hIAPP, significantly enhancing fiber-dependent membrane disruption while suppressing a pore-like mechanism. QCM, AFM, and NMR results show that Ca^{2+} ions promote a shallow membrane insertion of hIAPP, which leads to both the early accumulation of non-fibrillar oligomers on the membrane surface and later detergent-like removal of lipids from the bilayer triggered by fiber growth. Since both mechanisms are common to amyloid toxicity by most amyloidogenic proteins, it is likely that unregulated Ca^{2+} homeostasis, amyloid aggregation, peptide binding to lipids and membrane leakage are all parts of a self-perpetuating cycle fueling amyloid cytotoxicity.

2027-Pos Board B46
Specific Sequences within Beta-Amyloid Mediate Aggregation Associated with Lipid Membranes
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A hallmark of Alzheimer's disease (AD), a late onset neurodegenerative disease, is the presence of neuritic amyloid plaques deposited within the brain comprised of beta-amyloid (Aß) peptide aggregates. Aß forms a variety of nanoscale, toxic aggregates which have been shown to strongly interact with supported lipid bilayers, which may represent a key step in potential toxic mechanisms. Understanding how specific regions of Aß regulate its aggregation in the absence and presence of surfaces can provide insight into the fundamental interaction of Aß with cellular surfaces. We investigated the interaction of specific fragments of Aß (Aß1-11, Aß1-28, Abeta10-26, Aß12-24, Aß16-22, Aß22-35, and Aß1-40) with lipid membranes. These sequences represent a variety of chemically unique regions along Aß, i.e., the extracellular domain, the central hydrophobic core, and transmembrane domain. We determined how these Aß sequences alter aggregate morphology and induce mechanical changes of lipid bilayers using various scanning probe microscopic techniques, and compared these aggregates with those formed under free solution conditions. In free solution, oligomer and fibrillar aggregate species were formed with varied rate of formation and morphology, i.e. smaller fragments (Aß1-11, Aß12-24, Aß16-22, and Aß22-35) formed smaller oligomers, and shorter, less rigid fibrils. Interaction with model lipid bilayers resulted in distinct aggregates and changes in bilayer stability dependent on the Aß fragment. Aß10-26, Aß16-22, Aß22-35, and Aß1-40 caused disruption of the lipid bilayer structure upon exposure and resulted in a variety of distinct fibrillar aggregates. These interactions were associated with altered mechanical properties of the lipid bilayer. Conversely, Aß1-11, Aß1-28, and Aß12-24 had minimal interaction with a lipid membrane, forming only oligomers. These studies illustrate the potential role of specific amino acid sequences within Aß on aggregation and interactions with lipid membranes.

2028-Pos Board B47
The Interactin of Huntingtin Exon1 with Lipid Bilayers is Regulated by polyQ Length and polyQ Flanking Sequences
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Huntington's Disease (HD) is a neurodegenerative disorder that is defined by the accumulation of nanoscale aggregates comprised of the huntingtin (htt) protein. Aggregation is directly caused by an expanded polyglutamine (polyQ) domain near the N-terminus of htt, leading to a diverse population of aggregate species, including oligomers and fibrils. Furthermore, the length of the polyQ domain is directly related to onset and severity of disease. The first 17 amino acids on the N-terminus (N17) and the polyproline (polyP) domain on the C-terminal side of the polyQ domain have been shown to further modulate the aggregation process. Additionally, N17 appears to have lipid binding properties as htt interacts with a variety of membrane-containing structures present in cells, such as organelles, and interactions with these membrane surfaces may further modulate htt aggregation. To investigate the interaction between htt exon1 and lipid bilayers, in situ atomic force microscopy (AFM) was used to directly monitor the aggregation of htt exon1 constructs with varying polyQ-length or synthetic peptides with different combinations of polyQ domain flanking sequences associated with htt exon1 on supported lipid membranes comprised of total brain lipid extract. The exon1 fragments accumulated on the lipid membranes, causing disruption of the membrane, in a polyQ-length dependent manner. By adding N-terminal tags to the htt exon1 fragments, the interaction with the lipid bilayer was impeded. Synthetic peptides lacking the N17 flanking sequence had no appreciable interaction with lipid bilayers. Interestingly, polyQ peptides with the N17 flanking sequence interacted with the bilayer. This interaction was further modulated by the addition of the polyP domain.

2029-Pos Board B48
The Molecular Assembly of the Aerolysin Pore Reveals a Unique Swirling Membrane-Insertion Mechanism
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Aerolysin is the founding member of a super-family of ß-pore forming toxins for which the pore structure is unknown. We have combined X-ray crystallography, cryo-electron microscopy (EM), molecular dynamics and computational modeling to determine the structures of aerolysin mutants in their monomeric and heptameric forms, trapped at various stages of the pore formation process. A dynamic docking approach based on swarm intelligence was applied whereby the intrinsic flexibility of aerolysin extracted from new X-ray structures was utilized to fully exploit the cryo-EM spatial restraints. using this integrated strategy, we obtained a radically new arrangement of the prepore conformation and a near-atomistic structure of the aerolysin pore, which is fully consistent with all biochemical data available so far. Upon transition from the prepore to pore, the aerolysin heptamer shows a unique concerted swirling movement, accompanied by a vertical collapse of the complex, ultimately leading to the insertion of a transmembrane ß-barrel.

2030-Pos Board B49
Concentration Dependent Transition of Membrane-Bound Beta-Stranded KIGAKI Peptides from Unstructured Monomers into Immobilized Amyloid Fibrils Observed by Solid-State ^{19}F-NMR
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The structure, membrane alignment, flexibility, and aggregation behavior of the β-stranded antimicrobial peptide KIGAKI [with sequence $(KIGAKI)_3$-NH_2] has been determined in oriented lipid bilayers using circular dichroism (CD), oriented CD (OCD), and solid state NMR spectroscopy. Several IIe or Ala residues were replaced one at a time with CF_3-L/D-Bpg or Ala-d_3. At high