309-Pos Board B64
Assessing Polyglutamine Conformation and Aggregation with Molecular Dynamics Techniques
Riley J. Workman, Jeffry D. Madura.
Duquesne University, Pittsburgh, PA, USA.
Huntington's disease is one of nine neurodegenerative diseases characterized by gene mutations causing polyglutamine (polyQ) repeats in various proteins. Mutated proteins misfold, aggregate, and form amyloid-like fibrils in the neuron. As of now, the aggregation mechanism of these polyglutamine proteins is not well understood. Experimental techniques such as resonance Raman, circular dichroism, and ssNMR are used to analyze properties of polyglutamine solutions. Computational analysis is used in concert with experiment to allow investigation on the molecular level. In this work, short polyQ peptides are studied using molecular dynamics (MD) methods in order to better understand the mechanics of their aggregation. In prior work, we characterized the monomeric conformational ensemble of $D_2Q_{10}K_2$ peptides. Our next step is to investigate dimerization properties of these peptides. Adaptive biasing force paired with MD is used to evaluate the dimerization free energies and conformations of $D_2Q_{10}K_2$ peptides. On a larger scale, classical MD is used to evaluate the properties of multiple aggregate conformations, including facially stacked β-sheet and β-hairpin sheet systems. Ψ angle probability distributions will be generated from the resulting aggregate trajectories for comparison with experimental distributions. Results from both projects will be presented.

310-Pos Board B65
Role of Hydrophobicity in the Aggregation of Intrinsically Disordered Peptides
Agusti Emperador.
Institute for Research in Biomedicine, Barcelona, Spain.
We have studied the oligomerization of intrinsically disordered peptides using a coarse-grained model with implicit solvent. We have focused our study on the influence of hydrophobicity on the aggregation dynamics of the system, constituted by a high number of peptides in a box with periodic boundary conditions. We use an intermediate resolution coarse-grained model, with a fairly accurate representation of sidechain size, geometry and hydrophobicity.
Changing the strength of the hydrophobic term compared to the hydrogen bonding term, we have found that in the limit of high hydrophobicity the formation of high order oligomers is very slow. This is due to the folding of each peptide in a molten globule state with a strongly hydrophobic cluster and a hydrophilic surface that minimizes the free energy when completely solvated, therefore elusive to aggregation.

311-Pos Board B66
PH Reversible Conjugates of Graphene Oxide with Peptides and Proteins
Avanish S. Parmar[1], Douglas Pike[1], Patrick Nosker[1], Daniel Grisham[1], Nida F. Hasan[1], David Yin[1], Yuan Chen[2], Faith Njoku[2], Jenny Lockard[2], Vikas Nanda[1].
[1]Center for Advanced Biotechnology and Medicine, Department of Biochemistry and Molecular Biology Robert Wood Johnson Medical School, Rutgers University, Piscataway, NJ, USA, [2]Department of Chemistry, Rutgers University-Newark, Newark, NJ, USA.
Graphene Oxide (GO) is an attractive material for biomedical applications due to its unique properties, such as the abundance of surface polar groups, amphiphilicity, and biocompatibility. Much focus has been given to elastin-like polypeptides because of their stimulus responsive behavior, displaying lower critical solution temperature phase behavior, making them attractive for biomedical and biotechnological applications. Our aim is to engineer similar behavior in GO, combining it with engineered biomolecules such as peptides and proteins for pH, temperature and structural stimulus responses. Understanding the interaction between the GO and proteins at the molecular level is critical. However, this is a formidable challenge due to the complexity of proteins and the heterogeneous nature of the GO surface. In this work we elucidate the interaction between GO and biomolecules ranging from free amino acids to peptides and proteins. We used various biophysical methods like UV-Vis, Dynamic Light Scattering, Zeta Potential, Raman spectroscopy, optical imaging, etc. to characterize and understand the GO and biomolecule interactions in detail. Understanding the molecular interaction helps us to successfully form the pH reversible conjugates of GO in the presence of peptides and proteins. Our results suggest that electrostatic interactions are the driving force for the formation of amino acid and peptide-GO conjugates, but in the case of proteins, electrostatic interactions alone are insufficient; exposed hydrophobic groups or reduction in electrostatic repulsion between the proteins during denaturation in conjunction with electrostatics drive the formation of GO-protein conjugates. This property of GO may be utilized for many biomedical and nanotechnology applications such as drug delivery, biosensor applications and hybrid nano-biomaterials.

312-Pos Board B67
Single-Molecule AFM Force Spectroscopy Reveals the Difference in the Folding Patterns between Amyloid β 40 and 42 Within Dimers
Zhengjian Lv[1], Robin Roychaudhuri[2], Margaret M. Condron[2], David B. Teplow[3], Yuri L. Lyubchenko[1].
[1]University of Nebraska Medical Center, Omaha, NE, USA, [2]Department of Neurology, David Geffen School of Medicine at UCLA, Los Angeles, CA, USA, [3]Department of Neurology; and Brain Research Institute and Molecular Biology Institute, David Geffen School of Medicine at UCLA, Los Angeles, CA, USA.
Aβ42 and Aβ40 are the two primary alloforms of the amyloid β-protein (Aβ) found in humans. The two additional residues at the C-terminus of Aβ42 result in elevated neurotoxicity compared with Aβ40, but the molecular mechanism underlying this effect remains unclear. Here, we used single-molecule force microscopy (SMFS) to probe Aβ42 and Aβ40, and corresponding C-termini mutants to characterize their interpeptide interactions within dimers. The dynamic force spectroscopy (DFS) analysis revealed an elevated stability of Aβ42 dimers (0.18 s) compared with Aβ40 dimers (0.11 s). Probing the transient structures of both dimers demonstrated a dramatic difference in the folding patterns of monomers in dimers, with the primary contributions arising from the C-terminal interactions for Aβ42 and the N-terminal interactions for Aβ40. Mutations in the C-termini change the contributing interactions for both peptides. Gly33Val-Val36Pro-Gly38Val (VPV) substitutions increase the stability of transient dimers more than three-fold for both Aβ42 and Aβ40. This stabilization is explained by changes in the peptide folding patterns, observed by a decrease in the N-terminal interactions for Aβ40 and an increase in the central and C-terminal interactions for [VPV] Aβ42. The double substitution pP, Val36 to D-enantiomer Pro and Gly37 to L-Pro, decreases the stability of dimers, as evidenced by similar lifetimes of [pP]Aβ42 (0.11 s) and Aβ40 (0.11 s). Probing the dimer's structures revealed a surprising similarity of the interpeptide interaction patterns for [pP]Aβ42 and Aβ40, which was predominately through the N-termini. Overall, our findings demonstrate directly the effect of local sequence on the global structural reorganization of Aβ monomers within dimers and on their aggregation propensity.

313-Pos Board B68
Structural Studies of Septin Protein Assemblies by Direct Stochastic Optical Reconstruction Microscopy
Adriano Vissa, Theodore Pham, William S. Trimble, Peter K. Kim, Christopher M. Yip.
Biochemistry, University of Toronto, Toronto, ON, Canada.
Septins belong to a family of GTP-binding proteins that are thought to have various functions in mammalian cells including acting as scaffolds for protein recruitment, forming diffusion barriers in primary cilia, and participating in cytokinesis. These proteins, sometimes referred to as the fourth cytoskeletal component in part due to their colocalization with F-actin, assemble into homo- and hetero-oligomers such as the SEPT7-SEPT6-SEPT2 complex. This complex has been shown to form the symmetrically arranged hexamer SEPT7-SEPT6-SEPT2-SEPT2-SEPT6-SEPT7, with recent evidence pointing to the association of SEPT 9 at either end of the hexamer. These filamentous building blocks are thought to form higher-order assemblies such as bundles and upon actin depolymerisation, dissociate from actin to form characteristic ring structures. The organizational motif by which this assembly takes place is not currently well understood. To address this question, we use dSTORM (direct Stochastic Optical Reconstruction Microscopy), a single molecule localization-based approach to resolve diffraction-limited fluorescent signals in both space and time, with a theoretical lateral resolution of 10-20 nm and axial resolution of 50-70 nm. We report here the development and implementation of the dSTORM imaging technique on a combinatorial TIRF/confocal/AFM platform. Two-colour dSTORM imaging was performed by labelling F-actin/SEPT2 and SEPT2/SETP9 in human fibroblast cells using AlexaFluor 488 and AlexaFluor 647, with and without the presence of an actin depolymerising drug. The combinatorial diversity and higher-order structural assembly of septins are likely directly related to their specialized cytoskeletal functions and versatility. Therefore, a molecular scale investigation of the structural complexities of these proteins could lead to a better understanding of their cellular roles.