1242-Pos
Interface Dynamics In Hub Proteins
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Dynamical properties of proteins may have a significant role in regulating protein-protein interactions. In particular, intrinsic disorder and disorder-order transitions have been claimed to be especially involved in the binding of proteins known as ''hubs'' [1], which are characterized by a high level of connectivity in protein-protein interaction (PPI) networks. We therefore started to investigate the dynamical properties of hubs through molecular simulations, to assess the role of conformational flexibility in promoting ''promiscuity'' of interactions.
For this study, a dataset of proteins with known structure and interaction partners was first prepared. To cope with the incompleteness of the interaction data contained in the Protein Data Bank (PDB), we mapped the PPI database IntAct [2], which collects interactions from a wide range of experimental techniques, onto the structure-based database PiSite [3]. The proteins were then partitioned into 'classes' with increasing number of interactions.
A preliminary survey of the dynamical properties of each class was done using two independent approaches, namely tCONCOORD [4] and the Gaussian Network Model [5]. For each complex, the interfaces were extracted using the POPSComp method [6,7]. The availability of information on both the dynamics and the interaction properties will allow to determine possible correlations between flexibility and binding diversity.
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Protein Folding & Stability: Interactions with Membranes & Lipids

1243-Pos
Resolving the Native Structure of *Escherichia Coli* OmpA
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Modification of the sorting signal (residues 163-169) of outer membrane protein A (OmpA) of *Escherichia coli* by amphiphilic, water-insoluble oligo-(R)-3-hydroxybutyrates (cOHB) enables it to form narrow pores of ~ 80 pS in planar lipid bilayers at room temperature. Here we show that additional modifications of the C-terminal domain of OmpA in the periplasm enable it to refold into large pores in the outer membranes. Both narrow and large pore conformers migrate as 30 kDa proteins on SDS-PAGE gels. OmpA isolated from outer membranes (M-OmpA) refolds into large pores of ~ 450 pS after incubation in micelles or planar bilayers at elevated temperatures (E_a 33.2 kcal/mol), whereas OmpA isolated from cytoplasmic inclusion bodies (I-OmpA) treated in the same manner continues to form only narrow pores. Western blot immunoassay using anti-OHB IgG and ^{1}H-NMR indicate that chymotrypsin-generated C-terminal segment 264-325 of M-OmpA contains cOHB, whereas the same segment of I-OmpA does not. Importantly, the narrow to large pore transition also fails to occur when M-OmpA is exposed to disulfide bond reducing agents. The results indicate that cOHB-modification of the sorting signal in the cytoplasm and of the C-terminal segment 264-325 in the periplasm as well as C_{290} -C_{302} disulfide bond formation in the periplasm are all necessary steps in folding OmpA to its native large pore structure. They further suggest that cOHB modification may be an important factor in protein targeting and protein folding.

1244-Pos
Spectroscopic Study of Anchoring Aromatic Residues in Membrane Proteins and Peptides: Applications to Protein Folding and Vesicle Disruption
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Aromatic amino acids play critical roles in the stability and function of membrane-associated proteins and peptides. Here, we apply the site-selective vibrational tool, UV resonance Raman spectroscopy, to probe changes in the structure and microenvironment of tryptophan residues in integral membrane proteins and membrane-associated antimicrobial peptides. Alterations in molecular interactions, such as hydrogen-bonding states, cation-pi interactions, and local polarity, of tryptophan residues accompany the association and folding of these membrane-bound proteins to synthetic lipid bilayers. These results reveal the diversity of molecular interactions that help guide the in vitro assembly of membrane proteins and peptides in vesicles, and provide molecular clues to the mechanisms of membrane protein folding and vesicle disruption.

1245-Pos
Combining Genomic Information with Molecular Dynamics Simulation to Model Two-Component Signal-Transduction Systems
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Bacteria, archaea and some Eukaryotes employ so-called two-component signal transduction systems (TCS) as a means of adaptation to cellular and environmental stimuli. Typically, these systems feature a membrane bound sensor histidine kinase (SK), which modulates its autokinase activity in response to the stimulus and a transcription factor/response regulator (RR), which accepts a phosphoryl group from the SK and in turn mediates a cellular response. The phosphoryl-transfer requires the formation of a SK/RR complex ruled by transient interaction. This and other transient protein complexes involved in signal transduction are difficult to resolve by experimental means, such as X-ray crystallography or NMR, as evidenced by a lack of structural representatives for many such systems in the protein database PDB. The presented work demonstrates how theoretical methods can close this gap. A genomic direct coupling analysis extracts protein-protein interaction contacts from protein sequence databases. This information is integrated with experimentally determined structures of the unbound proteins in molecular dynamical simulation to understand protein docking and predict the structure of the protein complex. The reliability of this approach is demonstrated by achieving crystal resolution accuracy when reconstituting the known sporulation phosphotransferase complex between Spo0B and Spo0F, which is related to the TCS phsphotransfer complex. We introduce a structural model for the complex of TCS proteins TM0853 with TM0468, consistent with all available experimental data.

1246-Pos
Computer Simulations of Alzheimer's Beta Amyloid Interactions with Multicomponent Lipid Bilayers
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Amyloidogenic protein unfolding and subsequent aggregation on cell surfaces are linked to many protein misfolding diseases, e.g., Alzheimer's (AD) and Parkinson's. Beta-amyloid (betaA), a 39 to 43 residue peptide, is released from neuronal membranes upon sequential proteolytic cleavages of a large transmembrane amyloid precursor protein by two secretases. An understanding of the conformational transitions and stability mediated by the lipid surface interactions is important for developing new strategies for the prevention and treatment of protein misfolding diseases. Using all-atom MD simulation techniques, we explore the initial folding and lipid insertion kinetics of betaA of both 40 and 42 residue long on the surfaces of well-defined lipid nanodomains with different cholesterol contents that mimic the neuronal lipid membranes. Several molecular clusters consisting of different initial conformations, alpha-barrel, beta-sheet and globular-coil, of betaA and stable mixed lipid bilayer in explicit solvent have successfully been constructed. The conformational transitions and stability of betaA on the lipid surface and in a partially inserted state on lipid nanodomains of different lipid compositions were systematically studied. The protein-induced membrane disruptions were examined by calculating the lipid order parameter, water permeability and bilayer thickness profiles of the lipid bilayers. The time-dependent secondary structure of betaA was used to gauge the unfolding events of the protein and its dependence on the lipid composition of the interacting bilayers. Our results revealed that the cholesterol content in the lipid bilayer strongly affects the initial lipid surface-induced unfolding and bilayer-insertion and stability behavior of betaA in our model bilayer systems. Our computer simulation data may provide useful computational insights on the controversial sensitization and protective roles of lipid membranes on the protein aggregation events in AD.

1247-Pos
Lipid-Membrane Mediated Tau Misfolding and Aggregation
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Neurofibrillary tangles comprise of aggregated tau protein are a pathological hallmark of Alzheimer's disease (AD). However, the molecular basis of the early tau aggregation events, such as the nature of the structural fluctuations that trigger the cascade of misfolding and aggregation events, are unknown.