Gallic acid (GA) has been employed to explore the mechanism of inhibition of protein fibrillation. Various spectroscopic techniques such as UV-vis, fluorescence, circular dichroism and dynamic light scattering along with microscopic studies have been performed to investigate the anti-amyloidogenic property of GA on hen egg white lysozyme (HEWL). Results indicate a dose dependent inhibition of HEWL fibrillation by GA. Gel electrophoresis studies suggested that the ability of *o*-dihydroxy moiety present in the chemical structure of GA to be oxidized into the quinone moiety and H_2O_2 in the system plays a pivotal role in the inhibition. Covalent binding of quinones to the hydrophobic Trp residues of HEWL is the reason for which the hydrophobic regions of HEWL are not affected upon denaturing conditions. A new insight from this study using cyclic voltammetry revealed that GA imparts protection to the partially unfolded proteins by oxidizing the Met residues into highly polar sulfoxide-modified side chains through the *in situ* generation of H_2O_2. This helps to prevent self- association by stabilizing the partially unfolded proteins by the solvent molecules.

1159-Pos Board B68

Investigating Curli and Cellulose Interactions in the Spatial Context of Bacterial Biofilms

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Uropathogenic *E. coli* (UPEC) are responsible for the majority of urinary tract infections (UTIs), one of the most common bacterial infections in humans. Half of all women will contract at least one UTI in their lifetime, and the infection frequently recurs after treatment. UPEC enter the urinary tract then spread to the bladder as they colonize into bacterial biofilms encapsulated by the protective structure of an extracellular matrix (ECM). Biofilm formation provides a bacterial community with more resistance to environmental stresses as well as antibiotics. The main ECM constituents of UPEC are the functional amyloid protein curli, and bacterial cellulose, a β-1,4 linked glucose polymer, as well as a modified form of cellulose. Curli fibers mediate interaction, attachment, and colonization on the host while cellulose seems to provide cohesion and highly ordered assemblies within the biofilm architecture.

We set out to distinguish between curli and cellulose in the complex three-dimensional spatial context of bacterial biofilm. Utilizing microscopy, biochemistry, and solid-state NMR spectroscopy we probe the structural contacts between curli and cellulose and investigate the machinery responsible for production of these ECM components at the molecular level. Obtaining a higher resolution view of the order and interactions within the matrix encapsulating these persistent bacteria will allow for new strategies to disrupt biofilm formation.

Posters: Protein Dynamics and Allostery II

1160-Pos Board B69

Antigen Processing at the Atomic Level: MD Simulations of MHCI and its Peptide-Loading Complex

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Antigens exposed at the cell surface by major histocompatibility complex class I (MHCI) proteins enable self/non-self recognition by cytotoxic T cells, protecting the organism against viral infections and cancer-causing mutations. To perform their role, MHCI must first be loaded with an antigenic peptide inside the endoplasmic reticulum (ER), a process controlled by a multi-protein assembly called the peptide-loading complex (PLC). In the absence of any experimental structure of the PLC, we used molecular dynamics (MD) simulations to study its individual components, their assembly and their function. We have predicted the structure of the tapasin • MHCI interface, and shown how tapasin both protects empty MHCI from unfolding and catalyses the selection of high-affinity peptides through a molecular tug-of-war mechanism: tapasin pulls on a region of the MHCI peptide binding groove to open it, while the peptide simultaneously tries to close the groove. Low-affinity contenders ''lose'' this challenge and are exchanged until a high-affinity one binds to and closes the groove, thereby initiating complex break-down. We have also shown how tapasin recruits the transporter associated with antigen processing (TAP) into the PLC via transmembrane interactions. In addition, by truncating antigens or removing them from the MHCI binding groove, we made a spatially resolved map of MHCI plasticity, revealing how peptide loading status affects key structural regions contacting tapasin. Finally, we have integrated the previous elements to build a computational model of the full PLC. Our MD simulations explain experimental kinetics and mutagenesis data, and represent the first in-depth, atomic-level study of the mechanism underlying the PLC, an important step towards a better understanding of adaptive immunity.

1161-Pos Board B70

Protein Local Conformations at the Light of a Structural Alphabet

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Protein structures are classically described in terms of secondary structures. Even if the regular secondary structures have relevant physical meaning, their recognition from atomic coordinates has some important limitations such as uncertainties in the assignment of boundaries of helical and β-strand regions. Further, on an average about 50% of all residues are assigned to an irregular state, i.e., the coil. Thus different research teams have focused on abstracting conformation of protein backbone in the localized short stretches. Using different geometric measures, local stretches in protein structures are clustered in a chosen number of states. A prototype representative of the local structures in each cluster is generally defined. These libraries of local structures prototypes are named as ''structural alphabets''. We have developed a structural alphabet, named Protein Blocks, not only to approximate the protein structure, but also to predict them from sequence. Since its development, we and other teams have explored numerous new research fields using this structural alphabet. I will review here some of the most interesting applications: (i) the most efficient protein superimposition methods, (ii) new ways to analyse protein structures, and (iii) new tool for analysis of protein dynamics and allostery.

1162-Pos Board B71

Ancient Thioredoxins Evolved to Modern Day Stability-Function Requirement by Altering Native State Ensemble

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Thioredoxins - small globular proteins that reduce other proteins - are ubiquitous in all forms of life, from archaea to mammals. Ancestral Thioredoxins (Thrxs), which date back to almost 4 billion years, share high sequential and structural similarity with the modern day (extant) Thrxs, and yet they exhibit significantly different functional activity and stability. We investigate this by comparative studies of their (ancient and modern day Thrxs) folded state ensemble quantified by the Dynamic Flexibility Index (DFI). DFI - computed from well-converged (micro-seconds long) all-atom simulations of the native state - measures the relative resilience of an amino acid to perturbations in the rest of the chain when displaced out of equilibrium. Clustering proteins based on DFI profiles leads to groups that strongly resemble alternate classification scheme based on their activities and phylogeny. Despite the high structural similarity between the extant and its ancestor on each branch, the DFI profiles of the extant proteins are substantially different around alpha3, alpha 4 and catalytic regions. Moreover, the change in flexibility alters the allosteric coupling of the active site with rest of the protein in modern enzymes that may be responsible for their decreased catalytic activity. At a global level, we observed changes in DFI distribution profiles of Thrxs as they evolve. In particular, we note the population of low flexible (called hinges) and high flexible sites increases by minimal but subtle manipulations in the flexibility profiles without changing the actual hinges. Broadly speaking, the heterogeneity (quantified by the variance) in the DFI distribution increases with the decrease in the melting temperature typically associated with evolution of ancient proteins to their modern-day counterparts.

1163-Pos Board B72

Mechanisms of Protein-Protein Sliding: Coiled Coils as a Tool Model

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The *alpha*-helical coiled coil domain is a ubiquitous protein motif found in approximately 5% of all translated protein sequences. Due to the relative simplicity of the sequence/structure relationship, coiled coils have been extensively studied, leading the emergence of novel protein design and engineering techniques. Nevertheless, a complete understanding of coiled coils structure and their dynamics is still far from being achieved. For instance, it has been experimentally observed that coiled coils exhibit staggered conformations that are generated after sliding events between two helixes. Using structure-based modeling techniques as well as coiled coil design tools, we study the mechanisms of sliding for parallel and antiparallel homodimer coiled coil conformations. We are motivated at characterizing the molecular determinants that govern the kinetics and dynamics of sliding in coiled coils and to predict its prevalence in natural coiled-coil proteins.