Protein folds have been invented and adopted under various evolutionary constraints. The question arises how general mechanical attributes that control protein function have been sampled by the evolving protein domain repertoire in the varying biological environment.
We assessed three measures, namely flexibility, foldability, and mechanical strength, for ~100,000 protein domains, using geometrical sampling (Concoord), contact order, and coarse-grained protein unfolding, respectively. In general, we find high mechanical stability to involve low protein flexibility and low foldability.
In order to reconstruct protein architecture evolution, we used infor-mation embedded in a structural genomic census of fold architectures defined by a phylogenomic analysis of 185 completely sequenced genomes using advanced hidden Markov models. Interestingly, flexibility is found to increase in evolution, hinting towards a fine-tuning of protein function during evolution towards dynamic regulation.
Finally, our work can explain how proteins as an intricate network of inter-molecular bonds were designed by evolution as a response to mechanical stress.

1274-Pos Board B44
Ligand Recognition of Lipocalin-Type Prostaglandin D Synthase
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Lipocalin-type prostaglandin (PG) D synthase (L-PGDS) is a multi functional protein that functions as a PGD_2 synthesizing enzyme, a scavenger of various lipophilic ligands, and an amyloid β chaperone in the brain. L-PGDS is a member of the lipocalin superfamily and has the ability to bind various lipophilic molecules, such as prostanoid, retinoid, bile pigment, and amyloid β peptide. However, the molecular mechanism for this wide variety of ligand binding is not fully understood. In this study, we determined the NMR structures of recombinant mouse L-PGDS and a L-PGDS/PGH_2 analog. L-PGDS has a typical lipocalin fold, consisting of an eight-stranded β-barrel and a single α-helix. The interior of the barrel forms a hydrophobic cavity and the upper end of the barrel is open. The size of the barrel was found to be larger than those of other lipocalins and the cavity contained two pockets. The results of NMR titration, kinetic and molecular docking experiments revealed that PGH_2 and retinoic acid occupied the hydrophilic pockets 1 and 2, respectively. A structural comparison of the L-PGDS/PGH_2 analog complex with apo-L-PGDS showed that the H2-helix, CD-loop, and EF-loop located at the upper end of the β-barrel undergo a conformational change and cover the entrance of the cavity upon U-46619 binding. These results indicate that the two binding sites in the large cavity are responsible for the broad ligand specificity of L-PGDS in its induced-fit mechanism.

1275-Pos Board B45
Studying the Structure and Function of the Cold Shock Related Protein CspE, in Escherichia Coli through Molecular Modeling
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In the bacterium *Escherichia coli,* a single 4,639 base pair circular chromosome is compacted into about 100 independently supercoiled domains that will form a rosette structure, referred to as the nucleoid. One of the proteins shown to be involved in chromosome condensation is a 7-kDa protein called CspE. *Escherichia coli* K12, expresses nine Csp (Cold shock-related protein) gene products: (CspA-I) and they share between 29 and 83 percent identity in their amino acid sequences. The bacterium *Bacillus subtilis* has three Csp homologues with greater than 70% identity to CspE and studies have shown that Csp proteins bind to single stranded poly d(T) with a minimum of 6 thymines in an 8 base sequence. CspB in *Bacillus subtilis* has solved crystal structures both with and without it's DNA ligand. While the crystal structure of CspA from *Escherichia coli* has been elucidated, the remaining eight Csps have not. This work describes the use of structural information from the homologue CspB in *B. subtilis* to model the structure of the remaining eight unsolved structures of the nine Csps in *E. coli*. A single point mutation in the computationally predicted binding site of CspE results in the loss of its DNA binding function while a single point mutation outside of the predicted binding site shows no significant difference in DNA binding compared to wildtype.

1276-Pos Board B46
Probing the Nickel Coordination Environment in Helicobacter Pylori NikR
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Nickel homeostasis in gram-negative bacteria is often regulated at the transcriptional level by the nickel-dependent protein NikR, which belongs to the ribbon-helix-helix DNA-binding protein family. Generally, NikR is found as a homotetramer, with four nickel ions bound of the tetrameric interface at a metal binding domain (MBD). In Helicobacter pylori, NikR (HpNikR), unlike homologous NikR proteins, sits at a regulatory hub and has been found to have a variety of regulatory roles. HpNikR has been implicated in the regulation of proteins that function in acid acclimation, nickel uptake, virulence, and stress response. There are a number of questions about the nickel coordination environment in HpNikR due to a series of puzzling and sometimes contradictory structural and experimental results. Here we present results from complimentary computational and experimental techniques to help further resolve these questions. Specifically, we use molecular dynamics simulations and spectroscopic experiments to investigate the nickel coordination environment. We use circular dichroism to observe differences in the thermal stability of HpNikR depending on the nickel:-protein stoichiometry, with stabilization increasing as nickel concentration increases. Stability of HpNikR is found to be pH dependent. Elucidation of details about nickel coordination in HpNikR will improve our understanding of the functional role of genetic regulatory pathways in H. pylori pathogenicity.

1277-Pos Board B47
Toward Unraveling the Structure and Molecular Basis of Ferrous Iron Transport of Feo from *Klebsiella Pneumoniae*
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Feo from *K. pneumoniae* is a unique type of ferrous iron transporter for bacteria. It consists of three proteins, FeoA, a small SH3-domain like protein; FeoB, a large protein consisting of a cytosolic N-terminal G-protein domain and a C-terminal transmembrane domain that may serve as a Fe^{++} permease; and FeoC, a small putative transcriptional factor. Using X-ray crystallography and NMR we have solved the structure of FeoA, the intracellular domain of FeoB (NFeoB), FeoC and NFeoB/FeoC complex. In the structures, a canonical G-protein domain (G domain) is followed by a helical bundle domain (S-domain), which despite its lack of sequence similarity between species is structurally conserved. In the nucleotide-free state, the G domain's two switch regions point away from the binding site. This gives rise to an open binding pocket whose shallowness is likely to be responsible for the low nucleotide-binding affinity. Nucleotide binding induced significant conformational changes in the G5 motif which in the case of GMPPNP binding was accompanied by destabilization of the switch I region. In addition to the structural data, we demonstrate that Fe(II)-induced foot printing cleaves the protein close to a putative Fe(II)-binding site at the tip of switch I, and we identify functionally important regions within the S-domain. Moreover, we show that NFeoB exists as a monomer in solution, and that its two constituent domains can undergo large conformational changes. The data show that the S-domain plays important roles in FeoB function. The structures and their functional implications of FeoA, FeoC and NFeoB/FeoC complex will be discussed also.

1278-Pos Board B48
Structural Basis of the Specific Functions of Proteins Secreted from Mammary Gland during Different Progressive Stages such as Proliferative, Early-Lactation, Late-Lactation and Involution Phases
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The mammary gland undergoes repeated cycles of pregnancy, lactation and involution. Throughout these cycles, the interactions of mammary gland epithelial cells with their surrounding extracellular matrix (ECM), in particular the specialized ECM known as basement membrane (BM), contributes to the signals required for their proliferation, differentiation and survival. The structural and functional studies of prominent proteins secreted during these various phases have been carried out. The proteins included in these studies are insulin growth factor (IGF), lactoferrin (LF), Lactoperoxidase (LP), peptidoglycan recognition protein (PGRP) and signaling glycoproteins (SPX-40). Three-dimensional structural determinations using X-ray crystallographic methods