compensation a general characteristic of ssRNA binding, an inherent property of Py tracts, or a signature of sequence-specific ssRNA recognition? These possibilities are clarified by thermodynamic comparison of purine-tract association by these Py tract splicing factors, contrasted with the purine-specific protein PAB.

327-Pos Board B206

The DNA Bridging Protein H-NS and the SsrB Transcription Factor Counteract One Another to Silence and Activate Pathogenicity Island Genes in Salmonella

Yinjie Liu[1], Don Walthers[2], **Linda J. Kenney[2]**, Jie Yan[1].
[1]National University of Singapore, Singapore, Singapore, [2]University of Illinois-Chicago, Chicago, IL, USA.

The transcription factor SsrB activates transcription of genes located on Pathogenicity Island 2 (SPI-2) in *Salmonella enterica*. These gene products are responsible for forming a type-three secretion system that secretes effectors that modify the host macrophage vacuole, enabling *Salmonella* to replicate and then disseminate to the liver and spleen. Recent studies have reported that pathogenicity island genes are silenced by the nucleoid-like protein H-NS[1,2]. How transcription factors counter or relieve H-NS silencing is a major focus of study. The *sifA* gene is located outside of SPI-2 and encodes a product required for maintenance of the *Salmonella*-containing vacuole, providing an intracellular niche conducive to *Salmonella* replication and survival. Part of this process involves formation of *Salmonella*-induced filaments (Sifs). We set out to determine whether SsrB directly activates expression of the *sifA* gene, and whether H-NS could counter or prevent this interaction. In vitro transcription assays indicate that SsrB directly activates *sifA* transcription and this stimulation is prevented in the presence of H-NS. SsrB activation requires super-coiled templates; in the presence of linear DNA, no *sifA* transcripts are observed. Using atomic force microscopy, we show that H-NS forms multiple bridging complexes on super-coiled DNA. Additional experiments are underway to examine the effect of SsrB on these complexes. Supported by NIH GM-58746 and NSF MCB-0613014 to LJK and NUS R144000171712 to Y.J.

1. Navarre, W.W. *et al.*, (2006) Selective silencing of foreign DNA with low GC content by the H-NS protein in *Salmonella*. Science 313:236-238.
2. Lucchini, S. et al., (2006) H-NS mediates the silencing of laterally acquired genes in bacteria. PLOS Pathog 2:e81-89.

328-Pos Board B207

TBP Carries Out Specific DNA Binding Involving Information From Both Grooves

Cesar Millan-Pacheco, Victor M. Capistran-Licea, Nina Pastor.
Universidad Autonoma del Estado de Morelos, Cuernavaca, Morelos, Mexico.

The TATA binding protein (TBP) is a basal transcription factor that binds specifically to the minor groove of TATA boxes. TBP is required for efficient recruitment of the transcription machinery, as it bends DNA, generating binding sites for TFIIB and RNA polymerase II. Only ~10% of human protein coding genes possess a TATA box, and TBP is required for the transcription of the more common TATA-less genes also. We explored the energy contributions to TBP binding of a collection of 16 repeating DNA sequences, in a productive bent conformation. Binding energy is dominated by the cost of deforming DNA from the straight, B-DNA reference, to the conformation found in the complex. Interestingly, there is a positive correlation between the deformation energy of DNA and the interaction energy of DNA with TBP: poor interaction energies, derived mainly from avoided clashes with guanine amino groups and hydrophobic TBP sidechains, lead to greater deformation energies, due to clashes between exocyclic groups at the major groove of DNA. In order to uncouple deformation and interaction, we substituted guanines with inosines, eliminating putative clashes at the minor groove while keeping the major groove chemistry. Improving the interaction also ameliorated the deformation cost. Large deformation costs are incurred because we kept the complex structure fixed to a productive conformation. As high affinity binding has been shown not to be correlated always to transcription efficiency, we carried out molecular dynamics simulations of three TBP-DNA complexes, with alternating TA, CG and CI sequences. Preliminary results suggest that a large deformation cost is relieved by relaxing both TBP and DNA structures, generating increasingly unproductive complex structures.

Funding: Macroproyecto de Tecnologías de la Información y la Computación (UNAM). SEP-FOMES 2000 (Cómputo Científico), and CONACyT (91209 AC1).

329-Pos Board B208

Recognition and Signaling in DNA Mismatch Repair: MD Studies of MutS Complexes with DNA and ATP

Susan N. Pieniazek, Manju Hingorani, David L. Beveridge.
Wesleyan University, Middletown, CT, USA.

The MutS family of DNA binding proteins has been reported to play a critical role in mismatch repair (MMR). Crystal structures of MutS (*Escherichia coli* and *Thermus aquaticus*) as well MSH homologs including human MutSα reveal intricate and complex multi-domain protein structures comprised of greater than 1,500 residues. The DNA binding domain of these proteins recognizes mispaired or unpaired bases. It has been proposed that this recognition event results in the release of a signal that travels from the DNA binding domain to the ATPase site. While much has been learned from previous binding studies of MutS, the contribution of the protein dynamics on MutS complex formation and intraprotein communication events are not fully resolved at the atomic level. In this study, state-of-the-art molecular dynamics (MD) simulations are used to investigate the dynamical processes that occur during the interactions with DNA and ATP substrates. In particular, we are interested in how the DNA mismatch recognition/binding event is signaled, triggering the initiation of DNA repair. The computational challenge represented by the size and complexity of MutS-DNA complexes provides an opportunity to develop MD approaches for large multi-component biological systems.

330-Pos Board B209

In Silico Study Of Nonspecific DNA-protein Encounter Complexes

Mu Gao, Jeffrey Skolnick.
Georgia Tech, Atlanta, GA, USA.

In search of its specific targets, a DNA-binding protein associates with nonspecific DNA and subsequently diffuses along the DNA. Structural characterization of the nonspecific DNA-protein encounter complexes is of great interest. Due to weak interactions between the protein and nonspecific DNA, however, such characterization is experimentally challenging. Here, we describe the first comprehensive computational study on the encounter complexes of 44 specific DNA-binding proteins with nonspecific canonical B-DNA. In the analysis of these encounter complex models, we found that the recognition sites for specific DNA are usually favorable interaction sites for the nonspecific DNA probe and that nonspecific DNA-protein interaction modes exhibit some similarity to specific DNA-protein binding modes. These results led us to a novel method that predicts DNA-binding sites and binding-modes for a DNA-binding protein without knowing its specific DNA target sequence. In benchmark tests, the method achieves significantly better performance than three previously established methods, which are based on sophisticated machine-learning techniques. We further apply our method to protein structures predicted through modeling and demonstrate that our method performs satisfactorily on protein models up to 5 Å from their native structures.

331-Pos Board B210

Understanding DNA- and RNA-binding Proteins Using Sequence and Structural Features

Matthew B. Carson, Robert Langlois, Hui Lu.
University of Illinois at Chicago, Chicago, IL, USA.

The complex interactions between nucleic acid molecules and NA-binding proteins are an integral component of the gene regulation processes. Understanding which residues on these proteins bind nucleic acids is necessary in order to characterize these interactions. Due to the complicated nature of this problem, machine learning is often used to predict which residues are involved in the act of binding. The prediction of NA-binding residues can provide assistance in the functional annotation of NA-binding proteins. These predictions can also be used to expedite mutatgenesis experiments for the study of NA-binding proteins, guiding researchers to the correct binding residues in these proteins. In this work we focus on three goals. First, we use SVM and various ensemble methods based on the C4.5 decision tree algorithm to predict DNA- and RNA-binding residues within proteins with high, balanced accuracy by analyzing sequence and structural characteristics. Secondly, we show that our classifiers can achieve similar results on several data sets which were used in previous works to identify DNA- and RNA-binding residues. Thirdly, we show that we are able to distinguish DNA-binding residues from RNA-binding residues using structure- + sequence-based features and sequence-based features only.