2466-Pos
Theoretical Modeling of Protein Accessibility to the Chromatin Fiber
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Gene expression is orchestrated by a host of regulatory proteins that coordinate the transcription of DNA to RNA. Regulatory proteins function by locating specific sequences of DNA and binding to these sequences to form the transcription initiation complex. The eukaryotic genome is tightly packaged into a dense chromatin fiber. This packaged structure acts both to store the massive genome and to facilitate the accessibility of the genome to regulatory proteins. The interplay between the packaging proteins and the regulatory proteins is critical in normal cellular function and plays a pivotal role in a number of human diseases. We present a theoretical study on the dynamic accessibility of the chromatin fiber, providing an overview of several approaches to this multi-faceted problem. Our modeling efforts address both the structural properties of packaged DNA and the dynamic processes involved in target-site localization of regulatory proteins. Modeling of the 30-nm fiber reveals the impact of local nucleosome configurational properties on the fiber geometry, and we predict the mechanical properties of the assemblies and the resulting dynamic accessibility. We also discuss the role of linker histones and variant histones in the fiber assembly. Using our 30-nm fiber models, we address large scale condensation by epigenetic factors and discuss the role of histone methylation in the determination of heterochromatin and euchromatin states. Upon establishing our chromatin model, we study the dynamic processes involved in the target-site search of regulatory proteins within the complex chromatin structure. This effort combines our structural models with our protein-transport models in order to provide a novel perspective on regulatory-protein function.

2467-Pos
Nucleosome Organization is Quantitatively Described by Statistical Positioning Up- and Downstream of Transcription Start Sites
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The positions of nucleosomes in eukaryotic genomes determine which parts of the DNA sequence are readily accessible for regulatory proteins and which parts are not. A salient feature in recent genome-wide nucleosome maps is that nucleosomes appear well-positioned around a nucleosome free region (NFR) just upstream from the transcription start site (TSS). What determines this nucleosome organization is not known. One scenario is that the majority of nucleosome positions near the TSS are directly encoded in the DNA sequence. The alternative ''statistical positioning'' scenario, is that a few local barriers on the genome strongly constrain the positions of closeby nucleosomes, purely on statistical grounds. We use a physical model for the latter scenario, based on the Tonks gas of statistical physics, to quantitatively analyze recent data for yeast. We find that although the typical patterns on the two sides of the TSS are different, they are both quantitatively described by the same physical model, with the same parameters, but different boundary conditions. The inferred boundary conditions suggest that the first nucleosome downstream from the NFR is typically directly positioned while the first nucleosome upstream is statistically positioned via a nucleosome-repelling DNA region.

2468-Pos
DNA Loop Formation in Nucleosomes
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Many key processes in the cell nucleus, such as replication, transcription or DNA-repair, require the physical accessibility of specific DNA sequences. In eukaryotes, DNA is wrapped in 1.67 left-handed superhelical turns around a histone protein core to form the nucleosome. Although the function of histones in eukaryotes remains elusive, it is possible that histone mobility on DNA is responsible for 'opening up' DNA surfaces at different periods of chromatin organization. One of the mechanisms of nucleosome repositioning on DNA involve formation of local defects in the form of a loop and diffusion of it over the stretch of DNA attached to the histone. DNA loop formation has so far been discussed in the context of the worm like chain (WLC) model [1]. However, atomic force microscopy experiments [2] suggest that large angle bending energetics of DNA does not follow a WLC. Here we describe the energetics of loop formation on nucleosomes using a model which offers softer bending potential for large deflections, namely the sub-elastic chain (SEC), and compare with WLC. Results show that SEC favors small loop (~ 10 bp) formation, WLC favors large loop formation. Different energetics of loop formation also leads to different nucleosome repositioning behaviors for WLC and SEC. Nucleosomes, initially positioned in the middle of a DNA segment, jumps to the extremities for WLC and to the nearest neighboring positions for SEC.
1. Kulic I. M. , Schiessel H. 2003. Nucleosome repositioning via loop formation. *Biophys J.*, 84(5):3197-211.
2. Wiggins et. al. 2006. High flexibility of DNA on short length scales probed by atomic force microscopy. *Nature Nanotechnology*.1.137.

2469-Pos
The Third Level of Genome Functioning: Chromatin Folding
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Recent experiments have provided us with extensive amount of data which all suggest that chromatin folding is achieved by formation of chromatin-chromatin loops. This observed looping is highly dynamic and linked to cell differentiation. We propose that the chromatin folding constitutes a third level of genome functioning on top of the individual genes and their epigenetic control. Our interest is to study the principles of chromatin folding in the cell nucleus, above all how does chromatin folding relate to gene expression. In this we focus on the molecular mechanisms of formation of loops, dynamics and ergodicity of looping as well as to relation of dynamic looping to genome function. Our experimental approach relies on three-dimensional cell imaging to map the in vivo folding state of chromatin. For the interpretation of our data we use polymer-physics models. The most recent work of our group shows that chromatin folding status is different in genomic regions with higher transcription levels and in transcriptionally silent regions. In both cases the data can be fitted to a "random loop" polymer model(1).
1. Mateos-Langerak, J., Bohn, M., de Leeuw, W., Giromus, O., Manders, E.M.M., Verschure, P. J., Indemanns, M.H.G., Gierman, H.J., Heermann, D.W., van Driel, R., Goetze, S. 2009. Spatially confined folding of chromatin in the interphase nucleus. Proc Natl Acad Sci U S A. 106:3812-7.

2470-Pos
The Chromatin-Remodeling Complex ACF Functions as a Dimeric Motor to Space Nucleosomes
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The chromatin structure at a given locus is a key determinant of its transcriptional state. Evenly spaced nucleosomes directly correlate with condensed chromatin structures and gene silencing. The ATP-dependent chromatin assembly factor (ACF) generates such structures in vitro and is required for transcriptional silencing in vivo. ACF generates and dynamically maintains nucleosome spacing by constantly moving a nucleosome towards the longer flanking DNA faster than the shorter flanking DNA. But how the enzyme rapidly moves back and forth between both sides of a nucleosome to accomplish such bidirectional movement is not known. Using FRET to follow disruption of histone-DNA interactions in real time we show that nucleosome movement depends cooperatively on two ACF molecules, suggesting that ACF functions as a dimer of ATPases. Employing Electron Paramagnetic Resonance (EPR) to resolve different populations of the nucleosome-ATPase complex, we find that the nucleotide state determines whether the dimer closely engages one vs. both sides of the nucleosome. Furthermore three-dimensional reconstruction by single particle electron microscopy of the ATPase-nucleosome complex in an activated ATP state reveals a dimer architecture in which the two ATPases bind facing each other. Our results suggest a model in which the two ATPases work in a coordinated manner, taking turns to engage either side of a nucleosome. Such a mechanism would allow rapid sampling of both sides of the nucleosome and allow bidirectional movement without dissociation. This novel dimeric motor mechanism differs from that of other dimeric motors such as kinesin and dimeric helicases that processively translocate in one direction and reflects the unique challenges faced by motors that move nucleosomes.

2471-Pos
Role of DNA Fluctuations in RNA Polymerase Translocation through a Single Nucleosome
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We analyze two aspects of the physical behavior of a single nucleosome: the response of a single nucleosome core particle to tension and the translocation