2966-Pos Board B13

Disruptions of the Regular Structure in DNA Minicircles

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DNA bending and torsional deformations, which often occur during its functioning inside the cell, can cause local disruptions of the regular helical structure. The disruptions created by negative torsional stress have been studied in detail, but those caused by bending stress have only been analyzed theoretically. To address the problem, we probed the structure of very small DNA circles, 63 - 105 bp in length, by single-strand-specific endonucleases. We determined that bending stress disrupts the regular helical structure when the radius of DNA curvature is smaller than 3.5 nm. The experimental data suggest that strong DNA bending initiates kink formation while preserving base pairing. To get quantitative information about the disruptions we developed a statistical-mechanical model of the disruption formations in DNA minicircles. The model, used in the computer simulation, specifies the disruptions by three parameters: DNA bend angle at the disruption θ_d, local DNA unwinding caused by the disruption formation, and the free energy associated with the disruption formation in unstressed double helix, G_d. We obtained a relationship between values of G_d and θ_d under which the theoretical results are compatible with the experimental data. The relationship suggests that the free energy of a base pair opening, which includes flipping out both bases, is significantly higher than the generally accepted value.

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Twist and Tension-mediated Elastic Coupling between DNA-Bending Proteins

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The mechanical properties of DNA are harnessed through bending, looping, and twisting by proteins that play critical roles in both reading and packaging the genome. Cooperative binding of such proteins is required to assemble transcription factor complexes in promoter regions and to condense DNA into higher-order chromatin structures. We study, using theoretical modeling, the effective interaction between DNA-bending proteins that arises from elastic stresses in the linker when the DNA is subject to tension. Our model for the cooperative effects of introducing multiple bends into DNA utilizes the worm-like chain formalism with twist resistance and external force. We find that the coupling free energy between two proteins exhibits damped oscillations with increasing distance between the binding sites, due to a preference for the proteins to align in an anti-parallel fashion along a DNA helix that resists twist. The oscillations are superimposed on an attractive potential arising from the bending deformation energy, which promotes aggregation to minimize the curvature of the linker DNA. These results allow us to calculate the mean first encounter time for bending proteins sliding along DNA and indicate an optimal applied tension for protein assembly. Furthermore, our calculations are used to study the unwrapping transitions of a dinucleosome under tension. Our results highlight the need to consider DNA twist even when no torsion is applied and the DNA ends are free to rotate. We demonstrate a variable-range oscillatory coupling between DNA-bending proteins that may provide a versatile potential mechanism for tension-mediated gene regulation.

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Dynamic Conformation Fluctuations of λ-Phage DNA in an Optical Trap

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Dynamic fluctuations in the conformation of λ-phage DNA have been observed in a fiber optic based light trap. The light force is used to confine a polystyrene bead (diameter, 0.5μm - 2.8μm) in three dimensions at a fixed distance from a flat surface coated with anti-digoxygenin antibody. The DNA is fixed to this bead through a biotin - streptavidin linkage on the 3′end, whereas a digoxygenin label is present on the opposing 5′. Through this simple assay, the dynamics of the free ends have been quantified for various times and separation distances. The dynamics have been observed using a high frame rate camera and the intercalating fluorophore, YOYO-1. In contrast to a conventional optical trap, this trap lacks a highly focused beam, greatly reducing localized heating and the associated risk to the biological specimen which could potentially cause error in this type of measurement.

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Simulations of Copper-1,10-Phenanthroline Complexes Binding the DNA

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Copper complexes of 1,10-phenanthroline (phen) are chemical nucleases employed as footprinting reagents for determining ligand binding sites. The cleavage activity of the parent complex, $Cu(phen)_2$, occurs according to the following mechanism: a) reduction of $Cu(phen)_2^{2+}$ to $Cu(phen)_2^{+}$; b) non-coordinative binding of $Cu(phen)_2^{+}$ to DNA; c) $Cu(phen)_2^{+}$ oxidation to $Cu(phen)_2^{2+}$ by H_2O_2, and formation of Cu-''oxo'' and/or Cu-''hydroxyl'' species; d) oxidative attack leading to DNA-cleavage.

However, the potential clinical use of the parent compound is mainly prevented by two drawbacks: i) the low binding constant of the second phenanthroline; ii) the modest sequence selective DNA cleavage.

To improve $Cu(phen)_2$ efficiency, Pitié *et al.* used a serinol bridge to link the two phen rings leading to Cu(2-Clip-phen) and Cu(3-Clip-phen) derivatives, which cleave the DNA 2 and 60 times more efficiently than $Cu(phen)_2$. To address the modest sequence selectivity, the amine group of the serinol bridge was functionalized with sequence specific DNA minor/major-groove binding ligands such as cisplatin- and distamycin-like compounds, leading to encouraging results.

In this work, a combination of theoretical methods, including DFT, Docking and Molecular Dynamics, was employed to i) characterize the DNA binding of these complexes and ii) to determine the origin of their diverse DNA-cleavage efficiency. Our simulations clearly revealed that several factors such as planarity of the ligand, better interaction with DNA and minor-groove fit, contribute to the enhanced efficiency of Cu(3-Clip-phen) compared to the other structurally similar complexes.

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P53 Induced Dna Bending

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Specific p53 binding-induced DNA bending and its underlying driving forces are crucial for the understanding of selective transcription activation. Diverse p53-response elements exist in the genome; however, it is not known how p53 specific binding induced DNA bending and DNA sequence influences the bending extent. Molecular dynamics simulations were performed on a series of p53 core domain tetramer-DNA complexes with various DNA sequences with difference in the central 4-base pairs of each half site to compare the DNA bending extent and pinpoint the underlying driving forces. The results show that the specific interactions between p53 dimer and DNA and between p53 dimers triggered intra- and inter-half sites DNA bending . The central 4 base pairs were important to the bending extent due to its variant flexibility. Specifically, the more flexible CATG containing DNA was able to maintain the specific interactions with the p53 including those from residues Arg280, Lys120 and Arg248 while those with CTAG that had low flexibility were less capable of maintaining the specific interactions . As a result, base pairings for the CATG sequence were stable throughout the simulation trajectory while those for the CTAG sequence was partially dissociated for part of the trajectory, which affected the stability of nearby Arg280-Gua base interactions. Thus, DNA bending was induced by the specific p53-DNA and p53 dimer-dimer interactions and the bending extent was dependent on DNA sequence that was correlated with its flexibility and ability to maintain specific interactions in bent conformations.

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DNA Relaxation Dynamics in 1ID3 Yeast Nucleosome MD Simulation

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Nucleosomes are elemental structural units envolved in the formation of chromatin. Organization of chromatin has a profound effect on DNA transcription, replication, repair and recombination. Nucleosomes not only participate in the compaction of the genetic material but also regulate gene expression by controlling the accessibility of specific DNA binding sites to proteins. There are 33 crystal structures of nucleosomes currently available in the protein databank. Nucleosome typically consists of a 147 bp dsDNA wrapped around an octameric histone protein, (H2A.1-H2B.2)(H3-H4)2(H2A.1-H2B.2), in 1.65 left-handed superhelical turns. DNA interacts with histone octamer at 14 locations (every ~10 bp), forming a total of ~240 direct and indirect contacts and ~120 hydrogen bonds. Molecular Dynamics simulation is a useful technique for exploring dynamics at interaction