DNA Structure and Dynamics II

1398-Pos Board B128
The Effects of Base Stacking on DNA Flexibility
Lauren S. Mogil, Justin P. Peters, Jim Maher.
BMB, Mayo Clinic, Rochester, MN, USA.
DNA is the main genetic component of life and understanding its basic properties is crucial for advancement in applied sciences. DNA, a long macromolecule, must complex with proteins and other molecules for packaging and condensing to fit in the nucleus of a cell. Persistence length is a physical property that defines the stiffness of a polymer, ranging from rod-like to string-like. The persistence length of double-stranded DNA is ~150bp or ~50nm making it a relatively inflexible molecule. It is hypothesized that the two main contributors limiting DNA flexibility are mutual charge repulsions along the DNA backbones and attractive base stacking interactions. However, the relative contributions to DNA stiffness of electrostatic repulsion and base stacking forces are unknown. We use experimental T4 DNA ligase-mediated cyclization kinetics experiments to measure the physical properties of DNA, specifically DNA longitudinal and torsional flexibilities and helical repeat.
We measured the impact on DNA stiffness of enhancing or diminishing stacking energy involving modified adenine and guanine bases. Diaminopurine (DAP) is hypothesized to increase DNA bending stiffness by changing the base geometry and dipole moment so as to enhance stacking energy and inosine (dITP) is hypothesized to decrease DNA bending stiffness. Cyclization assays were performed with radiolabeled DNA probes varying in length from 211bp to 201bp. The free ends of the probes are complementary and, in the presence of T4 DNA ligase, yield various linear and circular species in vitro. The J-factor (related to persistence length through the worm-like chain polymer model) can be estimated in such assays. We found that the bending persistence length changed slightly relative to natural DNA but it is the twist persistence length that has a more dramatic change.

1399-Pos Board B129
Conformational Transition of Nanoslit Confined DNA at Low Ionic Strengths
JinYong Lee[1], Kyubong Jo[2].
[1]Department of Chemistry, Sogang UNIV, Seoul, Korea, Republic of, [2]Deparrtment of Chemistry and Interdisciplinary Program of Integrated Biotechnology, Sogang UNIV, Seoul, Korea, Republic of.
We investigate experimentally the effect of ionic strength on λ DNA molecule in glass/PDMS nanoslit by using fluorescence microscope. The radius of gyration Rg was measured as a function of slit height (250 ~ 1049 nm), and ionic strength (0.183 ~ 1.75 mM) as a result, we observed the Rg values increases as channel heights and ionic strengths decrease. Furthermore, based on Odijk's and de Gennes polymer physics theory, we interpret our experimental findings as a function of channel heights and ionic strengths. We also study the effect of EDTA concentration, radius of gyration of λ DNA change drastically in response to the varying EDTA concentration at low ionic strength condition. Our results are useful for understanding the manipulation of biomolecules in nanofluidic devices.

1400-Pos Board B130
Probing Sequence and Topological Specificity in the Binding of Tetra(Methylpyridyl)Porphines to DNAs
Stephen A. Winkle[1], Jennifer Barretta[1], Diane Edgar[1], Raul Castillo[1], Olga Roman[1], Roxana Roque[1], Jessica Millar[2], Maria Ballester[2].
[1]Chemistry and Biochemistry, Florida International University, Miami, FL, USA, [2]Math, Science and Technology, Nova Southeastern University, Fort Lauderdale, FL, USA.
The binding of a family of tetra(methylpyridyl)porphines to relaxed and supercoiled circular phiX197RF DNA has been probed using restriction endonuclease activity assays employing nine restriction enzymes (selected for different cleavage sequences and different flanking sequences), mung bean nuclease assays and E.coli topoisomerase assays. The restriction enzyme assays indicate that placement of the methyl substituent alters the sequence and topological specificity of binding. For example, the the restriction enzyme Mlu I (cleavage site ACGCGT), the ortho isomer yields partial inhibition of cleavage of supercoiled and relaxed DNA, the meta isomer yields inhibition of the relaxed DNA and no effect with supercoiled DNA, and the para isomer gives enhanced cleavage of supercoiled and relaxed DNA. With Dra I (TTTAAA), the ortho isomer again gives partial inhibition cleavage of supercoiled and relaxed DNA, the meta isomer gives inhibition of cleavage of relaxed AND supercoiled DNA, and the para isomer has no effect on cleavage of either form. Variations in flanking sequences affect the selectivity in binding. Mung bean digestion assays indicate that all three isomers alter the conformation of the supercoiled DNA, with the ortho isomer giving the greatest distortion. Topoisomerase studies suggest that these molecules unwind the DNA. Molecular modeling suggests that the placement of the methyl substituent on the pyridyl rings affects the planarity of the molecules, which may alter the binding to DNA and lead to the observations above.

1401-Pos Board B131
Molecular Dynamic Studies of Z[WC] DNA and the B to Z-DNA Transition
Michael G. Lerner, **Jinhee Kim**, Alexander K. Seewald.
Earlham College, Richmond, IN, USA.
Although DNA is most commonly found in the right-handed B-DNA structure, it is known that biologically active systems also contain left-handed ZII-DNA. We investigate the possibility that Z[WC]-DNA serves as an intermediate structure in the B to ZII transition. Molecular dynamics simulations indicate that Z[WC] nonamers are stable structures with the current AMBER nucleic acid force field. Steered molecular dynamics simulations indicate that, for collective transitions of the whole strand, the B-Z[WC]-ZII pathway may have a lower free-energy barrier than the direct B-ZII pathway. We then used both steered and targeted molecular dynamics in combination with umbrella sampling to produce potentials of mean force for the B to ZII transition along both pathways.

1402-Pos Board B132
Nanoconfined Circular DNA
Mohammadreza Alizadehheidari[1], Erik Werner[2], Charleston Noble[3], Lena Nyberg[1], Joachim Fritzsche[4], Bernhard Mehlig[2], Jonas Tegenfeldt[5], Tobias Ambjörnsson[3], Fredrik Persson[6], Fredrik Westerlund[1].
[1]Department of Chemical and Biological Engineering, Chalmers University of Technology, Göteborg, Sweden, [2]Department of Physics, Gothenburg University, Göteborg, Sweden, [3]Department of Astronomy and Theoretical Physics, Lund University, Lund, Sweden, [4]Department of Applied Physics, Chalmers University of Technology, Göteborg, Sweden, [5]Department of Solid State Physics, Lund University, Lund, Sweden, [6]Department of Cell and Molecular Biology, Uppsala University, Uppsala, Sweden.
Nanofluidic channels have become a versatile tool to manipulate single DNA molecules. They allow investigation of confined single DNA molecules from a fundamental polymer physics perspective as well as for example in DNA barcoding techniques.
Covalently closed circular DNA is found in many biologically relevant contexts, such as bacterial plasmids, viruses and eukaryotic mitochondrial DNA. Circular DNA is also interesting from a fundamental perspective. The circular topology forces two strands to be close to each other in the nanochannel, which changes the polymer physics behavior compared to linear DNA. Circular DNA is difficult to study with traditional single molecule techniques because it generally requires the attachment of handles.
Circular DNA in its folded state has less entropy and higher conformational free energy than the unfolded state. Unfolding of circular DNA is therefore entropically favorable. As a double-strand break occurs, circular DNA opens up and unfolds to its linear conformation inside the nanochannel. This study deals with the statics and dynamics of this transition from circular to linear DNA.
We observe that the opening of the circle has a strong preference to occur in the ends and discuss the mechanism behind this peculiar observation. We also investigate the difference in extension between the circular and linear state. We observe that their ratio decreases with decreased stretching, in agreement with theoretical calculations.
Finally we demonstrate how we can identify bacterial plasmids carrying antibiotic resistance genes using a recently developed mapping technique based on nanofluidic channels.

1403-Pos Board B133
Simulations of Crosslinking Efficiency and Sequence Specificity of Nitrogen Mustard Anticancer Drugs
Moon Joon Park, Michael E. Colvin.
University of California, Merced, CA, USA.
Cytoxan and ifosfamide are important anti-cancer drugs that function by forming covalent crosslinks between the two strands of double helical DNA. The cytotoxicity of these drugs depends on their efficiency in forming interstrand crosslinks versus intrastrand crosslinks or non-crosslinking DNA damage which can cause mutations in normal cells leading to future cancers. To better understand the factors favoring interstrand crosslinks, we have performed a series of near-microsecond timescale molecular dynamics simulations of different sequences of DNA 11-mers bound to the activated