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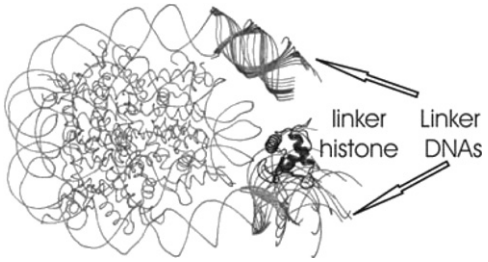
The linker histone is a protein that binds to the nucleosome and determines how the nucleosomes are linked to each other. To simulate the nucleosome-linker histone interactions, we applied a Brownian Dynamics (BD) technique together with normal mode analysis (NMA). NMA of the nucleosome revealed the most prominent modes of motion of its two linker DNAs. The results were used to generate conformations of the linker DNAs which were used in BD simulations of docking of a linker histone and its mutants to the nucleosome. From the simulations, two distinct binding sites on the linker histone were identified. The residues found to be most important for binding in the simulations with the linker histone mutants are consistent with experimental data. Moreover, a unique binding mode of the linker histone to the nucleosome was found for a wide range of conformations of the linker DNAs. As well as providing insights into the determinants of linker histone-nucleosome binding, the results are valuable for higher-order modelling of the chromatin.



431-Pos Board B310
Exploring The Spatiotemporaloral Dynamics of DNA Binding and Cleavage by Restriction Endonucleaes
Wei-Ting Wang, Jing-Shin Tsai, Chien-Ting Hsu, Tzu-Sen Yang*.
Taipei Medical University, Taipei, Taiwan.
Using restriction endonucleases to catalyze the double-stranded DNA (dsDNA) breakage at certain recognition sequences is an important molecular biology technique. The restriction endonucleases constitute an important defense mechanism of bacteria against viral attacks; this mechanism is to destroy invading foreign DNA molecules via cleaving a specific site (phosphodiester bond) of a dsDNA. By cleaving recognition sites on dsDNA with extraordinary specificity can lead to the DNA double strand breaks (dsb). We presented a novel single-molecule approach to investigate the interaction between DNA and restriction endonucleases, including DNA recognition and cleavage. To elucidate how fast restriction endonucleases recognize and cleavage DNA sequence, we constructed a high resolution dual-beam laser tweezers system to manipulate single DNA molecule, together with the site-specific restriction enzymes, namely, EcoRI (one-site endonuclease) and Cfr9I (two-site endonuclease), conjugated to nanometer-sized fluorescence particle. Because most endonucleases work in the presence of magnesium ions, we will apply optically based reaction mechanism to control and synchronize the restriction endonuclease activity in this study. Furthermore, both laser tweezers and fluorescence particle imaging will be used to probe whether the DNA double strand breaks occurred due to the molecular cutting. Hence, this single-molecule approach allows us to directly observe and visualize the spatiotemporal dynamics of DNA binding and cleavage by restriction endonucleases, and can be further applied to determine the DNA cleavage rate due to the presence of EcoRI and Cfr9I. Finally, we extend this approach, together with the light-induced molecular cutting, to investigate the DNA binding and cleavage by restriction endonucleases under tension at different temperatures.

432-Pos Board B311
Molecular Recognition Routes Of DNA By Anticancer Ligands: Mechanisms and Free Energies Explored Via Molecular Dynamics Simulations
Attilio Vittorio Vargiu[1,2], Alessandra Magistrato[3], Paolo Carloni[3], Paolo Ruggerone[1].
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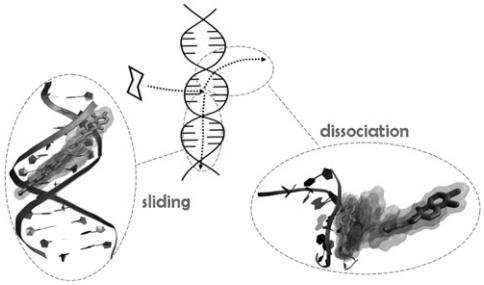
Molecular recognition of the DNA minor groove is a multi-route process which can involve many steps before the formation of the most stable adduct. In particular, many studies have pointed out the importance of events like sliding along the groove and dissociation (which is a relevant step in the translocation among different sequences) for the affinity and the specificity of minor groove binders.
In this contribution we present our recent work on the subject. Umbrella sampling and metadynamics were used to characterize mechanisms and free energy profiles of molecular recognition routes by the antitumoral agents anthramycin, duocarmycin and distamycin. Our results are in very good agreement with the available experimental data, and provide insights on the influence of factors like size, charge and flexibility on the molecular recognition process.



433-Pos Board B312
Computational Studies of Substrate Binding and Conformational Change in the Glycine Betaine Symporter BetP
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Max-Planck Institute of Biophysics, Frankfurt am Main, Germany.
The glycine betaine symporter BetP is an important protein for regulation of osmotic pressure in the microbe *Corynebacterium glutamicum*, a bacterium used extensively in biotechnology. BetP responds to changes in external osmolality by regulation of its transport activity. The recently solved X-ray crystallographic structure of this protein reveals that it is a homotrimer and that each monomer possesses its own substrate binding pocket. Available structural data for several secondary transporters suggest that these proteins may adopt one of several structurally-distinct states, namely outward- and inward-facing state conformations, as well as a so-called occluded state. Since the monomers in the BetP X-ray structure do not show any apparent substrate pathways, they are likely to represent an occluded state. To identify structural features of alternate states, which may have relevance for a range of secondary transporters, we constructed 3D models of outward- and inward-facing states of BetP using secondary transporters of known structure as templates and validated the modelling results through the rigid-body fitting of these models to low-resolution cryo-EM maps. In addition, to address several other remaining questions, including: the location of glycine betaine and Na^+ binding sites; the effect of the headgroup size and net charge of lipid molecules; and the importance of the trimeric state of the protein, we have performed all-atom molecular dynamics simulations of BetP. Finally, we combined the results of structural and simulation studies with those from sequence analysis of BCCT transporters in order to identify structural and functional roles for several important residues. The results of our computational studies may lead to a better understanding of key events in the transport cycle and they are being validated experimentally.

434-Pos Board B313
Substrate Binding Directs the Functional Hinge Bending Motion of Human 3-Phosphoglycerate Kinase
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3-Phosphogycerate kinase (PGK) is a two domain enzyme, with a binding site of the 1,3-bisphosphoglycerate on the N-domain and of the ADP on the C-domain. In order to transfer a phosphate group the enzyme has to undergo a hinge bending motion from open to closed conformation to bring the substrates to close proximity. Molecular dynamics simulation was used to elucidate the effect of ligand binding onto the domain motions of this enzyme. The simulation results indicate the presence of a relatively small amplitude hinge bending motion of ns timescale in the apo form while the time period of the hinge bending motion of the complex form is clearly over the 20 ns simulation time. Upon binding the ligands, the hinge bending shows more directed characteristics with one dominant hinge point in the vicinity of the substrates while the apo form exhibits several hinge points that contribute to the hinge bending motion. The correlation of interdomain atomic movements also increased upon substrates binding.

435-Pos Board B314
Molecular Dynamics Simulation Study of T Cell Receptor Molecular Recognition of Peptide-Major Histocompatibility Complexes
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The Ohio State University, Columbus, OH, USA.