# BP 4: DNA & DNA Enzymes

Time: Monday 14:00–17:00 Location: ZEU 260

**Invited Talk** BP 4.1 Mon 14:00 ZEU 260

**Single-molecule detection of DNA repair in real-time** — •Terence Strick[1], Kevin Howan[1], Nigel Savery[2], Seth Darst[3], and MM2M FP7 Consortium[4] — [1]CNRS Institut Jacques Monod Paris, France — [2]University of Bristol, UK — [3]Rockefeller Institute, NY, USA — [4]Erasmus Univ., Rotterdam

We describe the bottom-up reconstruction of DNA repair pathways using single-molecule nanomanipulation methods. This allows us to observe in real-time the initial steps of DNA repair and build up kinetic models for repair processes. We discuss a variety of DNA repair systems and show in which ways these systems are mechanosensitive or not.

BP 4.2 Mon 14:30 ZEU 260

**Partitioning of RNA polymerases in bacterial cells** — •Stefan Klumpp[1], Marco Mauri[1], and Terence Hwa[2] — [1]Max Planck Institute of Colloids and Interfaces, Potsdam — [2]University of California, San Diego

How frequently a gene is transcribed depends not only on its regulation, but also on the availability of the necessary molecular machinery, RNA polymerases (RNAPs) and their associated factors. The concentration of free RNAPs and factors, i.e. those that are available for the initiation of transcription, depends also on the demand by other genes, such that genes may compete for the transcription machinery. We used a model for the partitioning of RNAPs into several functional classes to address the effect of this competition [1]. The model has been tested against existing experimental data for the growth-rate dependence of constitutive transcription and the effects of RNAP overexpression. We find that the competition of genes for RNAPs generally plays a minor role, because a pool of RNAPs non-specifically bound to DNA buffers against such effects. For sigma factors, the component of the transcription machinery required for promoter recognition and binding, however, competition seems to play an important role and may actively be modulated by the cell during global switches in the gene expression program, such as in stress responses.

[1] S. Klumpp and T. Hwa, Proc Natl Acad Sci USA 105, 10245 (2008).

BP 4.3 Mon 14:45 ZEU 260

**A model for the degradation of messenger RNA in bacteria** — •Carlus Deneke, Angelo Valleriani, and Reinhard Lipowsky — Max-Planck-Institut für Kolloid- und Grenzflächenforschung, Department of Theory and Bio-Systems, Potsdam, Germany

In a cell, the amount of messenger RNA (mRNA) is kept in balance by the processes of transcription and degradation. In the bacterium E. coli, the stability of mRNA is limited due to the action of protein complexes called the degradosome. They consist of several endo- and exonucleases which cooperatively degrade the mRNA chain until it is eventually fully recycled.

In this contribution, we present a theoretical model that takes into account the stochastic nature of this process. To build the model, we have assumed that in bacteria the main degradation pathway is initiated by endonucleolytic cleavage, according to the standard view in the field. It exploits the fact that the coverage of mRNA with ribosomes depends on the age of the transcript and that ribosomes shield the transcript against degrading proteins.

One consequence of the model is that the mean life time of the transcripts decreases with the length of the coding sequence. This conclusion is in agreement with many experimental half-life measurements. We will show a comparison of our model to experimental half-life data and critically discuss the nature of these data.

BP 4.4 Mon 15:00 ZEU 260

**A Stochastic Model of DNA Replication Dynamics** — •Daniel Löb and Barbara Drossel — Institut für Festkörperphysik, TU Darmstadt

Inspired by detailed cell-biological data on the dynamics of DNA replication during S phase, we present a stochastic model of DNA replication dynamics that allows for a quantitative comparison between model parameters and data.

An important ingredient of the model is the structuring of DNA into zones of euchromatin and heterochromatin, which have different rates of replication initiation. The sizes of these zones affect the time course of replication of the two chromatin types.

Further important model features are the induced initiation of replication in the vicinity of replication forks and a limitation of the number of replication forks due to the limited availability of essential replication proteins.

BP 4.5 Mon 15:15 ZEU 260

**Unfolding mechanisms and the free energy landscape of the DNA i-motif** — •Jens Smiatek and Andreas Heuer — Institut für Physikalische Chemie, Westfälische Wilhelms-Universität Münster, D-48149 Münster, Germany

Since the discovery of the DNA i-motif, the formation and function of this specific structure has attracted broad interest. Actually the pH-dependent reversible folding/unfolding mechanism has been nowadays used in technological applications like in the construction of nanocontainers. We investigate the unfolding mechanism in high temperature unfolding simulations and characterized it in terms of its eigenvectors. Furthermore we present the results of Molecular Dynamics simulations for the free energy landscape for different reaction coordinates which has been computed by a generalized version of the metadynamics approach. Our results indicate that at room temperature the planar hairpin structure is more stable than the totally stretched chain.

**15 min. break.**

BP 4.6 Mon 15:45 ZEU 260

**Development of an inter-nucleotide potential for DNA based on Density Functional theory** — •Maria Fyta[1,2], Greg Lakatos[1], Pierfrancesco Rosini[3], Amanda Peters[1], Simone Melchionna[1,4], and Efthimios Kaxiras[1] — [1]Department of Physics and School of Engineering and Applied Sciences, Harvard University, Cambridge, MA, USA — [2]Physics Department, Technical University of Munich, 85748 Garching, Germany — [3]Laboratory for Multiscale Modeling of Materials, EPFL, Lausanne, Switzerland — [4]Istituto Applicazioni Calcolo, CNR, Rome, Italy

The structural and dynamical properties of double stranded DNA (dsDNA) play a critical role in a range of fundamental biological and technological processes. These include DNA translocation through artificial or biological nanopores, the wrapping of DNA around histone proteins, and the use of DNA molecules as nanotethers in nanoscale devices. To understand the behavior of DNA in these contexts, it is desirable to have a computational model capable of treating oligomers with hundreds to thousands of base pairs, on time scales of microseconds or longer. Utilizing accurate density-functional electronic structure techniques, we are developing a coarse-grained molecular model of dsDNA capable of reproducing the molecule's structural and dynamical properties on these length and time scales. Validations of the model indicate that it reproduces a number of experimentally measured structural features of DNA, including the persistence length under physiologic conditions. The generated potential model will be capable to investigate the behavior of dsDNA in interesting biophysical processes.

BP 4.7 Mon 16:00 ZEU 260

**Type III restriction enzymes use 1D diffusion to communicate the relative orientation of their distant target sites** — •Friedrich W. Schwarz[1], Julia Tóth[2], Kara van Aelst[2], Mark D. Szczelkun[2], and Ralf Seidel[1] — [1]BIOTEC TU-Dresden — [2]University of Bristol, UK

Type III restriction enzymes sense the relative orientation of their distant target sites and cleave DNA only if at least two of them are situated in an inverted repeat. The communication process is strictly dependent on ATP hydrolysis catalyzed by their superfamily 2 helicase domains. Given the similarity to Type I restriction enzymes, which couple ATP hydrolysis to directed motion on DNA, unidirectional loop translocation has been the suggested communication mechanism for Type III enzymes. Based on magnetic tweezers single-molecule cleavage experiments and ATPase measurements we suggest an alternative inter-site communication mechanism using 1D diffusion along the DNA contour. In order to verify this hypothesis we directly visualized the motion of Q-dot labeled Type III restriction enzymes along DNA. For this we used a setup that combines magnetic tweezers with total in-

ternal reflection fluorescence microscopy. The enzymes undergo a fast diffusive motion along DNA, capable of scanning kb distances per second. We also found that the affinity of the enzymes to non-specific and specific DNA is regulated by the presence of ATP, suggesting that ATP hydrolysis acts as a trigger for diffusion. Thus Type III restriction enzymes are the first DNA-modifying enzymes which communicate target site orientations over long distances via 1D diffusion.

BP 4.8 Mon 16:15 ZEU 260

**Probing the elasticity of DNA on short length scales by modeling supercoiled DNA under tension** — •Robert Schöpflin[1], Hergen Brutzer[2], Oliver Müller[1], Ralf Seidel[2], and Gero Wedemann[1] — [1]University of Applied Sciences Stralsund, 18435 Stralsund, Germany — [2]Biotechnology Center Dresden, University of Technology Dresden, 01062 Dresden, Germany

The worm-like-chain (WLC) is the most commonly used theoretical framework for modeling the flexibility of DNA. However, recently alternative so-called sub-elastic chain (SEC) models [1] were proposed that predict for large deflections a higher flexibility than the usual harmonic model. So far, no unambiguous verification of these models has been obtained since probing the elasticity of DNA on short length scales remains challenging. Here, we address this question by modeling single-molecule supercoiling experiments of DNA under tension [2] using high-resolution Monte Carlo simulations. DNA supercoiling under tension is accompanied by an abrupt buckling at the transition from the stretched to the superhelical, i.e. plectonemic, state. This transition is due to the extreme bending of the DNA in the end loop of the plectoneme and serves therefore as a sensitive benchmark for model evaluations. While simulations that employ regular WLC bending energetics quantitatively reproduce the buckling transition, the buckling almost disappears for SEC models. Thus, DNA bending is best described by a harmonic model down to bending radii of 3 nm.

[1] Wiggins, P., et al. Nat Nanotechnol. 1(2):137-41 (2006)
[2] Brutzer, H., et al. Biophys J. 98(7):1267-76 (2010)

BP 4.9 Mon 16:30 ZEU 260

**Optical Tweezers Force Spectroscopy of a Single DNA-bound Protein during Nanopore Translocation** — •Andy Sischka[1], Andre Spiering[1], Sebastian Getfert[2], Peter Reimann[2], Janine König[3], Karl-Josef Dietz[3], and Dario Anselmetti[1] — [1]Experimental Biophysics and Applied Nanoscience, Bielefeld University, Germany — [2]Condensed Matter Theory, Bielefeld University , Germany — [3]Biochemistry and Plant Physiology, Bielefeld University, Germany

We investigated the translocation of single protein molecules (RecA, Peroxiredoxin and EcoRI) bound to dsDNA through a solid-state nanopore controlled by optical tweezers and an electric field (nanopore force spectroscopy). During threading, we found distinct asymmetric force signals depending on the protein charge, the DNA elasticity and the counter-ionic screening [1]. A theoretical model of an isolated charge on an elastic polyelectrolyte strand experiencing an anharmonic nanopore potential compares very well with the measured force curves and explains a linear voltage dependency and a small hysteresis during back and forth translocation cycles. Translocation dynamics reflects the stochastic nature of the thermally activated hopping between two adjacent states in the nanopore [2]. This opens new and fascinating applications for label-free localization and discrimination of DNA-binding ligands, where positional and structural binding phenomena can be investigated in real-time at the single molecule level.

[1] A. Sischka et al.: J. Phys - Condens. Matt. 22: 454121 (2010)
[2] A. Spiering et al.: submitted (2010)

BP 4.10 Mon 16:45 ZEU 260

**The interplay of mutations and electronic properties in disease-associated genes** — Chi-Tin Shih[1,2], Stephen A Wells[3], Ching-Lin Hsu[4], Yun-Yin Cheng[1], and •Rudolf A Römer[3] — [1]Department of Physics, Tunghai University, 40704 Taichung, Taiwan — [2]The National Center for Theoretical Sciences, 30013 Hsinchu, Taiwan — [3]Dept. of Physics and Ctr for Scientific Computing,University of Warwick, Gibbet Hill Road, Coventry, CV4 7AL, UK — [4]Department of Physics, Chung-Yuan Christian University, Chung-Li, Taiwan

The electronic properties of DNA molecules are believed to play a crucial role in many phenomena taking place in living organisms, for example the detection of DNA lesions by base excision repair (BER) glycosylases such as Endonuclease III and MutY and the regulation of tumor-suppressor genes such as p53 by detection of oxidative damage. However, the reproducible measurement and modelling of charge migration through DNA molecules at the nanometer scale, in vitro or in vivo, remains a challenging and controversial subject even after more than a decade of intense efforts. Here we show, by analysing 162 disease-associated genes from a variety of medical databases with a total of almost 20000 known pathogenic mutations, a significant difference in the electronic properties of the population of pathogenic mutations compared to the set of all possible mutations. Comparison of the results for different models of charge transport suggests that it is the electronic properties of the coding strand, rather than the conductance of the double helix, that is significant.