AKB 18 Ion Channels and Nanopores

Time: Wednesday 15:30–16:45 Room: ZEU 255

Invited Talk AKB 18.1 Wed 15:30 ZEU 255

Synthetic Analogues of Biological Voltage-Gated Channels, Fabrication of Ion-Current Rectifiers and Protein Sensors — •Zuzanna Siwy — Department of Physics and Astronomy, University of California, Irvine, USA

We have fabricated a single asymmetric nanopore that mimics behavior of biological voltage-gated channels. The single pores have been prepared by the track-etching technique. The pores are conical in shape with diameter of the small opening down to several nm and the big opening in the micrometer range. We have designed two nanotube systems, which exhibit ion current rectification through two distinct mechanisms (i) through asymmetric potential energy profile for an ion inside the pore, and (ii) electro-mechanical gate placed at the entrance of the conical pore.

We have also designed single nanopore system, which produces voltage-dependent ion current fluctuations with the kinetics of opening and closing similar to voltage-gated biochannels.

I will also discuss application of synthetic voltage-gated nanopores as platforms in biosensing. The internal surfaces of the nanopores have been modified with a specific biochemical molecular-recognition agent (the *capture* agent, e.g., an antibody) which interacts specifically with a given biomolecule (the analyte) brought into contact with the nanotube. The binding interaction between the nanotube-bound capture agent and the solution-phase analyte is transduced as a change in the ion current that flows through the nanotube. We have demonstrated operation of the sensor for detection of ricin and immunoglobins.

AKB 18.2 Wed 16:00 ZEU 255

Gating charge effects in excitable membranes — •Gerhard Schmid, Igor Goychuk, and Peter Hänggi — Institut für Physik, Universität Augsburg

Voltage dependent ion channels mainly determine the electric properties of axonal cell membranes. The ion channels thereby do not only allow the passage of ions trough the cell membrane but they also contribute to the additional charging of the cell membrane resulting in the so-called capacitance loading. The switching of the channel gates between an open and a closed configuration is always connected with movement of gating charge within the cell membrane. At the beginning of an action potential the gating current is opposite to the direction of the ion current through the membrane. Therefore the excitability is reduced due to capacitance loading. Our stochastic Hodgkin-Huxley modelling takes into account both, channel noise – the fluctuations of the number of open ion channels [1] – and the capacitance fluctuations due to gating charge. We investigate the spiking dynamics of small membrane patches and analyze the statistics of the spontaneous spiking. In doing so, we find that such gating charge effects yield a drastic reduction of the spontaneous spiking rate. This work is supported by DFG (SFB 486).

[1] G. Schmid, I. Goychuk, P. Hänggi, Europhys. Lett. **56**, 22-28 (2001).

AKB 18.3 Wed 16:15 ZEU 255

Direct force measurements on DNA in a solid-state nanopore — •U. F. Keyser, B. N. Koeleman, D. Krapf, R. M. M. Smeets, S. G. Lemay, N. H. Dekker, and C. Dekker — Kavli Institute of Nanoscience, Delft University of Technology, The Netherlands

Amongst the variety of roles for nanopores in biology, an important one is enabling polymer transport, for example in gene transfer between bacteria and transport of RNA through the nuclear membrane. Recently, this has inspired the use of protein and solid-state nanopores as single-molecule sensors for the detection and structural analysis of DNA and RNA by voltage-driven translocation. The magnitude of the force involved is of fundamental importance in understanding and exploiting this translocation mechanism, yet so far has remained unknown. Here, we demonstrate the first measurements of the force on a single DNA molecule in a solid-state nanopore by combining optical tweezers with ionic current detection. The opposing force exerted by the optical tweezers can be used to slow down and even arrest the translocation of the DNA molecules. We obtain a value of 0.24*0.02 pN/mV for the force on a single DNA molecule, independent of salt concentration. Our data allow the first direct quantitative determination of its effective charge of 0.53*0.05 electrons per base pair, corresponding to a 73% reduction of the bare DNA charge. Our novel single-molecule technique further is a major step forward in biotechnology (towards rapid DNA sequencing) and biophysics (study of unfolding of RNA or DNA-protein binding).

AKB 18.4 Wed 16:30 ZEU 255

Antibiotic translocation through OmpF — •Tivadar Mach[1], Karin Türk[1], Luminita Damian[1], Sergei M Bezrukov[2], and Mathias Winterhalter[1] — [1]International University Bremen, Germany — [2]National Institutes of Health, Bethesda, MD, USA

The first step for antibiotics to reach their target in gram-negative bacteria is crossing the outer membrane. Several studies have concluded that general diffusion porin OmpF plays an important role in the uptake of some antibiotics, and that this uptake can often be considered the limiting step in their functionality. We use the analysis of the ion current through a single trimeric OmpF porin reconstituted into a planar lipid bilayer to study binding and translocation of fluoroquinolone antibiotics of different structure and hydrophobicity. Because the size of the antibiotic molecules is close to the size of the constriction zone in the OmpF pore, longer residence times of the fluoroquinolones in the channel cause random transient blockages in the ion current. Analysing these fluctuations we calculate chemical binding constants, affinity, and transfer probability. Our findings complete and corroborate earlier indications and indirect measurements of pathways of entry, providing a test-case for a direct, controlled measurement method also to be expanded to other antibiotic structures.