and trends between the various parameters that are important to ion-channel experiments, and compare the results to our theory.

2825-Pos Board B517

Screening for Influx through Membrane Proteins on the Single-Molecule Level using an Automated and Parallel Lipid Bilayer Platform

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Planar lipid bilayers can be automatically formed by remotely actuated painting in parallel on Micro-Electrode-Cavity-Arrays (MECA)-Chips. Voltage-clamp recording of ionic current through bacterial channels in bilayers is a powerful technique, yielding information on both the physicochemical characteristics and the physiological role of membrane proteins. Antibiotics penetrate bacterial membranes through general diffusion protein channels, like OmpF in E. coli. The MECA chip approach supports the analysis of translocation events in high resolution by detection of the transient block of ionic currents when a single molecule resides in the pore.

Here, reconstituting the porins CarO from A. Baumannii, OmpC and OmpF from E. coli and orthologs in K. pneumoniae/E. cloacae, we resolved the transport of antibiotics and thereby study kinetics of antibiotics permeation through porins. This information can be useful in optimizing the design of new antibacterial drugs effective against resistant bacteria.

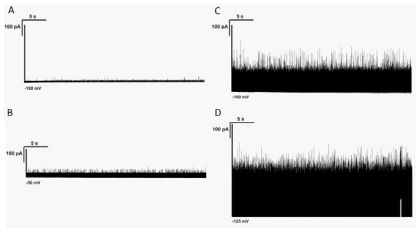

Figure: single OmpF in bilayers formed using the MECA platform (in 1 M KCl at RT=25°C). A. in absence of antibiotics at −100 mV (control), B, C,D. Recording at −50, −100 and −125 mV in presence of 5 mM of Norfloxacin.

2826-Pos Board B518

Molecular Dynamics Simulation of Fast Water Transport though Aquaporin-Mimic Nanpores

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The aquaporin water channel allows efficiently fast water transport while rejecting ions and protons. We perform molecular dynamics simulations with flexible hourglass-shaped pores by mimicking aquaporin water channels. The simulation model includes a uniform applied external force field to mimic osmotic gradient environment and an added harmonic constraint to achieve nanopore flexibility. We estimated the osmotic permeability from simulation and it is consistent with experimental results to confirm the validity of our model. We performed non-equilibrium molecular dynamics simulation with Nosé-Hoover thermostat, the SPC/E water model, and carbon pore atoms. The occupancy (number of water molecules inside the pore) is maintained as a fixed value as long as the pore geometry is the same. The simplified model also successfully captures the important physics of real aquaporin, e.g. a single-file arrangement of water molecules, the reorientation of water dipoles. We found that the model pore has the capacity to house more water molecules. We also found that water flow is up to 5 times than predicted by continuum hydrodynamic theory. This finding suggests that deforming straight nanopores into the hourglass shape (if possible) enhances the ability of water transport in thin membranes. The future work includes the simulations with a longer hourglass-shaped nanopore (maybe up to 1 μm) and changing the chemical characteristics (e.g. surface charge, hydrophobicity) of a membrane surface.

2827-Pos Board B519

1/F Fluctuations of Amino Acids Generate Non-Poisson Water Transportation in AQP1

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Aquaporins (AQPs), which transport water molecules across cell membranes, are involved in many physiological processes. Most AQPs are exclusively permeated by water molecules, rejecting charged molecules and ions at aromatic/arginine selectivity filter (ar/R region). However, the effects of the conformational fluctuations of amino acids on water transportation remain unclear. Using all-atom molecular dynamics simulations, we analyze water transportation and fluctuations of amino acids within AQP1. The amino acids exhibit 1/f fluctuations, indicating possession of long-term memory. Moreover, we find that water molecules crossing the ar/R region obey a non-Poisson process. To investigate the effect of 1/f fluctuations on water transportation, we perform MD simulations of restrained AQP1 molecules and simple Langevin stochastic simulations. 1/f fluctuations of amino acids contribute to water transportation in AQP1. These findings appreciably enhance our understanding of AQPs and suggest possibilities for developing biomimetic nanopores.

2828-Pos Board B520

Thermodynamics of Water Entry in Aquaporins

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Permeation of water across cell membranes is facilitated by highly efficient, selective water channels called aquaporins. While a large number of experimental and computational studies over the last decade have provided great details about the mechanisms of permeation and selectivity in these proteins, some of the fundamental aspects of the mechanism are still unknown. Here, we report on a computational study employing various simulation and analysis techniques to characterize the thermodynamics of water molecules inside the aquaporins in its most detailed manner. Extensive molecular dynamics simulations on various aquaporins have been performed and the spatial translational and rotational components of diffusion and entropy were computed using the 2 phase thermodynamic method (2PT). For the first time, we find that the spontaneous filling of AQPs by water is primarily the result of an entropic gain. Our results also indicate a strong interconversion of the entropy components as water molecules enter the channel; while water molecules exhibit the highest rotational motion in the extra/intra cellular mouths of the pore, their translation is slow compared to the bulk value. The frequencies in the power spectra of translational and rotational motions overlap, indicating a strong coupling of these motions inside the pore. A shooting mechanism with near diffusive behavior is observed in the extracellular mouth of the pore, which can explain the fast conduction of water in aquaporins.

2829-Pos Board B521

Ion Transport Modelling for Quality Assessment of Transmembrane Protein Structures

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Currently only 2% of transmembrane protein structures are known, while transmembrane proteins correspond to 25% of genes. One of the main goals of bioinformatics is to predict the structure of a protein based on its sequence. An important step in the modelling process is quality assessment of the models. In this work we propose a function based flow method which can be compared to the experimental channel characteristics. Two basic methods for modelling ionic channel function are used: the 3D Poisson-Nerst-Planck model (3D PNP), which is a fast continuum method and Brownian Dynamics (BD), which is a discrete-continuum algorithm.

We show that the 3D PNP solver is a sufficient tool for the quality assessment of the potassium channel structural models, which rejects 97% of misfolded structures within 2-3 CPU-minutes each structure. Deficiency of the PNP-based approach is rejection of 60% of proteins with low RMSD and good electrostatic properties. To improve the sensitivity, we develop a novel BD tool for ion transport simulation. In this study, we evaluate two methods for calculating the pore accessibility and two representations of ion sizes (real radius and single voxel). In both cases the size of ions is involved in the diffusion coefficient calculation. The tool uses the electrostatic potential distribution obtained from the 3D PNP simulations. We tested the results of our approach on the alpha-hemolysin (7ahl) and the potassium (3fb8) channels.