Rather than focusing on the structural details of a specific natural protein, we are designing general protein structural scaffolds (“maquettes”) to accommodate a variety of functions. Here we will present transmembrane electron transfer via AP6, an amphiphilic tetra-helical maquette that binds up to 6 helices. We demonstrate that AP6 self-assembles with phospholipids into vesicles. Our stop flow experiments confirm that the AP6 maquette significantly increases the electron transfer rates between oxidizing interior and an external redox mediator dye, as shown below.

**711-Pos  Board B511**

Expression and Characterization of Cytochrome C6 from Chlamydomonas Reinhardtii using a Designer Gene

Nicole L. Vanderbush, Brian St. Clair, Marylyn Davis, Dan Davis.

Cytochrome C6 is a luminal redox carrier in oxygenic photosynthesis. We have constructed a synthetic gene, expressed, purified, and co-oxidized an initial characterization for the cytochrome c6 from Chlamydomonas reinhardtii. The synthetic gene was constructed by the removal introns and the substitution of synthetic gene was constructed by the removal introns and the substitution of a UV-Vis spectrophotometer and include a reduced peak at 553nm, a peak at 523nm, and a Soret band at 417nm. The midpoint potential at pH 7 was determined by redox titrations and found to be 365 mV. Differential scanning calorimeter experiments also reveal that the folding of the protein only slightly favors the product state, the solvent and counterions not be resolved in the 3.35 Å resolution X-ray structure.

The glycine betaine/sodium symporter BetP responds to changes in external osmolality by regulation of its transport activity. A recent X-ray structure of the three protomers have different conformations. These results suggest that the isolated monomer BetP trimers in order to identify interface residues crucial for maintaining the trimeric state. Mutations across the width of nanowires. We will present electron transport measurements on intact biofilms and the contribution of nanowires to their overall conductivity.

**Membrane Transport**

**714-Pos  Board B514**

Computer Simulation of TolC Ground State Dynamics and Spontaneous Binding of the AcrB Docking Domain

Martin Raunest, Nadine Fischer, Christian Kandt.

In Escherichia coli the AcrAB-TolC efflux pump expels a broad range of drugs and other molecules. While AcrB is the engine in this system, the outer membrane protein TolC functions as an efflux duct interacting with numerous inner membrane translocases. TolC occurs in at least two states, one that is impermeable for drugs and one where drug passage is possible. We performed a series of five independent, unbiased 150ns molecular dynamics (MD) simulations of wildtype TolC 1EK9 embedded in a phospholipid/water environment at 0.15M NaCl concentration. One of these runs was extended to 300ns in three independent copies. Whereas TolC remains closed between a 1st bottleneck region outlined by Asp-374 &371, we observe opening and closing motions in a 2nd bottleneck region near Gly-365. While previous studies reported a frequent binding of potassium ions stabilizing a closed TolC conformation in the bottleneck II region, we observe a frequent passage of sodium ions. However, in one simulation a consecutive binding of two sodium ions occurs between Gly-365 and Asp-374 leading to a closed TolC conformation at the AcrB interface, which was stable for more than 175ns. To gain insight into TolC-AcrB interaction, we performed five independent unbiased 150ns MD simulations of TolC and the AcrB docking domain (AcrB_dd). Initially placed 1 nm away from TolC and identically oriented as in the AcrAB-TolC Symmons docking structure, AcrB_dd spontaneously docks to TolC in one run. Extending this simulation to one micro second, we find an TolC-AcrB_dd docking interface characterized by a larger contact area and a slight asymmetry not present in the Symmons model. At the same time TolC opens in the bottleneck II region. All simulations were performed using GROMACS 4.0.3 and the G53a6-GROMOS96 force field.

**715-Pos  Board B515**

Computational and Experimental Studies of Substrate Binding, Conformational Change and Importance of the Trimeric State in the Glycine Betaine Transporter BetP

Kamil Khabizov, Camilo Perez, Ching-Ju Tsai, Christine Ziegler, Lucy R. Forrest.

The glycine betaine/sodium symporter BetP responds to changes in external osmolality by regulation of its transport activity. A recent X-ray structure of BetP, the structures of the alternate states (e.g., open to the outside of the cell), molecular mechanisms of substrate and Na+ binding and transport, as well as the functional implications of the trimeric state remain poorly understood. To address these questions, we carried out computational studies using a range of techniques to derive hypotheses that were then tested experimentally. First, to identify structural features of the alternate states, we developed a procedure for feasible fitting of the X-ray structure of BetP into a lower-resolution cryo-EM map of BetP in a more native lipid environment, in which the three protomers have different conformations. These results suggest that: (i) the protomers adopt distinct conformational states relevant to the transport cycle; and (ii) there is conformational coupling between the protomers. Second, we performed all-atom molecular dynamics simulations and in silico alanine scanning of BetP trimers in order to identify interface residues crucial for maintaining the trimeric state. Mutations of these residues to alanine were introduced experimentally revealing that the isolated monomers are functional, and that the trimeric state is important for the regulation and higher activity of the protein. Finally, using molecular modeling and biochemical experiments we identified two Na+ binding sites in BetP that could not be resolved in the 3.35 Å resolution X-ray structure.

**716-Pos  Board B516**

Mathematical Model of the Regulatory Cell Volume Decrease

Aleksandr V. Il'yaskin, Galina Batyrina, Evgeniy I. Solonov, Aleksandr Ershov, Dmitriy Medvedev, Denis Karpov.

Renal collecting cells perform vasopressin regulated water reabsorption and form the composition of tubular fluid. The osmotic pressure of the extracellular fluid varies significantly. To maintain viability in hypotonic