potential of mean force (PMF), was calculated along the path. The electrostatic coupling between the excess proton and chloride ion was also explored. These studies therefore provide a more detailed picture of the proton transport process in the ClC-ecl antiporter.

1713-Pos

Ion Selectivity in the Aspartate Transporter Glt_{PH}

Michael Thomas, Dylan Jayatilaka, Ben Corry.
University of Western Australia, Perth, Australia.

The aspartate transporter Glt_{PH} is an integral membrane protein that catalyses the movement of aspartate across lipid bilayers. Glt_{PH} utilises established ion gradients, transporting two sodium ions with each aspartate molecule. Previous studies have shown that the ion binding sites demonstrate selectivity for Na^+ over both Li^+ and K^+ ($Na^+ > Li^+ > K^+$) [1]. The sodium binding motif is similar to that of another sodium dependent leucine transporter, LeuT. Computational studies have attributed different mechanisms to ion selectivity in each of the two sodium binding sites in LeuT [2]. Selectivity in the first site results from the binding of the negatively charged carboxylate group of the substrate resulting in strong electrostatic interactions while selectivity in the second site is enforced by an almost rigid cavity of coordinating ligands held in place hydrogen bonding networks.

Using various computational techniques, we describe the thermodynamic contributions to the free energy of binding that give rise to the experimentally observed selectivity sequence $Na^+ > Li^+ > K^+$ in Glt_{PH} and compare and contrast them to those in LeuT.

[1] Boudker, O. et al. Nature 2007, 445, 387-393
[2] Noskov, S.; Roux, B. J. Mol. Biol. 2008, 377, 804-818

1714-Pos

Microscopic Mechanism of Ion Selectivity in the Nak Pump

Haibo Yu[1], Pablo Artigas[2], Benoit Roux[1].
[1]Department of Biochemistry and Molecular Biology, University of Chicago, Chicago, IL, USA, [2]Department of Cell Physiology and Molecular Biophysics, Texas Tech University Health Science Center, Lubbock, TX, USA.

The sodium/potassium pump establishes the Na+ and K+ concentration gradients across the plasma membrane of animal cells and therefore plays an essential role in maintaining cell volume and secondary active transport of other solutes. The crystal structures of the Na+/K+ pump provide atomic insight into the binding of K+ ions and conformational transitions during the functional cycle. However, important details about the ion-selectivity remain to be addressed. In particular, 2 out of the 3 binding sites are shared between Na+ and K+ and it is not clear how this pump selects K+ over Na+ when in the outwardly facing conformation (E2P) or Na+ over K+ when in the inwardly facing conformation (E1). We have undertaken free energy calculations to understand the physical principles that govern the ion selectivity in Na+/K+ pump and dissected various factors that may contribute to the selectivity. We found that the pump elegantly modulates the electrostatic environment of the binding sites to achieve the corresponding selectivity. Our results are consistent with available experimental data and provide new hypothesis to test experimentally. [Supported by NIH grant GM062342].

1715-Pos

The Role of Architectural and Structural Forces in Ion Selectivity

Haibo Yu[1], Sergei Y. Noskov[2], **Benoit Roux**[1].
[1]University of Chicago, Chicago, IL, USA, [2]University of Calgary, Calgary, AB, Canada.

A novel theoretical framework is presented to clarify the role of architectural and structural forces in ion selectivity by expressing the relative free energy of bound ions in terms of a reduced local system coupled to a potential of mean force (PMF) representing the influence of the surrounding environment. The PMF is separated into two contributions. The first includes all the harsh forces keeping the ion and the coordinating ligands confined to a small microscopic region, but do not prevent the ligands from adapting to ions of different radii. The second regroups all the remaining forces that serve to dictate a precise geometry of the coordinating ligands best adapted to a given ion. In the limit where the precise geometric forces are dominant, the binding site is almost rigid and ion selectivity is controlled by the ion-ligand interactions according to the classic ''snug-fit'' mechanism of host-guest chemistry. In the limit where the precise geometric forces are negligible, the ion and ligands behave as a self-organized ''confined droplet'' that is free to fluctuate and adapt to a smaller ion. But selectivity can also occur under such conditions. In the small and crowded volume, ion selectivity is determined by the ion-ligand and ligand-ligand interactions and is controlled by the number and the chemical type of ion-coordinating ligands. The theoretical framework is used to analyze K+ binding sites in the KcsA channel and Na+ binding sites in the LeuT transporter.

1716-Pos

Mechanisms of Ion Permeation through Gramicidin a Channels

Yuhui Li[1], Olaf S. Andersen[2], Benoit Roux[1].
[1]Department of Biochemistry and Molecular Biology, The University of Chicago, Chicago, IL, USA, [2]Department of Physiology and Biophysics, Weill Medical College of Cornell University, New York, NY, USA.

Gramicidin A (gA) channels make an ideal system to test all-atom molecular dynamics (MD) of membrane proteins and mechanisms of ion permeation. In addition to being the most studied membrane ''protein'', gA channels are tiny, allowing for long MD runs and calculations of potential of mean force (PMF) in tractable time. The binding sites at either end of the gA channel can both hold a single cation. At low concentration, permeation occurs as a series of independent events in which one cation at a time moves across the pore. Ion permeation usually is described using the ion position z in the direction of the pore axis as a ''reaction coordinate''. But it is not known whether z is a good reaction coordinate to describe the process. A powerful tool to characterize the mechanism of ion permeation in the gA channel is the ''committor'' probability: the fraction of trajectories initiated from a given position that first commit to the left or right binding site of the channel. We evaluate the committor probability distribution function to identify the physical reaction coordinates of a K+ in gA using extensive MD calculations. At high concentration, permeation is dominated by 2-ion processes where cations are bound at either ends of the small pore. To understand the impact of double ion occupancy on the mechanism of ion permeation, we calculate the 2-ion PMF. The results show that if the first ion resides in the inner binding sites at one end of the channel, then the outer and inner binding sites for the second ion at the other end of the channel become shallow. The energetics of double occupancy is explained by considering the dipole moment fluctuation of the single-file water molecules inside channel. [Supported by NIH grant GM070971].

1717-Pos

Thermodynamically Dominant Hydration Structures of Ions and their Role in Ion-Specificity

Safir Merchant, **Dilip Asthagiri**.
Johns Hopkins University, Baltimore, MD, USA.

To understand the basis of ion-specific effects in biology, it is necessary to first understand the hydration structure and thermodynamics of ions. Based on a multi-state organization of the potential distribution theorem, we present new insights on the role of ion-water interactions and water density fluctuations at the size-scale of the ion in determining the ion-hydration structure and thermodynamics. We find that the hydration free energy of the ion depends on three quantities: 1) the hydration free energy of the ion in a specified n-coordinate state, where in the n-coordinate state n water molecules are present within the coordination volume of the ion; 2) the probability, xn, of observing that n-coordinate state around the ion; and 3) the probability, pn, of observing n water molecules in the coordination volume in the absence of the ion. Based on this development we find that only a small subset of water molecules in the first hydration shell of the ion sense the chemical type of the ion. Further, these core-water molecules tend to attenuate the interaction of the ion with the rest of the medium, and thus the higher coordination states of the ion more sensitively reflect density fluctuations of the solvent medium at the size scale of the observation volume. The relevance of this development in understanding ion-pairing and the selective binding of ions to biological molecules is discussed.

1718-Pos

On the Domain of Applicability of Currently used Force Fields for the Calculation of the Activity of Alkali Ions at Physiological Ionic Strength

Chao Zhang[1], Simone Raugei[1], Bob Eisenberg[2], Paolo Carloni[1].
[1]SISSA, Trieste, Italy, [2]Rush Medical Center, Chicago, IL, USA.

Alkali ions are present in virtually all biological processes. Their energetic properties have been so far predicted mostly by MD or MC calculations based on effective potentials derived for infinite diluted conditions (i.e. a single ion surrounding solely by water molecules) [1]. However, in physiological conditions, the concentration of K+ is sub-molar in the cytoplasm [2], and it may be by one, or even two, orders of magnitude larger near globular proteins or nucleic acids and in the active sites of of enzymes or channels [3-5]. The presence of a large ionic strength I is likely to limit the accuracy of the currently used potentials.

Here we will discuss recent calculations of the activity coefficients for K+,Na+ ions at increasing I. Such coefficients have been obtained by calculating the excess chemical potentials from thermodynamics integration [6], with several commonly used biomolecular force fields. Preliminary results show that classical force fields generally overestimate the activity coefficients of ions.

[1] M. Patra and M. Karttunen. J. Comput. Chem., 25:678-689, 2004.