Molecular Dynamics Study on the Opening Behavior of Bacterial Mechano-sensitive Channel MscL Effected by Membrane Thickness

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One of mechanosensitive channels Mscl is homopentamer with two transmembrane inner (TM1) and outer (TM2) helices and activates by sensing membrane tension. The major issue of Mscl is to solve its gating mechanism. Previous studies revealed that Mscl embedded in a thinner lipid bilayer opens more easily. However, it remains unclear why the channel opening depends on the bilayer thickness. Thus we performed MD simulations of Mscl embedded in three types of the bilayer to explain the dependence of the opening behavior in atomic detail. As a result, Mscl in a thinner membrane actually expanded widely. Also it was found that a tilt angle of the tranemembrane helices increased as the thickness decreased, leading to smaller interaction energy between Mscl and the membrane.

Mechanosensitive Channel Mscl Using Molecular Dynamics Simulations

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One of mechanosensitive channels, Mscl, is homopentamer of a subunit with transmembrane inner and outer helices. The major issue of Mscl is to understand the gating mechanism driven by membrane tension. To address this question, MD simulations have been performed; however, it remains unclear the relationship between tension sensing and the gate opening. Thus, we performed opening simulations of the channel and analyzed thermal fluctuations using principal component analysis and get insight into the coupling between a mechanosensor and the gate. We modeled wild type, F78N and G22N mutant Mscl, and performed simulations for 40 ns to sample 5000 coordinate data sets. As a result, it was found that a fluctuation coupling of between 22nd and 78th amino acids is important.

Mechanosensor and gate is tightly coupled in the opening process of the bacterial mechanosensitive channel Mscl.

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The bacterial mechanosensitive channel Mscl is known to gate by membrane tension and we previously found that F78 acts as a tension sensor. In this study we performed MD simulations of several Mscl mutants to get insights into the relationship between the tension sensor F78 and the gate. The GOF mutant G22N is easier to open, while the LOF mutant F78N cannot be opened upon strong membrane stretch. To test whether the behavior of G22N is independent of the tension sensing at F78, we performed simulations of the double mutant G22N/F78N and found that G22N/F78N Mscl did not open the gate, suggesting that the tension sensor and the gate of Mscl is tightly connected and that the interaction between the tension sensor and lipids is essentially important for the Mscl opening.

H²Ca²⁺ Exchange Transporter in Outer Membrane of E. coli

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The multidrug efflux transporter AcrB and its homologs are important in the multidrug resistance of Gram-negative pathogens. ABI-PP is AcrB and MexB-specific inhibitor that does not inhibit MexY; MexB and Y are principal multidrug exporters in Pseudomonas aeruginosa. We have determined the first inhibitor-bound structures of AcrB and MexB. ABI-PP tightly binds to a narrow pit composed of a phenylalanine cluster located in the binding site and sterically hinders the functional rotation. We found that the difference of affinity for the inhibitor between AcrB/MexB and MexY is the volume of the residues of the end of the pit. The structure of the hydrophobic trap described in this study will contribute to the development of universal inhibitors of MexB and MexY.

Structural Basis for the Counter-Transport Mechanism of a H⁺/Ca²⁺ Exchanger

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Ca²⁺/cation antiporators catalyze the exchange of Ca²⁺ with various cations across biological membranes to regulate cytosolic calcium levels. Here, we report the crystal structure of a H⁺/Ca²⁺ exchanger from Archaeoglobus fulgidus (CAX_Af) in the two representatives of the inward-facing conformation at 2.3 Å resolution. The structures suggested Ca²⁺ or H⁺ binds to the cation-binding site mutually exclusively. Structural comparison of CAX_Af with previously a reported CaCA protein revealed that the first and sixth transmembrane helices alternate create hydrophilic cavities on the intra- and extracellular sides. The structures and functional analyses provide insight into the mechanism of how the inward- to outward-facing state transition is triggered by the Ca²⁺ and H⁺ binding.

Two alternative conformations of a voltage-gated sodium channel

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We present two conformations of a voltage-gated sodium channel (Nav) from C. thermarum reconstituted into lipid bilayers in one crystal at 9Å resolution based on electron crystallography. Despite a voltage sensor arrangement identical to that in the activated form, we observed two distinct pore domain structures of a prominent form with a relatively open inner gate and a closed inner gate conformation similar to the first prokaryotic Nav structure. Our analyses together with mutational and electrophysiological experiments indicated that widening of the inner gate was dependent on interactions among the S4-S5 linker, the N-terminal part of S5 and its adjoining part in S6, and on inter-helical repulsion by a negatively charged C-terminal region subsequent to S6.