a genetically altered strain of *E. coli* with a non-functioning AcrAB-TolC pump. Graphical representations of data revealed four possible distinctive patterns or ''stages'' of influx of the EtBr compound into the live cells. Data also suggests the Langmuir model of adsorption as being a potential mathematical model for the first diffusion stage. Development of a working model that allows for accurate determination of the kinetic behavior of the influx of EtBr into the cell allows for a basis of comparison in strains of *E. coli* with fully functioning efflux pumps. This would allow for the quantification of an efficiency value can then be used to help determine which compounds are most effectively removed by the AcrAB-TolC pump.

1135-Pos

Reconciling Membrane Protein Simulations with Experimental Spectroscopic Data

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Scarcity of membrane protein biophysical characterization necessitates that we salvage all information available from laboratory experiments and computational simulations. Spectroscopy experiments such as double electron-electron resonance (DEER), through residue pair probes, provide distance distributions that indicate conformational heterogeneity in membrane proteins. Atomistic molecular dynamics (MD) simulations, can structurally determine inward-open, occluded, and outward-open conformations of transporter membrane proteins, among other partially open/closed states of the protein. Yet, we find that there is no direct consensus between distributional data from DEER experiments and MD simulations, which has challenged validation of structures obtained from long-timescale simulations. We hypothesized four potential sources of mismatch for observed discrepancies: (1) Increased rotamer dynamics from mutational cysteines and MTSSL probes. (2) Overall protein dynamics affected by mutations and probe additions. (3) Conflicting experiment and MD simulation protein environments because DEER spectroscopy experiments are performed with proteins embedded in either, detergent micelles or lipid nanodiscs. (4) Effect of lipid composition, solution and buffer conditions. These factors have not been systematically investigated for their contribution to mismatch between experiments and simulations. Current coping strategies for comparisons rely on heuristics, such as mapping nearest matching peaks between two ensembles or biased simulations, such as restrained MD. In this work, we perform atomistic MD simulations of transporter proteins, LeuT and PepTSo, from two major membrane protein families. Both proteins have been previously characterized via DEER experiments. Our simulations include various protein-membrane complexes - membranes with heterogeneous and homogeneous bilayers, micelle, and nanodisc environments; reveal differences in flexibility of the overall protein as well as specific sites chosen for probing. Our results will accelerate developing potential mitigation strategies to improve simulation observables match with spectroscopy experiments. Our insights will also be crucial in designing experimental micelle compositions similar to protein's native environment.

1136-Pos

Lipid Membrane Deformation Induced by Transmembrane Peptides

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The plasma membrane has an essential role in life activities of living cells. For example, the lipid membrane enables to enclose molecules in inner compartment to isolate from outer environment. Inner/outer molecules are sometimes exchanged by dynamic membrane deformation as endo- or exocytosis. Extensive proteins including membrane proteins complicatedly interact in the deformation event. Matrix-2 (M2) protein in influenza A virus mediates a part of infection pathway by budding and scission. It was reported that the partial sequence of M2 protein has the deformability to liposome which contains model cell-membrane. Besides, a few studies reported that melittin, one of antimicrobial peptides (AMPs), also induced liposome deformation, but the detailed mechanism of the deformation-induced peptides has been still unclear. In this study, as the first step to elucidate the mechanism, we focus on short membrane binding peptide, AMPs, and cell-penetrating peptides (CPPs), and evaluate their deformability. The AMPs are known as pore-formation in plasma membrane, and CPPs translates across the lipid membrane. In our experiments, we prepared liposome, added peptides to liposome solution, observed them by microscopy, and then performed the 2D shape-deforming analysis. Also, we developed planner lipid membrane and measured temporal membrane capacitance (Cm). In this paper, we will discuss on (a) these peptides have deformability (b) the deformability difference among each peptide. We believe that our methods will be a useful tool to elucidate the peptide deforming mechanism.

1137-Pos

Mechanisms of Negative Membrane Curvature Sensing and Generation

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Certain proteins have the propensity to bind to negatively curved membranes and generate negative membrane curvature. The mechanism of action of these proteins is much less studied and understood that that of positive curvature generators. In this work, we use implicit membrane modeling to explore the mechanism of two important negative curvature sensing and generating proteins: the main ESCRT III subunit Snf7 and the IRSp53 I-BAR domain. We find that Snf7 monomers alone can sense negative curvature and that curvature sensitivity increases for dimers and trimers. We have observed spontaneous bending of an Snf7 oligomer into a circular structure with 42 monomers and a radius of $\sim$20 nm. Snf7 oligomers bind with the same interface to both flat and curved membranes. Simulations of IRSp53 I-BAR domain on cylindrical and spherical membranes show that the I-BAR dimer is only weakly curvature sensing and prefers to orient parallel to the tube axis for tubes less than $\sim$ 40 nm. I-BAR dimers tend to make higher oligomers by interacting laterally in a staggered fashion. Such higher oligomers tend to have a bent shape and fit better on a negatively curved membrane. These studies shed some light onto the elusive mechanisms of negative curvature sensing and generation.

1138-Pos

Red Blood Cell Curvature is Controlled by the Non-uniform Distribution of Myosin-Mediated Forces and Membrane Tension

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The biconcave disk shape of the mammalian red blood cell (RBC) is unique to the RBC and is vital for its circulatory function. Recent experiments have demonstrated that the biconcave shape of the RBC relies not only on the physical properties of the membrane but also depends on the molecular constituents of the membrane skeleton, including the contractile activity of nonmuscle myosin IIA (NMIIA) motor protein. Here, we use the classical Helfrich model for the RBC membrane and incorporate heterogeneous force distributions along the membrane to mimic the contractile activity of NMIIA. By considering this new energy term compared to the classical Helfrich model, we find that the biconcave shape of the RBC depends on the ratio of forces per unit volume in the dimple and rim regions of the RBC. Experimental measurements of NMIIA densities at the dimple and rim validate our prediction that (a) membrane forces must be non-uniform along the RBC membrane and (b) the force density must be larger in the dimple region than the rim region to produce the observed membrane curvatures. Furthermore, we find that the tension of the RBC membrane plays an important role in regulating this force-shape landscape. Our findings of heterogeneous force distributions on the plasma membrane for RBC shape maintenance can have implications for shape maintenance of many cell types.

1139-Pos

Visualizing Opa1-Mediated Changes to Inner Mitochondrial Membrane Morphology

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Mitochondrial membrane morphology is directly related to organelle function. Previous studies show that the shape of the inner mitochondrial membrane (IMM) is directly implicated in regulating the release of cytochrome C to initiate apoptosis. However, the mechanism of this release is unclear. We applied *in situ* cryo-electron tomography to study the shape of the IMM under different protein states. We studied the effects of varying expression levels of the inner membrane fusogen Opa1 on membrane structure. Using focused-ion beam sectioning of Opa1-knockout, wild type, and overexpression eukaryotic cells, and subsequent transmission electron cryomicroscopy (cryo-EM), we observe mitochondria from these cell lines in their cellular environment. Tomographic reconstructions of these sections capture morphological changes resulting from altered protein expression. Our results show that *in situ* electron cryo-tomography is suitable for studying the effect of protein expression on mitochondrial ultrastructure. We expect that comparative tomographic analysis may reveal OPA1-directed changes in cristae structure.