finally membrane-associated autophagy signaling, which was regulated in a virulence-selective manner. Collectively, our data shows that Mtb can fine-tune its interaction with host cell membranes and proteins governed by thenature of exposed lipid in its outer membrane. These findings will deepen the understanding of host-pathogen interactions and facilitate discovery of the host membrane-associated novel therapeutic targets.

800-Plat
Transition States of Passive Lipid Transport are Characterized by Hydrophobic Contacts
Julia R. Rogers, Phillip L. Geissler.
Department of Chemistry, University of California, Berkeley, Berkeley, CA, USA.
Cell homeostasis requires the maintenance of heterogeneous membrane compositions through both vesicular and non-vesicular transport mechanisms. Yet, our biophysical description of non-vesicular lipid transport remains incomplete. To help fill these gaps, we aimed to identify an accurate reaction coordinate for passive lipid exchange between membranes. Towards this goal, we investigated the elementary steps of lipid exchange, lipid desorption and insertion into a membrane, using molecular simulation. From over 1,000 lipid insertion trajectories of all-atom and coarse-grained lipid models, we discovered a free energy barrier for lipid insertion and identified multiple pathways characterized by splayed lipid intermediates. This barrier appears hidden when only the lipid's displacement normal to the bilayer, which has traditionally been used to describe lipid exchange, is monitored. In contrast, an accurate reaction coordinate measures the breakage and formation of hydrophobic lipid-membrane contacts, which give rise to a barrier for lipid insertion. At the transition state, hydrophobic contacts are just as likely to form as they are to break. Consistent with this fact, membrane distortions and solvent fluctuations, which can both enable and prevent hydrophobic contact formation, are observed in the transition state ensemble. Overall, our results demonstrate that the formation and breakage of hydrophobic contacts is rate limiting for passive lipid exchange and provide a foundation to understand the catalytic function of lipid transfer proteins.

801-Plat
Differential Actin Binding Affinity Leads to Protein Sorting in a Reconstituted Active Composite Layer
Abrar A. Bhat[1], Amit Das[2], Kabir Husain[2], Madan Rao[2], Darius V. Koester[3], Satyajit Mayor[1].
[1]National Centre for Biological Sciences, Bangalore, India, [2]Simons Centre for the Study of Living Machines, National Centre for Biological Sciences, Bangalore, India, [3]Centre for Mechanochemical Cell Biology and Division of Biomedical Sciences, Warwick Medical School, University of Warwick, Coventry, United Kingdom.
Various functional cell surface proteins bind to cortical actin and undergo actomyosin driven clustering. GPI-anchored proteins, T-cell receptors, glycoproteins such as CD44, and E-cadherins are some reported examples of such patterning. Given the density and diversity of proteins in the plasma membrane, the question arises how the local accumulation of specific molecules is controlled spatiotemporally. Here we explore the possibility that molecular patterning and cluster formation of membrane proteins arises from the combination of differential binding of surface molecules to actin filaments and non-equilibrium actomyosin dynamics. Using a reconstitution system of actomyosin networks tethered to supported lipid bilayers, we provide evidence for the proposed active sorting mechanism. We show that given pairs of actin-binding proteins with differential affinity for actin can be driven into defined patterns by actomyosin dynamics; the patterns disperse in the absence of activity. Our experimental findings are well explained by theoretical simulations that incorporate actomyosin activity, the binding affinity and the relative abundance of the membrane-actin linkers, spanning a phase space of different sorting scenarios. This patterning mechanism could be a key in understanding the spatio-temporal organization and regulation of various processes at the living cell surface.

802-Plat
Non-equilibrium Thermodynamics and Hydrodynamics of Lipid Membranes
Amaresh Sahu, Joel Tchoufag, Yannick Azhri Din Omar, Yulong Pan, Kranthi K. Mandadapu.
Chemical and Biomolecular Engineering, UC Berkeley, Berkeley, CA, USA.
Biological lipid membranes make up the boundary of the cell and many of its internal organelles, including the nucleus, endoplasmic reticulum, and Golgi complex. Such membranes are not simply static, semi-permeable barriers protecting their internal contents, but rather play a dynamic role in many cellular processes. Importantly, lipid membranes are unique materials where lipids flow in-plane as a two-dimensional fluid, yet the membrane bends out-of-plane as an elastic shell. Many past works neglected in-plane fluidity when describing lipid membrane shapes and their stability, thus ignoring important hydrodynamic couplings between in-plane and out-of-plane behavior. We develop an irreversible thermodynamic framework for arbitrarily curved and deforming lipid membranes, and determine their dynamical equations of motion—including the aforementioned coupling. We find in-plane viscous stresses arising from lipid flows lead to an out-of-plane force, despite the membrane bending elastically out-of-plane. We non-dimensionalize the dynamical equations and find a new dimensionless number, named the *Scriven–Love number*, comparing out-of-plane viscous forces to well-known bending forces. Works ignoring membrane fluidity implicitly set this number to zero, however we calculate the Scriven–Love number in past experimental works and find many instances where it is large. In such cases, membrane dynamics are governed by out-of-plane viscous forces, bending plays a negligible role, and we can find novel membrane shape changes and instabilities. For example, in tubes the Scriven–Love number mediates a hydrodynamic instability involving in-plane lipid flows. At low Scriven–Love numbers, tubes undergo a pearling instability. At high Scriven–Love numbers, however, time-oscillating solutions are admitted and moreover the tube is convectively unstable: local perturbations are carried with a base flow of lipids to yield nontrivial membrane shapes. Our results may be relevant in understanding various biological situations, such as lipid flows along an axon body and tubes shooting from the endoplasmic reticulum.

803-Plat
Molecular Transport and Spatial Sorting of Membrane-bound DNA Nanostructures by a Biological Reaction-diffusion System
Beatrice Ramm[1], Alena Khmelinskaia[2], Philipp Blumhardt[1], Hiromune Eto[1], Kristina A. Ganzinger[3], Petra Schwille[1].
[1]Cellular and Molecular Biophysics, Max Planck Institute of Biochemistry, Martinsried, Germany, [2]University of Washington Institute of Protein Design, Seattle, WA, USA, [3]AMOLF, Amsterdam, Netherlands.
Lateral heterogeneity and spatial patterning of proteins observed in biological membranes have been accredited to a wide variety of phenomena including lipid rafts, phase separation and curvature recognition, but remain poorly understood.Here, we found that a biological reaction-diffusion system, the *Escherichia coli* MinDE system, induces patterns and gradients of completely unrelated membrane-bound macromolecules by a non-specific mechanism. A paradigm for pattern formation, the MinDE system is based on the ATPase MinD, its activator MinE and the membrane as a reaction matrix. This minimal oscillator defines midcell in *E. coli*. Using a well-established *in vitro* reconstitution assay on supported lipid bilayers we show that MinDE dynamics are able to spatiotemporally regulate functionally unrelated membrane proteins. Intriguingly, the ATP-driven MinDE self-organization induced directed and active net transport of lipid-anchored proteins, establishing large-scale gradients on the membrane. To interrogate the phenomenon in a defined manner we employed this simplistic transport mechanism for positioning of a synthetic cargo: membrane-anchored DNA nanostructures. By varying the number of membrane anchors and the size of the highly controllable DNA origami we determined the influence of cargo properties on its spatiotemporal positioning by MinDE. We find that the diffusion coefficient and the membrane footprint of the target molecule determine the extent of the regulation. Using this knowledge, we are able to sort and spatially separate DNA origami according to the number of membrane anchors by MinDE self-organization. These findings imply that MinDE are able to position a much larger set of proteins in the cell than previously known. We further speculate that also other reaction-diffusion systems are capable of regulating a large set of proteins by similar non-specific interactions, hinting towards a generic mechanism to couple ATP consumption to protein patterning and sorting.

804-Plat
The Combined Hydrodynamic and Thermodynamic Effects of Immobilized Proteins on the Diffusion of Mobile Transmembrane Proteins
Rohit R. Singh[1], Ashok Sangani[2], Susan Daniel[1], Donald Koch[1].
[1]Dept Chem & Biomolec Eng, Cornell Univ, Ithaca, NY, USA, [2]Syracuse Univ, Syracuse, NY, USA.
The diffusivity of transmembrane proteins in the plane of the membrane has been shown to be one or two orders of magnitude lower in the plasma membrane than in reconstituted vesicles. One hypothesis for this observation is that the plasma membrane contains immobilized transmembrane proteins which slow the diffusion of mobile proteins. Previous studies have described the retardation of the diffusivity of mobile proteins by hydrodynamic and thermodynamic interactions with immobile proteins acting independently. Using a multipole solution for the hydrodynamic mobility in the array and a finite element solution of the concentration of mobile proteins, we show that the long-time diffusivity of a mobile protein resulting from coupled hydrodynamic