

## New and Notable

### Quantifying Membrane Asymmetry

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Molecular transmembrane asymmetry plays a major role in such diverse cellular aspects as morphology, adhesion, and signaling. In general, surface receptor proteins have a different molecular architecture on their cytoplasmic and extracellular side corresponding to the respective function. Likewise, lipid asymmetry between the two monolayers of eukaryotic plasma membranes and membranes from organelles, bacteria, and viruses is well established (Devaux, 1991; Dolis et al., 1997).

As a morphological response to a transbilayer asymmetry in lipid species and/or different aqueous environments, a cellular membrane generally adopts a curved equilibrium configuration. During the last decade, an elegant description of the morphology of membranes and in particular of free vesicle shapes has emerged from a general theory of bending elasticity (Seifert, 1997). Within the area-difference elasticity model, asymmetry is quantified via both the area difference between the two monolayers of a vesicle and the spontaneous curvature of the membrane. These quantities may be combined into an effective differential area (Mui et al., 1995; Döbereiner et al., 1999).

In an excellent paper published in this volume, Heinrich et al. apply the area-difference elasticity model or the generalized bilayer-couple model, as the authors call it, to the analysis of vesicle deformations by an axial load. The authors describe in amazing detail the morphological changes of a vesicle as it is strained axially with micropipettes. It is well known that a sufficiently large point force acting on a fluid membrane leads to the formation of a long tubular appendix. The authors' main finding is that the precise morphological scenario with which such a tether is pulled out of a vesicle in fact depends quite delicately on membrane asymmetry. It is therefore possible to measure membrane asymmetry with this technique. Tether-pulling experiments are complementary to morphological measurements on fluctuating vesicles (Döbereiner et al., 1999). Both techniques allow quantitative determination of the tendency of a membrane to bend. Due to the relatively high forces involved in tether-pulling, the regime of strong membrane

asymmetry, which is not easily accessible by monitoring freely fluctuating vesicles, may be studied.

Heinrich et al. provide a profound theoretical basis for the analysis of such tether-pulling experiments, which in the past have relied on a number of ad hoc assumptions and approximations. The authors have developed a numerical technique that allows computation of complex membrane morphologies characterized by strongly varying curvature (e.g., a sphere with a thin tether) with very high precision. The calculation of such shapes has not been possible before now. Tether-pulling experiments yield important mechanical characteristics of membranes, like bending moduli, in addition to the effective differential area encoding membrane asymmetry. Further, it is possible using this technique to acquire information on dynamic features, e.g., driven lipid flip-flop and intermonolayer friction. This versatile usage of tether-pulling is nicely summarized in the rather complete historical account the authors give in their Introduction. The elastic interactions of the plasma membrane with cytoskeletal proteins may be studied more quantitatively than has been possible so far using the theoretical analysis of Heinrich et al. For instance, it is now feasible to perform a comprehensive analysis of the buckling instability of microtubules pushing against membranes (Kuchnir Fygenon et al., 1997). In summary, the results of Heinrich et al. open far-reaching possibilities for further quantitative work on tether-pulling. This technique will continue to play a major role in the quest to understand the behavior of biomembranes.

In particular, the determination of membrane asymmetry by tether-pulling and observations of fluctuating vesicles will deepen our knowledge about cellular processes. One may speculate about a cross-coupling of the morphology of a membrane with its signaling function via molecular asymmetry. One example of such a relationship may be the inositol phospholipid pathway or another biochemical cascade involving lipid-derived second messengers (Ghosh et al., 1997). In the former pathway, the phospholipase C cleaves off the head group of inositol phospholipids to produce inositol triphosphate and diacylglycerol. The latter lipid has quite a small head group and, thus, membrane asymmetry clearly changes during this process. The variation in membrane asymmetry could, in turn, trigger morphological transformations of the bilayer. It may also be used to couple distinct biochemical reactions. An asymmetry-induced change in the mechanical stress profile of the bilayer membrane could alter the activity of enzymes or ion

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channels that are some distance apart (Keller et al., 1993). It remains to be seen whether these effects are in fact widely employed by nature.

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