DOI:10.36471/JCCM_March_2025_02

Tension doesn't always propagate in a biological cell membrane

1. **Cell Membranes Resist Flow**
   Authors: Zheng Shi, Zachary T. Graber, Tobias Baumgart, Howard A. Stone, Adam E. Cohen
   Cell 175, 1769–1779 (2018)

2. **Cell protrusions and contractions generate long-range membrane tension propagation**
   Authors: Henry De Belly, Shannon Yan, Hudson Borja da Rocha, Sacha Ichbiah, Jason P. Town, Patrick J. Zager, Dorothy C. Estrada, Kirstin Meyer, Hervé Turlier, Carlos Bustamante, and Orion D. Weiner
   Cell 186, 3049–3061 (2023)

Recommended with a Commentary by Christoph F. Schmidt, Department of Physics, Duke University

Biological cells are contained and insulated against their environment by a cell wall. In animals, this cell wall consists of a lipid bilayer supported by a cytoskeletal cortex consisting mostly of actin, a filamentous protein polymer. The lipid bilayer typically contains about 50% membrane proteins by weight. This cell membrane is generally believed to respond like a 2D viscous fluid, with a relatively high viscosity, comparable to a substance like honey at room temperature. In a fluid, pressure, or in the case of a 2D membrane, tension gradients would lead to flow and were always expected to rapidly relax. Because a fluid lipid or lipid-protein layer should have very small compressibility, local tension changes were thought to be transmitted as phonons in the fluid across the whole cell membrane within milliseconds [1]. This was thought to create a rapid physical communication mechanism from one end of the cell to the other. One particular type of recipients of such communications are mechanosensitive channels, membrane-embedded proteins, a subset of which is considered to open to allow ion flux through the membrane in response to an increase in membrane tension [2]. This view is challenged by the recommended papers from Adam Cohen and collaborators and Orion Weiner and collaborators. In spite of the apparently contradictory titles, both papers agree in their experimental results that local tension spikes, created by membrane tethers drawn with micropipettes, stayed local and did not propagate rapidly to other parts of the cell, unless the lipid membrane was detached from the cell cortex in membrane bulges called blebs, in which case tension did propagate rapidly from one spot on the bleb to another one (Fig. 1). The experimental geometry in both studies was similar,

tethers were pulled from surface-adherent cells in different locations and the question was how much tension, if any, was propagated for one thether to the other. Obviously, force balance holds all the time and if inertia in the system and drag between cell and medium are negligible, the vector sum of the three forces, tether one, tether two and cell-substrate adhesion must always be zero. The interesting finding of both papers is that if, say, tether one is pulled out a bit further, that additional force is not changing tension in tether two, even when the two tethers were only 5 µm (Cohen paper) or 2 µm (Weiner paper) apart, but were evidently balanced by increased substrate-adhesion force instead.

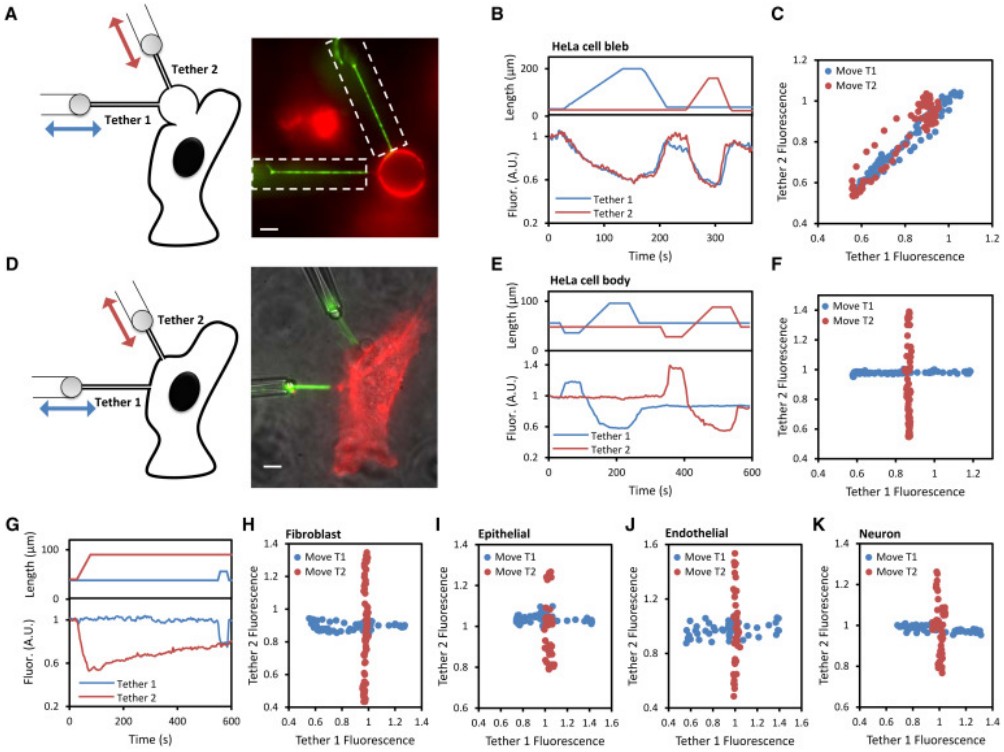

Figure 1: (A and D) Schematic (left) and fluorescence image (right) showing a pair of tethers pulled from (A) a cell-attached bleb or (D) the cell body of a HeLa cell expressing GPI-eGFP. Green: fluorescence under patterned illumination (restricted to dashed boxes); red: fluorescence under wide-field illumination. In (D), a transmitted light image (gray) is combined with the fluorescence images. Scale bars 5 μm. (B and E) The two tethers were stretched sequentially (top), and the fluorescence of each tether was monitored (bottom) in (B) a HeLa cell bleb and (E) an intact HeLa cell. (C and F) Relation between the intensities of the two tethers when either the first or second tether was stretched in (C) a HeLa cell bleb and (F) an intact HeLa cell. (G) Test for slow coupling between tethers in a HeLa cell. A change in length of tether 2 did not affect fluorescence of tether 1 within a 500-s measurement window. (H–K) Repetition of the experiment in (D)–(F) in (H) NIH 3T3 fibroblasts, (I) MDCK epithelial cells, (J) mouse brain endothelial cells, and (K) rat hippocampal neurons. T1: tether 1; T2: tether 2. Reproduced with permission from the first recommended paper.

It is useful to consider time scales and lipid flows at this point. Density fluctuations in

3D-solids and fluids propagate with the speed of sound. In bulk lipids the speed of sound is on the order of 1000m/s [3], meaning that a sound wave would travel in less than a microsecond from one end a cell to another. This is not the appropriate estimate, though, since in a thin film, surface acoustic waves (SAW) dominate [4] and those couple strongly to the viscous embedding medium which would dampen the waves. The two papers we discuss do not look at short time scales, but consider tension gradients that drive membrane flow. If a tension on the membrane in one part of the cell is maintained over time scales longer than any transients, e.g. associated with SAWs, last, in this case by mechanically pulling a membrane tether out of the cell membrane, then flow sets in and tension gradients are maintained until flow stops. Flow velocity is determined by the resistance to flow. The Cohen paper makes the point that the logarithmically decaying flow field (in a 2D fluid) around obstacles, such as the dense array of membrane proteins anchored to the actin cortex, creates an enormous flow resistance. Therefore, the increase of force exerted by tether one will strongly couple to the actin cortex surrounding the base of tether one and from there through adhesion molecules to the substrate. Slow lipid flow into tether one was seen in both studies, as a slow increase of tether diameter after a step displacement (Cohen paper) and a steady increase in tether length under constant pulling (Weiner paper). The Cohen paper then models the propagation of tension as a diffusive process by balancing elastic and viscous forces in the membrane in a Darcy-flow scenario. It is not so obvious if one should not also consider local reservoirs of lipid, which are known to exist, e.g. in the form of caveoli [5, 6], that could feed the tether without pulling lipid from across the cell. Neither of the two studies could experimentally measure any tension propagation, which could be a point in case for local membrane reservoirs providing the path of least resistance to obtain lipid for extending the tether.

In general, it is important to keep tension in the actin cortex separate from lipid membrane tension and from overall cell wall tension which is the sum of the two. While the two layers are dynamically coupled as explained in the Cohen paper, the elastic resting state may be different (and constantly changing in the living cell), the cortex is more or less acting like a shear elastic solid, albeit adaptive and changing on long time scales, and also activated by motor proteins, and the lipid membrane likely has variable surface area due to coupling with caveoli or other cell-internal membrane systems. The point is illustrated by the finding in the Weiner paper that local tension changes involving the whole cell wall do rapidly propagate across the cell (Fig. 2). It is slightly confusing that the authors call this membrane-tension propagation while it is clearly cell-wall tension propagation. The model proposed in the Weiner paper then assigns elasticity to the lipid membrane and viscosity (and activity) to the actin cortex which seems to be the opposite of what one would expect, although the tight coupling between lipid membrane and cortex makes it hard to distinguish the behavior of the two layers.

The Cohen paper goes on to test the activation of mechanosensitive channels in response to membrane tether pulling by fluorescently monitoring intracellular Ca^{2+} levels. They find that channels were triggered close to the tether base, but not elsewhere in the cell. This result clearly challenges the rather well established assumption (extensively discussed in the Cohen paper) that tension-gated mechanosensitive channels react to global membrane tension and points towards an entirely local mechanism of channel activation.

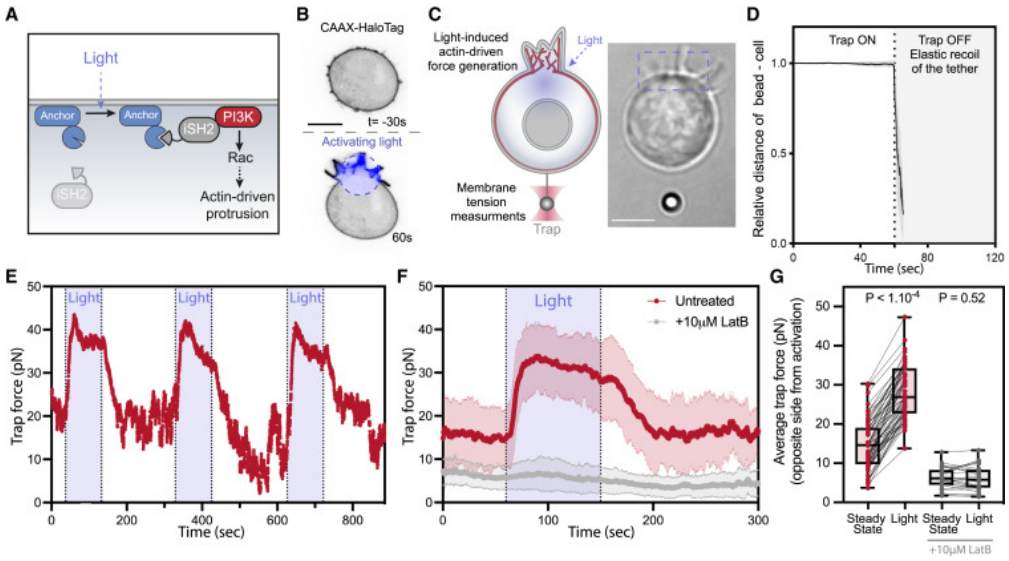

Figure 2: (A) Optogenetic control for light-induced activation of phosphoinositide 3-kinase (PI3K) via localized recruitment of inter SH2 domain (iSH2), resulting in Rac GTPase activation that initiates actin-driven cell protrusions (see STAR Methods). (B) Time-lapse confocal images of a neutrophil-like HL-60 cell expressing opto-construct (Opto-PI3K) and membrane marker (CAAX-HaloTag), showing localized membrane protrusion upon light activation. (C) After light-activated protrusion on one side of the cell (top of frame), changes in membrane tension on the opposite side (bottom of frame) are measured via a membrane tether held by an optical trap. (Right) Brightfield image of a protruding cell during tether pulling assay. (D) After tether pulling measurements, the trapping laser is turned off, and the elastic recoil of the bead toward the cell is observed to confirm the absence of cytoskeleton in the tether (means ± SD; n > 15, N = 5). (E) Representative time trace of trap force (a direct readout of cell membrane tension change) reveals robust and sharp increase in membrane tension over repeating cycles of light-activated protrusion on the opposite end of the cell (as in C); light: 90 s on (shaded area). (F) Red: averaged time trace of trap force before (steady state), during (light), and after activating cell protrusion (means ± SD; n > 60, N = 8). Gray: as a control, averaged trace from cells treated with actin polymerization inhibitor (10 μM latrunculin B) shows little membrane tension change upon optogenetic activation. (G) Averaged trap force before (steady state) and during activation. Box and whiskers: median and min to max; p values from Wilcoxon paired Student's t test. Scale bars: 5 μm. Reproduced with permission from the second recommended paper.

In summary, both papers convincingly show that lipid flow relative to the actin cortex is extremely impeded due to the coupling by anchored membrane proteins. Tension in the lipid membrane therefore propagates very slowly across the cell and is not a rapid long-range communication path as was assumed in the past. These studies are an important step forward in our understanding of the complex mechanics of the cell wall with its protein-packed viscous lipid membrane coupled to the semiflexible, active and adaptive actin polymer network of the cortex.

References

[1] A. Diz-Muñoz, D. A. Fletcher, and O. D. Weiner, Trends in cell biology **23**, 47 (2013).

[2] C. D. Cox, N. Bavi, and B. Martinac, Cell reports **29**, 1 (2019).

[3] G. Hustad, T. Richardson, W. Winder, and M. Dean, Chemistry and Physics of Lipids **7**, 61 (1971).

[4] H. Überall, in *Physical acoustics*, Vol. 10 (Elsevier, 1973) pp. 1–60.

[5] B. Sinha, D. Köster, R. Ruez, P. Gonnord, M. Bastiani, D. Abankwa, R. V. Stan, G. Butler-Browne, B. Vedie, L. Johannes, *et al.*, Cell **144**, 402 (2011).

[6] O. L. Gervásio, W. D. Phillips, L. Cole, and D. G. Allen, Journal of cell science **124**, 3581 (2011).