From DNA damage to epithelial integrity: new roles for cell forces

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Forces on and within Cells is the theme of the 14 papers accepted by the publication deadline for this special issue of *Molecular Biology of the Cell*. The experimental systems examined range from stem cells and T-cells to yeast, worm, fly, and fish cells, all of which highlight the conceptual approach. The field concerns itself with forces and resistance to shape change at scales from the molecular or nuclear to the embryonic and organ scale. Forces are generated actively by cells—mostly the cytoskeleton—as the cells pull and push to reshape themselves or move their organelles and/or themselves in around movable or immovable surroundings. Forces are also generated externally, by nearby cells or perhaps at larger scales by tissue forces or exterior fluids, or contact pressures, with similar questions for cell and molecular consequences and responses. A key aspect of this highly quantitative science is mathematical modeling rooted in physics (rules that apply independent of the detailed molecules) that makes some testable assumptions, and generates concrete predictions to drive cell biology in rational and hopefully exciting new directions. For example, who would have thought that DNA damage relates to forces in or on a cell…?

Cytoskeletal forces that control chromosome position in division should take care not to compromise genome integrity. **Cassi Estrem and coworkers** use imaging of chromatin and DNA damage repair factors in budding yeast during nuclear migration and squeezing through the bud neck. This mitosis is closed within an intact nuclear envelope, and so very clever genome engineering is used to visualize and measure strain in chromatin during squeezing. Surprisingly, high forces and strains generated by the microtubule system cause a delay in repair and thereby increase foci of DNA damage. Rescue is achieved by decreasing force and also by increasing neck diameter, which decreases nuclear strain. The results resonate with findings reported in this issue for interphase stem cells migrating through small pores. **Lucas Smith and coworkers** propose such a model for what can happen as a stem cell migrates through dense matrix to a site of injury prior to differentiation and tissue regeneration. Nuclear envelope rupture leads to loss of transcription factors and DNA repair factors, which associates with increased DNA damage and suppressed cell cycle, but rupture can be rescued by inhibiting actomyosin, which slows migration. For muscle stem cells, rupture suppresses differentiation, while for mesenchymal stem cells, osteogenesis is increased, which is potentially a new mechanism of bone formation and of calcification in fibrotic tissues such as diseased muscle.

Forces on the nucleus and its nuclear lamina are often exerted by the cytoskeleton through highly conserved linkages of so-called KASH domain proteins that help cross the double bilayer space of the nuclear envelope. **Zeinab Jahed and coworkers** combine genetic manipulations of the model worm *Caenorhabditis elegans* with atomistic simulations in studies of the different lengths of cytoskeleton-binding KASH domain proteins. Longer is stronger in tension, they show: swapping to short domains that also lack some key interacting amino acids fails to allow cytoskeleton-driven movements of nuclei. This defect is evident in multinucleated tissue, where interphase nuclei don't disperse as normal but instead aggregate together. Such aggregated nuclei differ of course from polyploid and aneuploidy nuclei that generally result from a failure to properly segregate chromosomes during mitosis. Mirror symmetric division with one cytokinetic furrow is the typical outcome of two centrosomes that set up a suitable force balance. **Tomo Kondo and coworkers** study a *C. elegans* mutant that mostly exhibits three or more centrosomes per cell, which potentially sets up a complex, multipole tug-of-war. They observe nonetheless that mirror symmetric–type division predominates in experiment, with calculations indicating that this results from polarized cell shapes and cortex-derived tensions.

Cytokinetic rings also generate actomyosin contractile forces, but forces are orthogonal to cell polarization and highly dynamic, with a key question being what sets the rate of constriction to generate daughter cells. **Shuyuan Wang and coworkers** use molecularly explicit simulations to explain the abnormally rapid constriction of rings that are unanchored from the yeast membrane. A specific organization and barbed-end anchoring scheme is implicated because, in the absence of membrane resistance, the tensionless filaments shorten at the high velocity typical of load-free myosin. The latter concept has been enshrined for decades in muscle physiology as A.V. Hill's equation that relates the speed of contraction inversely to the force of contraction. Nonmuscle cells display muscle-type sarcomeric structures, but the preferred description is that striated muscle with its high contractility is a high-density, highly evolved form of the more ubiquitous nonmuscle cell biology. **Shiqiong Hu and coworkers** image the latter in embryonic fibroblasts and show how sarcomere-resembling stacks of myosin filaments emerge from linear fibers. This proceeds and builds with alpha-actinin, which probably acts like a uniform cross-tie between railroad tracks, but the process is blocked somehow by tropomyosins that bind along actin filament tracks. Surprisingly, highly ordered actomyosin leads to lower cell tension, smaller adhesions, and smaller traction forces—which perhaps hints at additional molecular mechanisms

DOI:10.1091/mbc.E19-06-0338
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(such as isoform switching to cardiac or muscle myosins) for specialization of high contractility.

F-actin can of course drive outward protrusions of the cell membrane, without myosin-II, which is a problem that George Oster and his students showed fit very well with the physics of a "Brownian ratchet" in which thermal fluctuations of a membrane are blocked on one side as actin monomers assemble on that side. **Oleg Igoshin and other former students and colleagues of George Oster** summarize some of the many contributions that George made to molecular, cell, and tissue biology in an article that they dedicate to the memory of George Oster (who the present author had the privilege of knowing as a graduate teaching assistant for several years at UC Berkeley). Another actin polymerization problem that George and his students worked on most recently with **outgoing *MBoC* Editor David Drubin** is endocytosis, which is driven by F-actin polymerization. **Masoud Nickaeen and coworkers** calculate the physics of this process in yeast by accounting for the multiple actin-binding proteins involved in the directed assembly of a cross-linked and entangled, visco-active gel and by also accounting for the turgor pressure that opposes volume decrease. Forces and shapes are reasonable, and the theory-based predictions will no doubt drive the experimentalists to make more measurements and deepen the understanding of endocytosis not just in yeast but in other cell types under diverse conditions. **Jeff Hardin and coworkers** model computationally a much larger scale process of convergent extension in development—where George Oster also made pioneering theoretical contributions. Stretching of almost any substance in one direction tends to cause lateral shrinkage in orthogonal directions, which conserves volume. This so-called "Poisson effect" captures some key aspects of convergent extension, even moreso when coupled to epithelial cells modeled by lines and vertices that actively rearrange under tension. Such explanations drive the field forward by identifying gaps in measurements including both the passive physical properties of tissues on relevant time scales and the active responses that underlie reshaping of epithelial cells in context.

Compliance of epithelial layers must be balanced by resilience to maintain the epithelium's barrier function that separates and protects inner tissue from hostile environs. **Lathiena Manning and coworkers** characterize two adherens proteins that back each other up in maintaining epithelial integrity during cell–cell remodeling. *Drosophila* fly embryos show that junctional remodeling such as in mitosis, or in cell intercalation, or under actomyosin cable forces maintains junctional–cytoskeletal linkage and apical contractility because of fly homologues of the proteins Afadin and ZO-1. One of these can be deleted but not both without major disruption. **Jovany Franco and coworkers** study the related process of wound healing in live zebrafish. Sacrifice might be key as local cell crowding from directed migration after wounding is seen to extrude seemingly healthy cells from the epithelium. Mechanically regulated stretch-activated ion channels contribute, with perturbations suppressing the extrusion and favoring proliferation. The observations are surprising in that healing requires loss (not gain) of healthy cells, and they seem a major challenge for theoretical models that should eventually seek to explain the cell and molecular basis for epithelial barrier integrity. **Amity Eaton and coworkers** also focus on this adhesion-actomyosin super-assembly that is referred to as the apical junctional ring, and they specifically examine epithelial cells on the bladder's lumen. Drink a lot, and the ring expands dependent on actin assembly and on exocytic trafficking (which should add membrane) but independent of myosin-II contractility; take a trip to the restroom, and the ring quickly contracts, dependent on contractility as well as actin polymerization and endocytosis. Throughout, neighboring cells maintain integrity and thus prevent leakage into the wrong spaces, which is all good to keep in mind in daily duties.

Immune cells and other blood components constitute a fluid tissue that is subject to very different types of forces, stresses, and strains compared with solid tissues. **Robert Pullen and coworkers** model computationally the dynamics of T-cell interactions with the surface of an antigen-presenting cell, which is a very timely topic given the tremendous progress in T-cell–based therapies. Cell surfaces aren't smooth, and microvilli on T-cells certainly complicate the forces on receptor–ligand bonds. Additional complexity arises from so-called "catch bonds" that strengthen under force and seem unique to biology; most inanimate materials simply break under high enough force. The net effect leads to a nontrivial feedback between binding and microvillus motion, which selectively stabilizes scanning microvilli for antigen discrimination. **Zhenhai Li and coworkers** devote their attention to the molecular physics of the large blood polymer Von Willebrand factor, which has long been known to not just extend under flow but then be cleaved and activated by protease. Single-molecule force measurements are applied to various constructs to uncover the stretch-dependent interactions and their force-dependent lifetime, revealing site interactions that depend on forced conformation and sometimes calcium too. The highly reductionist studies certainly underscore the richness of force-dependent transitions that likely underlie the more complex cell and tissue systems elaborated throughout the rest of this special issue.

Finally, the editors of this special issue appreciate the opportunity, made possible by *MBoC*'s leadership, to collect such a set of papers in one convenient volume. There are no doubt many additional 2019 papers that could also fit into this issue. Society journals have long had a crucial role in identifying fields of growing interest to subsets of members of the society. Mechanobiology is one such topic in resurgence, one might say, as it increasingly integrates ever-improving microscopy-derived observations of shape changes at many scales with increasingly facile molecular manipulations that perturb the systems and thereby provide data sets for theory that, if done well, can predict what's next. Please enjoy!

ACKNOWLEDGMENTS

This work was supported by the National Cancer Institute under Physical Sciences Oncology Center Award U54 CA193417, National Heart, Lung, and Blood Institute award R01 HL124106, Human Frontier Science Program grant RGP0024, the United States–Israel Binational Science Foundation, a National Science Foundation Materials Science and Engineering Center grant to the University of Pennsylvania, and also grant agreement CMMI 15-48571.

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