concentrations and substrate stiffness. Low shear stress levels in the range of 0.2 Pa are able to trigger a cellular response and hence can be used to effectively drive cells to follow predetermined sequences of steps including straightforward motion, immobilization and reversal. In addition, a wide range of extracellular calcium concentrations have been investigated in our study. We found that soluble calcium in the extracellular environment plays an important role in mechanotactic cell guiding and control. The existence of an optimal value for the calcium concentration for both the average cell speed and the directionality can be clearly observed.

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Molecular Tattooing of Live Cells and Animals: Spatial and Time Specific Effects of Azidoblebbistatin by Two-Photon Microscopy

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Until now the greatest limitation in the application of bioactive compounds has been the inability of confining them specifically to single cells or subcellular components within the organism. Recently, we synthesized photoinducible forms of bioactive compounds which can be covalently attached to their target enzymes, resulting in prolonged effects. Furthermore, we revealed that photoactivation can be initiated by two-photon excitation, thereby the bioactive compounds to their targets can be covalently attached in vivo, localizing their effects to femtoliter volumes in cellular or subcellular level. We specifically inhibited myosin II molecules in different parts of the migrating posterior lateral line primordium (pLLp) of zebrafish embryos by the use of our recently published photoinducible myosin inhibitor - azidoblebbistatin - and a two-photon microscope. Our results demonstrate that local inhibition of myosin II suppresses the cytokinesis of the neuromast's cells but does not affect directly the movement of pLLp. We also found that the dividing cells of pLLp are located in the frontal zone of the primordium.
Molecular tattooing by using photoactivable azidoblebbistatin enables us to control cell migrations in cellular and subcellular level.

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Cell Size and Shape Regulates Epithelial-Myofibroblast Transition

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Myofibroblasts aid in wound healing and exert large contractile forces in order to assist in wound closure. Aberrant activation of myofibroblasts can lead to the development of pathological conditions including fibrosis, cancer, and the foreign-body response to implanted biomaterials. Myofibroblasts can develop from epithelial cells through an epithelial-mesenchymal transition (EMT). During EMT, epithelial cells loosen attachments to their neighbors and acquire an elongated morphology. These phenotypic changes are accompanied by alterations in gene expression patterns including upregulation of cytoskeletal proteins which contribute to the ability of myofibroblasts to exert large contractile forces. Here, the relationship between cell shape and cytoskeletal tension and the expression of cytoskeletal proteins in transforming growth factor (TGF)-β1-induced EMT is determined. We employ a microcontact printing approach to control cellular shape and we find that permitting cell spreading promotes the increased expression of myofibroblast markers while restricting cell spreading prevents EMT. Furthermore, we find that the subcellular localization of key mechanoresponsive molecules is regulated by cell shape and plays an important role in regulating the expression of cytoskeletal associated proteins. Results provide insight into how mechanical signals can control the development of myofibroblasts and may suggest ways to engineer therapeutic solutions for fibrosis and cancer.

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Experimental Measurement and Simulations of the Cytokinetic Ring Tension in Fission Yeast

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Cytokinesis in animals and fungi requires the assembly and constriction of an actomyosin contractile ring at the site of cell division, but the ring tension, the ring's principal mechanical property, has rarely been measured. Thus it has not been possible to relate the organization of the ring to its principal function, and the mechanism of ring constriction remains poorly understood. Here we combined experimental measurement and mathematical modeling to characterize the tension of the cytokinetic contractile ring and its relation to organization. We used *Schizosaccharomyces pombe* protoplasts, whose cell walls have been enzymatically digested, to measure the tension. Based on extensive biochemical and genetic characterization of the components of the *S. pombe* ring, we developed a detailed computer simulation incorporating the key components. The simulation calculated the tension and organization of the ring for direct comparison with our experiments. We report the first measurements of contractile ring tension in fission yeast, using micropipette aspiration of protoplast cell membranes and optical microscopy analysis of membrane deformation by the tense ring as it constricts. The measured values are in close agreement with our ring simulations. The simulations show that the ring components undergo a remarkable process of continuous self-organization into a tight tension-generating bundle. The bundle produces high tension by orienting actin filaments parallel to the ring, and by concentrating actin filaments and myosin-II clusters to enhance the number of force-producing interactions. The self-organization naturally provides the mechanism to continuously remodel the ring as it constricts without loss of tension, a long-standing puzzle concerning the mechanism of ring constriction.

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3D Model of Cytokinetic Ring Assembly in Fission Yeast

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During fission yeast cytokinesis, actin filaments are nucleated in random directions by cortical Cdc12 and captured by myosin-II motors from neighboring nodes, exerting forces that pull them together into a contractile ring. Contractile ring assembly has been previously described using a stochastic 2D computational model based on a "search, capture, pull and release" mechanism (SCPR). In this model, actin filaments are assumed to polymerize along the cell membrane, consistent with enhanced concentration of actin filaments near the cell membrane in experiments. Recent observations in mutant and wild-type cells have suggested that actin filaments nucleated in the cytoplasm and in non-medial locations can also participate into the assembly of the cytokinetic ring as well as in clump, star, and meshwork actomyosin morphologies. The relative contributions of the non-medially nucleated actin filaments has been debated. In order to examine these effects that involve 3D dynamics and to better understand the mechanisms that confine actin filaments along the cell surface, we extended the SCPR model to 3D. In this model semiflexible actin filaments (represented as beads connected by springs) grow from nodes and they can be captured by myosin in neighboring nodes. Cross-linking by alpha-actinin and fimbrin is represented by an attractive interaction between filament beads. We also implemented cytoplasmic formin nucleation and actin cable incorporation into the contractile ring. We quantified the resulting distribution and orientation of actin around the band of myosin nodes and studied the effects of varying model parameter values. We describe the conditions under which ring, clump, transient star and meshwork structures may form along the cell membrane and compare to prior experiments.