highly influences cell spreading, morphology and cell fate. Moreover, when cells spread on constraining adhesive micropatterned substrates, reproducible cell cytoskeleton and adhesion site organizations are observed. This suggests that a simple physical energy minimizing process drives cell adhesion when considering two cellular tensions at the interface, one along and the other normal to the cell interface. These active tensions are due to myosin II activity that maintains cell shape and which relies on the stabilization of actomyosin bundles.
To characterize cellular active tensions, we investigate how peripheral contractile bundles are formed, stabilized and rearranged during cell spreading. Experimental quantitative analyses are carried out on living or fixed cells spreading on adhesive patterns, where protrusions and adhesions are spatially restricted. Using fluorescently labelled F-actin on living cells, we observe that curvature radius of peripheral bundles increase during the spreading phase concomitantly with the increase of the bundle spanning distance. This supports the idea that bundle tension may depends on its length. Once cells have covered the whole adhesive pattern, further increase of the curvature radius often happens, suggesting that cells relax to a tensile steady state on longer period.
On fixed cell populations, similar correlations between curvature radius and spanning distance are observed. Close analysis of bundle strength and myosin II distribution suggest the existence of these dynamical processes. Analysis of the assembly of extracellular matrix proteins into fibrils revealed that tangential tension (i.e. along peripheral bundles) may be prominent compared to inward assemblies, mainly observed on spreading cells on homogeneous substrate.

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Evaluating Tension in Actomyosin Bundles at the Cell Periphery
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During cell spreading, interactions between cells and their local environment drive key processes such as motility, differentiation and division. Both chemical and mechanical factors regulate these cellular responses including the substrate topology that highly influences cell spreading, cell morphology and cell fate. Cell shape and cytoskeletal tension are maintained by the activity of myosin II. When cells spread on micropatterned adhesive substrates, reproducible cell cytoskeleton organizations into curved actomyosin bundles at the cell periphery are observed. To characterize cellular active tensions and their dependencies on geometries and on myosin II activity, the tension of peripheral contractile bundles is determined using different techniques of cellular force measurements.
Traction force microscopy (TFM) allows measurement of cellular forces exerted on the adhesive substrate by registering fluorescent beads displacements embedded in soft gels, and by then computing the corresponding cellular stress applied on the substrate. We combine micropatterning technique and TFM to investigate the tension of the peripheral contractile bundles depending on the pattern and cell geometries.
By using ultra-soft cantilevers and three-dimensional patterned substrates, we perform mechanical probing on peripheral contractile bundles enabling, at small deformations, to obtain direct tension measurements. By adjusting indentation magnitude and deformation speed, we aim to determine viscoelastic properties of actomyosin bundles on living cells.
Both the passive (TFM) and active (cantilever-based) experiments suggest a range of bundle tensions from 10 to 100 nanonewtons. Using drugs to vary the contractility level of the cell, we can reconstruct the interdependencies between cell contractility (i.e. bundle tension and cortical tension), cell shape (i.e. bundle curvature) and substrate properties (i.e. adhesive geometry and substrate stiffness).

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'Hum'-Corrected Comparison of Viscoelastic Properties of Normal, Tumorigenic, and Metastatic Breast Cells
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We tested the hypothesis that breast cancer cells have lower viscoelastic moduli than their normal counterparts. The moduli were evaluated by tracking peroxisomes within three human mammary epithelial cell types: normal, tumorigenic, and metastatic. Peroxisomes in these cells were labeled with GFP, and then imaged at 100 fps. From the resulting movies of peroxisome motion, mean squared displacements (MSDs) of the peroxisomes were evaluated. To assess the contribution of motor-driven 'hum' of the actomyosin network to the measured MSDs, peroxisome motion was also determined in the three cell types after they were treated with blebbistatin or sodium azide and 2-deoxy-D-glucose. Blebbistatin is a myosin II inhibitor, and sodium azide and 2-deoxy-D-glucose deplete cellular production of ATP. Our results indicate that peroxisome MSDs are larger in the presence of ATP and active myosin motors than when driven solely by thermal energy.
By inserting these MSDs into the generalized Stokes-Einstein equation, apparent viscoelastic moduli were obtained for the cytoplasms of normal, tumorigenic, and metastatic breast cells with and without the motor-driven 'hum'. The generalized Stokes-Einstein equation assumes that the only energy that drives motion of the tracked peroxisomes in the cytoplasm is thermal. Peroxisomes in all three cell types report lowered apparent cytoplasmic viscoelastic moduli in the presence of uninhibited myosin II motors than when driven solely by thermal energy. The viscoelastic moduli determined from metastatic and tumorigenic cells without hum are significantly lower than those from their normal counterparts.
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Regulation for Phosphatidylinositol Lipids Signaling System by Talin
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Cellular motility is a basic function underlying various important physiological phenomena. Actomyosin system generally works as an internal force generator, and external stimulations received via receptor-mediated signaling systems and an adhesive system modulate the motility. The force-generating, signaling and adhesive systems work in cooperation to make a united anterior-posterior polarity, which ensures an effective and adaptive cell migration. The actin polymerization is induced by PI(3,4,5)P3-enriched domains on the cell membrane, which arise from a chemotactic signaling system including PI(3,4,5)P3-producing PI3-kinase and egrading PTEN in a self-organized manner. However, how the adhesive system is correlated to the PtdIns system is unknown, which should to be an essential mechanism in regulating a migration mode dependently on whether the cells move solitarily or collectively. We examined effects of loss-of-function of focal adhesion on the PtdIns dynamics in single-celled amoebae and multicellular structures of *Dictyostelium discoideum*. amoebae of a mutant lacking a component of focal adhesion, talin, by disruptions of *talA* and *talB*, did not adhere to the substratum and exhibited an enhancement of PI(3,4,5)P3 domains traveling on the cell membrane. Due to the conditional up-regulation of PI(3,4,5)P3 production, a response to chemoattractant stimulation was abrogated. Both the PI(3,4,5)P3 dynamics in resting and stimulated cells was recovered by a treatment with PI3-kinase inhibitor, indicating talin suppresses PI3-kinase activity. A single disruption of *talB*, not *talA*, reconstructed the abnormal PI(3,4,5)P3 dynamics, suggesting *talB* is more responsible. The *talB* lacking cells failed to move collectively in the multicellular structures. Therefore, talin may be coupling the adhesion and PtdIns signaling systems via PI3-kinase, and multicellular collective motion is possibly mediated through the coupling by talinB.

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Mechanical Properties of Vimentin Intermediate Filament Networks
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One marker of mesenchymal cells is the expression of vimentin intermediate filament (VIF) proteins, which assemble into networks. The cytoskeleton is typically comprised of these intermediate filament networks, actin, and microtubules. While some aspects of VIF networks have been characterized in bulk, their mechanical properties in situ have not been well defined. Here we use microrheological techniques to study VIF network mechanics in cells in which microtubules and actin have been removed so that VIF can be studied independently of the other cytoskeletal components. Simultaneous imaging of the network allows us to correlate network structure with its local mechanical properties.

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B Cell Receptor Clustering and Signaling Activation are Modulated by Physical Parameters of the Surface
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Antigen binding to the B cell receptor (BCR) induces receptor aggregation into signaling microclusters, actin dynamics and cell spreading, which trigger B cell signaling activation. Recent studies have shown that gathering of surface