Title: Why mechanical forces matter in health and disease: Quantifying cell-generated mechanical forces in high spatial resolution

Abstract

A robust nanopillar platform with increased spatial resolution reveals that perinuclear forces originated from stress-fibers spanning the nucleus of fibroblasts are significantly higher on these nanostructured substrates than the forces acting on peripheral adhesions. Many perinuclear adhesions embrace several nanopillars at once, pulling them into β1-integrin and zyxin rich clusters, which are able to translocate in the direction of cell motion without loosing their tensile strength. The high perinuclear forces are greatly reduced upon inhibition of cell contractility or actin polymerization, and disruption of the actin-cap by KASH dominant-negative mutant expression. *LMNA* null fibroblasts have higher peripheral versus perinuclear forces, impaired perinuclear β1-integrin recruitment as well as Yap nuclear translocation, functional alterations that can be rescued by lamin A expression. These highly-tensed actin-cap fibers are required for YAP nuclear signaling and thus playing far more important roles in sensing nanotopographies and mechano-chemical signal conversion than previously thought. Our ability to control cell behavior by properly engineered materials and microenvironments is tightly coupled to understanding the mechanisms of cell-matrix interactions. Anisotropy of extracellular matrix (ECM) drives cell alignment and directional migration during processes like development and wound healing, but also in cancer cell migration and invasion. So far, migrational persistence whether on flat isotropic or anisotropic surface patterns and fibers was mostly studied as local phenomenon asking how specific integrins are responsible for the recognition of the spatial ECM cues. Yet, cell alignment and directional migration in response to external ECM cues requires signal integration across length scales. By producing patterned 2 μm ECM stripes, which lead to cell alignment, and testing the previously described pan-integrin null fibroblast cells, we indeed observed that cell alignment and directional migration on patterned stripes were lost when β1 integrin was absent. By combining nanopillar arrays with printed cell-adhesive fibronectin (FN) stripes, we could probe subcellular force distributions at submicron resolution. Importantly, directional migration along adhesive patterned stripes was also impaired for lamin A/C knockout cells, incapable of forming an actin cap and restored upon lamin A/C rescue. Together, our data suggest that β1 integrin is required for the recognition of spatial ECM cues and that force transmission to the nucleus via lamin A/C is essential for subsequent directional migration.