



Elasticity, mechanosensing and traction by cells under carrying morphologies

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Abstract: It is unclear whether morphology difference, dictated by the geometry of attachment is critical for cell rheology and mechanosensing. Thus, we studied the rheology and mechanosensitivity of bone cells under different morphologies using atomic force microscopy and our two-particle assay for optical tweezers. Primary osteoblasts were isolated from avian bone using a sequential collagenase treatment. MLO-Y4 osteocytes and MC3T3-E1 osteoblasts were incubated at room temperature in CO₂-independent medium without serum immediately before measurements. Elasticity was determined using a Hertzian contact model for both flat and round morphologies. We found that the elastic modulus of MLO-Y4 osteocytes when flat and adherent (> 1kPa) largely differed when round but partially adherent (< 1kPa). The elasticities of round suspended MLO-Y4 osteocytes, MC3T3-E1 osteoblasts, and primary osteoblasts were similarly < 1kPa. The mechanosensitivity of round suspended MLO-Y4 osteocytes was investigated by monitoring nitric oxide (NO) release, an essential signaling molecule in bone. These cells were stimulated by oscillatory undulations of the integrin-bound spheres upto 30pN force. Interestingly, the NO released increased in response to 5pN force stimulation, in contrast with flat cells, which required 20nNforce stimulation while releasing lesser NO. To simulate a well controlled mechanosensing environment, round suspended cells were attached at opposite ends to fibrin-coated spheres using optical tweezers. Force traction fluctuations by round suspended cells increased amplitude positively correlating to increased optical trap stiffness. In this assay, the trap stiffness simulated extracellular matrix attachment at controlled sites. Our results suggest that a round cellular morphology supports a less stiff cytoskeleton configuration compared with flat cellular morphology.



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