Nanomechanics of the extracellular matrix of lung and heart tissues

Daniel Navajas

Institute for Bioengineering of Catalonia, School of Medicine - University of Barcelona, and CIBER of Respiratory Diseases. Barcelona, Spain
dnavajas@ub.edu

Cells sense and actively respond to mechanical features of their microenvironment. Moreover, mechanical cues have been shown to mediate critical cell functions including proliferation, differentiation, gene expression, contraction, and migration. Therefore, a precise definition of the mechanical properties of the extracellular matrix (ECM) is needed to further our understanding of the cell-microenvironment interplay. We use atomic force microscopy (AFM) to study nanomechanical properties of lung and heart ECM. Thin slices (10-20 μm thick) of decellularized rat lung parenchyma and mouse heart left ventricle are probed with a custom-built AFM attached to an inverted optical microscope. The Young's modulus (E) of the ECM is computed by fitting the tip-ECM contact model to force-indentations curves recorded on the ECM. The complex shear modulus (G*) is measured by placing the tip at an operating indentation of ~500 nm and superimposing small amplitude (~75 nm) multifrequency oscillations composed of sine waves (0.1-11.45 Hz). G* is computed in the frequency domain from the complex ratio between oscillatory force and indentation. We found that lung ECM exhibits scale-free rheology with a storage modulus (G', real part of G*) increasing with frequency as a weak power law [1]. G' values in the lung parenchyma ECM ranged from 6 kPa in the alveolar septum to 15 kPa in the pleural membrane. The loss modulus (G'', imaginary part of G*) displayed a parallel frequency dependence at low frequencies, but increased more markedly at higher frequencies. We assessed the effect of different decellularization procedures on the local stiffness of the acellular lung by measuring E at different sites of rat lungs subjected to four decelularization protocols with/without perfusion through the lung circulatory system and using two different detergents [2]. Lung matrix stiffness revealed considerable inhomogeneity, but conventional decellularization procedures did not result in substantially different local stiffness. We measured E of ECM in healthy and bleomycin-induced fibrotic mouse lungs [3]. The local stiffness of the different sites in acellular fibrotic lungs was very inhomogeneous and increased according to the degree of the structural fibrotic lesion. We also studied ECM nanomechanics of different histological regions of the left ventricle wall of healthy and infarcted mouse hearts [4]. The ECM of the ventricular wall was 2-fold stiffer than the lung parenchyma with G' ranging from 10 kPa in the epicardium and collagen-rich regions of the myocardium to 30 kPa in elastin-rich regions of the myocardium. Importantly, infarcted ECM showed a predominant collagen composition and was 3-fold stiffer than collagen rich regions of the healthy myocardium. ECM rheology of both lung and heart tissues was very well characterized by a two power law model composed of a weak power law with an exponent 0.05, accounting for a viscoelastic solid regime dominant at physiological frequencies, and a second power law with an exponent of 3/4, accounting for a viscoelastic liquid regime at high frequencies. Our AFM measurements define intrinsic mechanical properties of the ECM at the length scale in which cells sense and probe their microenvironment. Regional changes in mechanical properties of the ECM could provide differential mechanical cues to regulate the spatial distribution, differentiation and function of lung and heart cells.

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