

Understanding the biophysics of diabetic nephropathy: modelling TGF β 1 evoked changes in viscoelasticity of epithelial cells of the proximal tubule.

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Aim: To investigate the biophysical mechanisms underlying proximal tubule fibrosis and the impact cell mechanics have on disease progression, we measured the mechanical stress applied to single cells treated with TGF β 1 as a function of time and quantified viscoelastic properties using stress-relaxation mathematical models.

Methods: Cytoskeletal reorganization was assessed, and single cell stress-relaxation testing performed by Atomic Force Microscopy (AFM) based force spectroscopy on model human proximal tubule cells (HK2) +/- TGF- β 1 (10ng/mL) at 37°C. Fitted with a spherical bead, AFM cantilevers functioning under constant force mode measured mechanical forces over time and viscoelastic properties were calculated using the Maxwell model of viscoelasticity.

Results: Data suggest that both control and TGF β 1 treated cells exhibit transient (5sec) force relaxation of 0.7 and 0.3nN respectively, in response to physical deformation. The initial phase was followed by stabilization in force over a subsequent 30sec period. TGF β 1 treated cells demonstrated a reduction in relaxation characteristics, suggesting that complex viscoelastic components are strongly affected by reorganization of the actin cytoskeleton. Data indicate that viscosities after treatment vary up to 35% (12cells, n=3, p<0.001). Treated cells showed a three-fold reduction in experimental decay of the force time curve that leads to significant changes in the adhesive deformation behaviour.

Conclusion: TGF β 1 (10ng/mL) triggered complex nanomechanical changes in the viscous-elastic behaviour of single cells. Our research suggests that the progression of the disease instigates intricate physical changes that may in part, mediate altered cell-ECM interactions linked to altered cell phenotype in tubular injury.

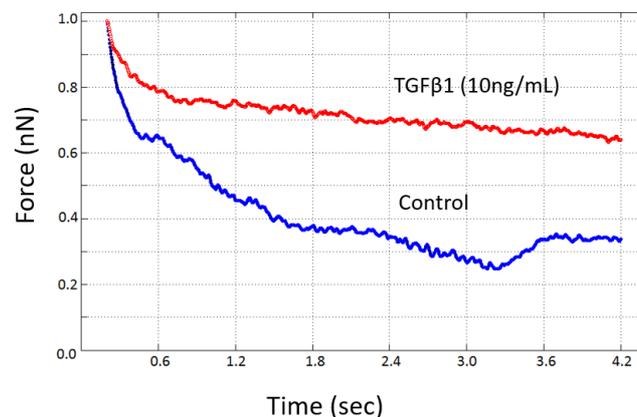


Figure: Experimental Stress-Relaxation data of HK2 +/- TGF- β 1 (10ng/mL).