ANALYSIS OF THE ACTIVE RESPONSE OF CELLS TO CYCLIC LOADING USING A MODIFIED AFM SYSTEM

Noel H. Reynolds, Paul P. Weafer, William Ronan, Patrick McGarry
Department of Mechanical and Biomedical Engineering, NUI, Galway, Ireland

Introduction

Cell function and tissue maintenance are regulated by a dynamic loading environment. Several *in-vitro* studies have reported realignment of the actin cytoskeleton due to cyclic deformation.

In the current study a modified AFM system (Figure 1A) is used to obtain quantitative force data measured under cyclic loading conditions [Weafer, 2012a, b]. It is demonstrated that a bio-chemo-mechanical framework for stress fibre contractility and remodelling [Deshpande, 2006; Ronan, 2012] accurately predicts cell forces under dynamic loading conditions.

Methods

Experimental: Osteoblasts, either untreated or treated with cytochalasin-D (cytoD), are cyclically deformed for 2 hours at a frequency of 1 Hz (Figure 1B). For the first hour, a cyclic strain range of 10-35% is applied (regime 1). For the second hour, a strain range of 0-25% is applied (regime 2). It should be noted that the strain rate is identical for regimes 1 and 2.

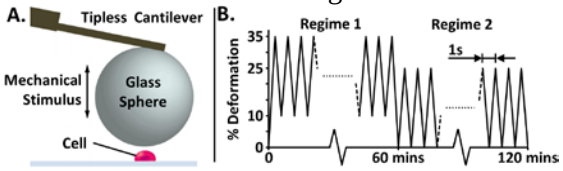

Figure 1: Schematic of modified cantilever (A) and graphical representation of cyclic deformation (B).

Computational: Passive cell behaviour is simulated using a non-linear visco-hyperelastic constitutive formulation:

$$\dot{\sigma} = \alpha\beta e^{\alpha\varepsilon}\dot{\varepsilon} + \gamma(\delta + \sigma - \beta\phi)\{\dot{\varepsilon} - (1/\lambda)\ln[1 + (\sigma - \beta\phi)/\mu]\}$$

where α, β, δ, γ, λ and μ are material constants and $\phi = (e^{\alpha\varepsilon} - 1)$.

Stress fibre contractility in untreated control cells is simulated using an active framework. Tension dependant dissociation of the actin cytoskeleton is predicted using a first order kinetic equation [Deshpande, 2006]:

$$\dot{\eta} = (1-\eta)Ck_f - (1-\sigma/\sigma_0)\eta k_b$$

where η is the dimensionless activation level of a stress fibre bundle. Formation is driven by an exponentially decaying signal C.

The contractile response of the bundles is modelled using a Hill-like equation:

$$\sigma/\sigma_0 = 1 + (k_v/\eta)(\dot{\varepsilon}/\dot{\varepsilon}_0); \qquad -\eta/k_v \le \dot{\varepsilon}/\dot{\varepsilon}_0 \le 0$$

Results

As shown in Figure 2A, cytoD treated cells reach a steady state maximum (max) force of ~30nN during regime 1. At the onset of regime 2, the max force reduces instantaneously and subsequently recovers to only 40% of the regime 1 steady state value. For untreated contractile cells (Figure 2B) a higher steady state max force is measured in regime 1 (~180nN). An instantaneous reduction in force at the onset of regime 2 is followed by a recovery to ~80% of the regime 1 value.

The limited recovery of cytoD cells is captured by the visco-hyperelastic model. The stress fibre contractility model accurately predicts steady state behaviour for untreated cells.

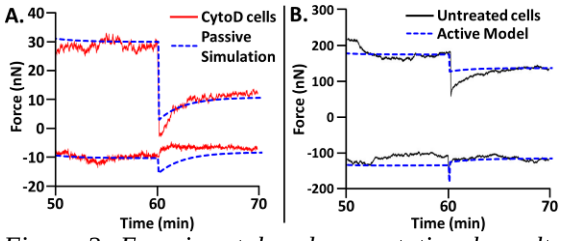

Figure 2: Experimental and computational results for cytoD treated (A) and untreated cells (B). Results shown are ± 10 mins of the regime change region. Maximum and minimum forces at the end of loading and unloading half-cycles are plotted.

Discussion

Forces generated during the cyclic deformation of control cells differ significantly from those of cytoD cells, demonstrating the important role of the actin cytoskeleton in the dynamic response of cells. During constant strain rate cyclic loading, the applied strain range has little effect on the response of contractile control cells, in contrast to the response of non-contractile cytoD cells. Computational predictions suggest that passive visco-hyperelastic material models are suitable only for cells in which the actin cytoskeleton has been removed.

References

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