JOURNAL OF THE MECHANICAL BEHAVIOR OF BIOMEDICAL MATERIALS 14 (2012) 143–157

Available online at www.sciencedirect.com

SciVerse ScienceDirect

journal homepage: www.elsevier.com/locate/jmbbm

Research Paper

Numerical investigation of the active role of the actin cytoskeleton in the compression resistance of cells

William Ronan[a], Vikram S. Deshpande[b], Robert M. McMeeking[c], J. Patrick McGarry[a,*]

[a]Department of Mechanical and Biomedical Engineering, National University of Ireland Galway, University Road, Galway, Ireland
[b]Department of Engineering, University of Cambridge, Trumpington Street, Cambridge CB2 1PZ, UK
[c]Department of Mechanical Engineering, University of California, Santa Barbara, CA 93106-5070, USA

ARTICLE INFO

Article history:
Received 3 February 2012
Received in revised form
22 May 2012
Accepted 24 May 2012
Available online 21 June 2012

Keywords:
Stress fibre
Active contractility
Constitutive formulation
Cell compression

ABSTRACT

Numerous in-vitro studies have established that cells react to their physical environment and to applied mechanical loading. However, the mechanisms underlying such phenomena are poorly understood. Previous modelling of cell compression considered the cell as a passive homogenous material, requiring an artificial increase in the stiffness of spread cells to replicate experimentally measured forces. In this study, we implement a fully 3D active constitutive formulation that predicts the distribution, remodelling, and contractile behaviour of the cytoskeleton. Simulations reveal that polarised and axisymmetric spread cells contain stress fibres which form dominant bundles that are stretched during compression. These dominant fibres exert tension; causing an increase in computed compression forces compared to round cells. In contrast, fewer stress fibres are computed for round cells and a lower resistance to compression is predicted. The effect of different levels of cellular contractility associated with different cell phenotypes is also investigated. Highly contractile cells form more dominant circumferential stress fibres and hence provide greater resistance to compression. Computed predictions correlate strongly with published experimentally observed trends of compression resistance as a function of cellular contractility and offer an insight into the link between cell geometry, stress fibre distribution and contractility, and cell deformability. Importantly, it is possible to capture the behaviour of both round and spread cells using a given, unchanged set of material parameters for each cell type. Finally, it is demonstrated that stress distributions in the cell cytoplasm and nucleus computed using the active formulation differ significantly from those computed using passive material models.

Abbreviations: SF, stress fibre; RVE, representative volume element; E, Young's modulus; ν, Poisson's ratio; SMCs, smooth muscle cells; MSCs, mesenchymal stem cells; FBs, fibroblasts; *nuc*, nucleus; σ_{vm}, tensile equivalent stress, or, von Mises stress

*Corresponding author. Tel.: +1 353 91 493165; fax: +1 353 91 563991.
E-mail addresses: w.ronan1@nuigalway.ie (W. Ronan), vsd@eng.cam.ac.uk (V.S. Deshpande), rmcm@engineering.ucsb.edu (R.M. McMeeking), patrick.mcgarry@nuigalway.ie (J.P. McGarry).

1751-6161/$ - see front matter © 2012 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.jmbbm.2012.05.016

1. Introduction

Previous in-vitro studies have established that cells react to their physical environment and to applied mechanical loading. Cells can sense and actively respond to the stiffness of an underlying substrate, with substrate stiffness affecting cytoskeletal remodelling (Byfield et al., 2009). Furthermore, it has been demonstrated that changes in cell shape or substrate stiffness can cause an increase in apparent cell stiffness (Janmey et al., 2011). However, the mechanisms underlying this active response of cells to the mechanical environment are poorly understood.

Cell compression has been used to quantify differences in the mechanical response of different cell types and shapes. Previous studies have used finite element models in tandem with experimentally measured geometries and compression forces to show that spread cells have a higher apparent stiffness than round cells for a range of different cell types (Caille et al., 2002; Darling et al., 2008). However, these models only consider the cell as a passive entity and assume either elastic, viscoelastic, or biphasic material behaviour. For such passive cell models, a given, unchanged set of material parameters cannot be used to replicate experimentally observed compression forces for round and spread cells. The apparent stiffness of spread cells must be artificially increased to account for the significant cytoskeletal remodelling that the cell undergoes as it changes from a round to a spread configuration (McGarry, 2009; McGarry and McHugh, 2008). In order to gain insight into the mechanisms underlying cellular cell stiffening, it is necessary to employ an active material model that predicts the distribution and contractility of the actin cytoskeleton.

The cytoskeleton has previously been modelled using pre-positioned passive filaments and contractility has been included as a prescribed thermal strain (Mohrdieck et al., 2005; Storm et al., 2005). However, these attempts have not considered the cellular processes that drive cytoskeletal remodelling and contractility. A recent study has proposed a novel computational model of contractile stress fibre (SF) behaviour based on the biochemistry of SF formation (Deshpande et al., 2007). This model is entirely predictive; i.e., the SF distributions and contractility are dynamically governed by cellular signalling and tension dependent dissociation. Models predicting SF formation and contractility have also been proposed by Kaunas and Hsu (2009), and Vernerey and Farsad (2011). All such models have been confined to 1D or 2D formulations; restricting SF formation to a single plane and restricting the ability to simulate in vitro experiments. The models of Kaunas et al. and Vernerey and Farsad differ from that of Deshpande et al. (2007): Vernerey and Farsad assume that the rate of SF formation is increased by fibre tension; Kaunas et al. assume that fibre dissociation occurs when a fibre has been stretched past a critical length. The Deshpande formulation has been used successfully to simulate SF distributions in cells on patterned substrates (Pathak et al., 2008). In a study by McGarry et al. (2009) this formulation is shown to accurately predict the scaling of active cell tractions with cellular contractility and with substrate stiffness for cells adhered to arrays of microposts.

In the current study, this formulation is expanded into a fully 3D framework that allows for the simulation of realistic round and spread cell geometries. This 3D implementation is used to investigate differences in SF evolution in a range of cell types with varying contractility. Simulations are performed for axisymmetric round and spread cells, and for a fully 3D elongated or polarised cell. The effect of cell shape and contractility on the compression response of cells is examined. The results of this study are compared to previous experimental data to illustrate the predictive capabilities of the model. Our findings highlight the importance of SF distribution and contractility in the mechanical response of a cell to applied compression.

2. Methods

2.1. Stress fibre contractility

Stress fibre (SF) formation consists of three coupled phenomena: an activation signal which triggers the formation of the SFs, dissociation of fibres due to a reduction in tension, and a Hill type law relating the contractility of a SF to strain rate.

The role of cellular signalling has been closely linked to cytoskeletal remodelling and mechanotransduction. In this study, the complete signalling pathway which triggers the SF formation is phenomenologically represented as an exponentially decaying signal (Roberts et al., 2001):

$$C = e^{(-t_1/\theta)} \tag{1}$$

where θ is a constant that controls the decay rate of the signal and t_1 is the time since the most recent signal.

Cytoskeletal tension is essential for sustaining SF bundles and a reduction below a defined isometric level leads to fibre dissociation (Franke et al., 1984; Kolega, 1986). The contractile behaviour of assembled SF bundles is similar to that of skeletal muscle. The tension in the SF bundle, which is generated by cross-bridge cycling of actin–myosin pairs (Warshaw et al., 1990), is related to the bundle contraction rate using the following Hill-like equation:

$$\frac{\sigma_f}{\sigma_0} = \begin{cases} 0 & \frac{\dot{\varepsilon}}{\dot{\varepsilon}_0} \le -\frac{\eta}{\bar{k}_v} \\ 1 + \frac{\bar{k}_v}{\eta}\frac{\dot{\varepsilon}}{\dot{\varepsilon}_0} & -\frac{\eta}{\bar{k}_v} \le \frac{\dot{\varepsilon}}{\dot{\varepsilon}_0} \le 0 \\ 1 & \frac{\dot{\varepsilon}}{\dot{\varepsilon}_0} > 0 \end{cases} \tag{2}$$

where σ_f is the stress in the SF bundle, σ_0 is the isometric tension, and $\bar{k}_v$ is the reduction in stress upon increasing the shortening strain rate $\dot{\varepsilon}$, by $\dot{\varepsilon}_0$. The dimensionless activation level of a SF bundle, η, at any orientation, also defines the isometric tension, σ_0, where $\sigma_0 = \eta\sigma_{max}$. σ_{max} is the maximum tension in a fully activated bundle. Fig. 1 shows the variation in tension with strain rate for each part of Eq. (2).

SFs are described by defining the dimensionless activation level η:$\eta(0 \le \eta \le 1)$, where $\eta = 1$ corresponds to the maximum possible SF activation level allowed by the biochemistry. The signal induced formation and tension dependent dissociation of the actin cytoskeleton is captured using a first order kinetic equation (Deshpande et al., 2007):