**SUPPLEMENTARY MATERIAL**

# A Multiscale Model to Predict Neuronal Cell Deformation with Varying Extracellular Matrix Stiffness and Topography

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## Supplementary Material: Finite element formulation and analysis

The multiscale mechanisims involved within neuronal cell and their interaction with ECM through molecular receptors has several modeling challenges. As a result, we model the neuronal cell with essential morphologies that are specific to predict neuronal cell mechanosensing behavior (refer to the mechanotransduction in section 2.1). Neuronal cell, dendrite and ECM 3D models are constructed using SOLIDWORKS® (refer to Fig. 2 for the neuronal cell dimensions). Finite element analysis on the neuronal cell and the ECM is analyzed using ANSYS R19.1®. The neuronal cell (nucleus, cytoplasm, and cell membrane) is discretized with 20-node hexahedral solid elements (element type ANSYS: SOLID186) and is able to predict large deformation and viscoelastic material behavior. The Neo-Hookean and Maxwell material model is used to understand the effects of hyperelastic and viscoelastic behavior on the nuclei, cytoplasm, and cell membrane (refer below section for parameters of the material model). Dendrites and ECM are discretized with the 8-node linear hexahedral solid element (SOLID185) and assumed with the linear elastic modulus ($E_{ECM} = 4 - 5000$ Pa, and $E_{dendrite} = 2.5$ KPa). A nonlinear longitudinal spring (COMBIN39) is used to model receptors such as integrin and NCAM, and the necessary

force-deformation relation is derived from the receptor analysis using steered molecular dynamics (SMD). We assign frictionless (friction coefficient, $\mu$=0) element contact behavior between neuronal cell-ECM and dendrite-ECM; since receptors are responsible for the interface contact behavior. Boundary conditions of neuronal cell contractility, ($U_{cell}$=0.3 μm) are applied to the cell membrane by circumferential displacement of outer layer nodes (refer to **Fig.7**). Experimental studies have shown intracellular and intercellular sensing occurs in the range of microscales[1-3]. We have therefore applied contractility of neuronal cell at a rate of 1 $\mu$m/s. We fix the bottom surface of the ECM in all directions to prevent rigid body movement in the model. An implicit analysis for the applied cell contractility in the neuronal cell, dendrite, and ECM was conducted on the multiscale model to solve for stress and displacements. We use the sparse direct solver technique with a force convergence of approximately ±0.5% residual error. FEA procedure is outlined in the flow chart to predict the effect of ECM stiffness and topography on the neuronal cell (refer **Fig. S1**).

A preliminary mesh convergence analysis was conducted to ensure that the element size and number of elements do not impact the results in the multiscale model. The mesh convergence study was performed by refining the element size or number of component elements until von Mises stresses converge $\leq$ 5.0%. For details on the size of the final element and number of elements for each component in neuronal cell and ECM refer to **Table S1**.

**Table S1:** Results of mesh convergence analysis for neuronal cell, dendrite, and ECM.

| Component | ANSYS Element type | Mesh size range | Number of elements |
|---|---|---|---|
| Nucleus | SOLID186 | 0.2 μ$m$ | 100546 |
| Cytoplasm | SOLID186 | 0.2 μ$m$ | 53452 |
| Cell membrane | SOLID186 | 0.2 μ$m$ | 36789 |
| Dendrite (4 no's) | SOLID185 | 0.1 μ$m$ | 113298 |
| Flat ECM | SOLID185 | 0.5 μ$m$ | 100000 |
| ECM asperity with $\phi = 2.5$ μ$m$ | SOLID185 | 0.1 μ$m$ | 105000 |

| | | | |
|---|---|---|---|
| ECM asperity with $\phi = 1.0\ \mu m$ | SOLID185 | 0.1 $\mu m$ | 123000 |
| ECM asperity with $\phi = 0.5\ \mu m$ | SOLID185 | 0.1$\mu m$ | 149761 |

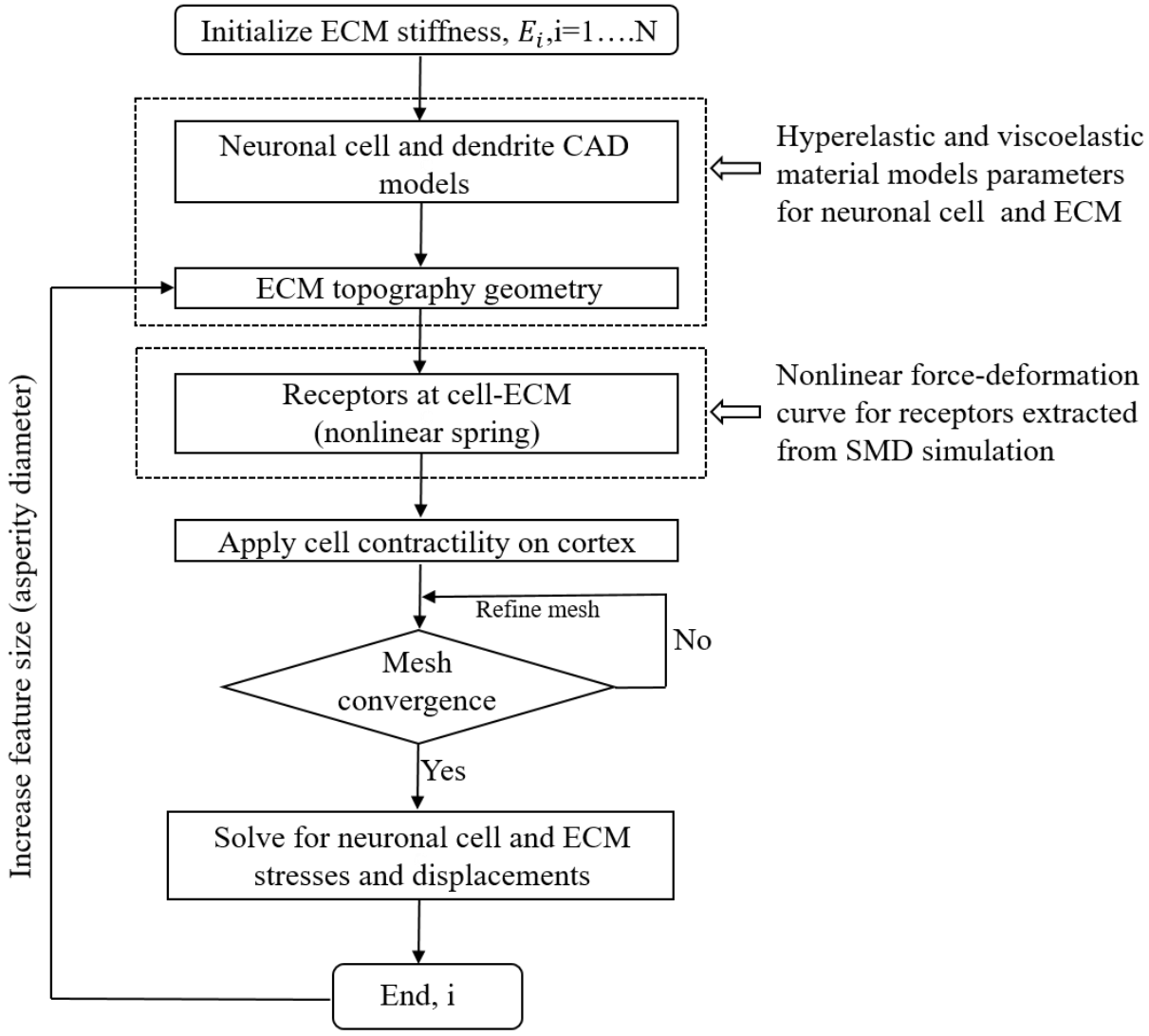


**Fig S1** Flowchart of the FEA methodology used in the multi-scale model to estimate the effect of ECM topography and stiffness on neuronal cell mechanical behavior.

**Supplementary Material: Microscale material properties**

Nanoindentation studies on neurons from rat cerebral cortices provided information on individual mechanical characteristics (force-displacement) of the cell membrane, cytoplasm, and the nucleus. Based on Jerusalem, et al[2]. the material properties of the cell membrane, cytoplasm and nucleus are extracted from neuronal cell indentation results. These experimental results show that cell behavior is nonlinear and viscoelastic. To capture nonlinear characteristics in our computational model and reduce the number of material model parameters, we use a hyperelastic Neo-Hookean model. We implement a Neo-Hookean model since it has shown the ability to predict cell mechanical behavior with a minimal number of material constants[1]. This model is based on the strain energy potential, which is given by:

$$W = \frac{\mu_0}{2}(\bar{I}_1 - 3) + \frac{1}{d}(J-1)^2 \qquad (1)$$

Where,

W = strain energy per unit reference volume

$\bar{I}_1$ = 1st deviatoric strain invariant

$\mu_0$ = long term shear modulus of the material

$d$ = parameter related to the incompressibility of the material

$J$ = determinant of elastic deformation gradient $F$

For predicting the passive viscoelastic behavior of the neuronal cell, we use a generalized Maxwell model. The Maxwell model consists of both Hooke and Newton models and is connected both in series and parallel, as shown in **Fig. S2 a.** However, to minimize the number of constants, we assume a single Maxwell model. This assumption reduces the complexity and considers the viscous effect in neuronal cell components:

$$G(t) = \mu_0 + \mu_1 e^{-\frac{t}{\tau_1}} \quad (2)$$

Where $\mu_1$ is the shear modulus and $\tau_1$ is the relaxation time related to the Maxwell model. The material constants are extracted based on experimental results[3] and are listed in **Table S2**. Instantaneous shear modulus is higher at the microsecond range and decays to a lower shear modulus within the first few seconds to a stable state (refer **Fig. S2 b**). However, the cell membrane rate dependence is negligible, and the shear modulus is approximately constant even at higher timescales. We assume linear elastic material properties for dendrite and ECM due to limitations in the availability of experimental data for rate dependency and hyperelastic behavior.

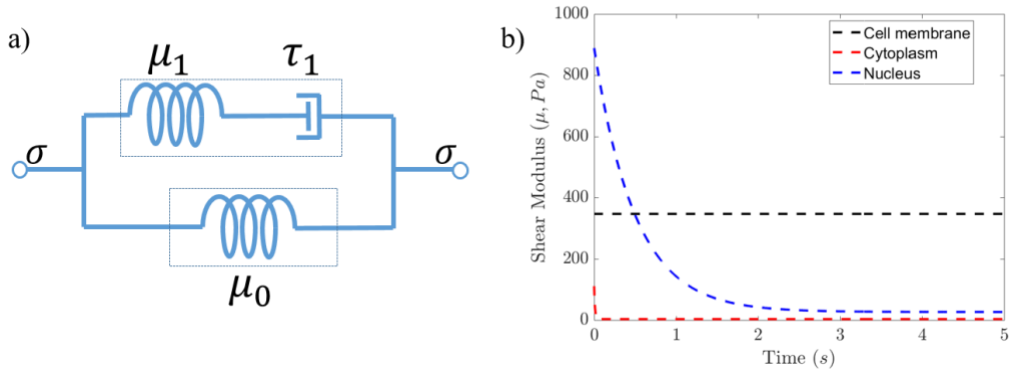


**Fig. S2** a) First order generalized Maxwell model representation for applied stress (σ). b) Shows the rate dependency of the neuronal cell components such as cell membrane, cytoplasm, and nucleus.

**Table S2:** Parameters fitted to the force-displacement results of the neuronal cell experimental data2.

| Material Models | Hyperelastic model (Neo-Hookean) | $c_o^{mem}$ (Pa) | $d_o^{mem}$ ($Pa^{-1}$) | $c_o^{cyto}$ (Pa) | $d_o^{cyto}$ ($Pa^{-1}$) | $c_o^{nuc}$ (Pa) | $d_0^{nuc}$ ($Pa^{-1}$) |
|---|---|---|---|---|---|---|---|
| | | 166.67 | 0 | 13.89 | 0 | 1.67 | 0 |
| | Rate-dependent (Maxwell) | $\mu_1^{mem}$ (Pa) | $\tau_1^{mem}$ (s) | $\mu_1^{cyto}$ (Pa) | $\tau_1^{cyto}$ (s) | $\mu_1^{nucl}$ (Pa) | $\tau_1^{nucl}$ (s) |
| | | 13.89 | 3000 | 107.78 | 0.01 | 862.22 | 0.5 |

**Supplementary Material: Effect of random distribution of receptors on neuronal cell**

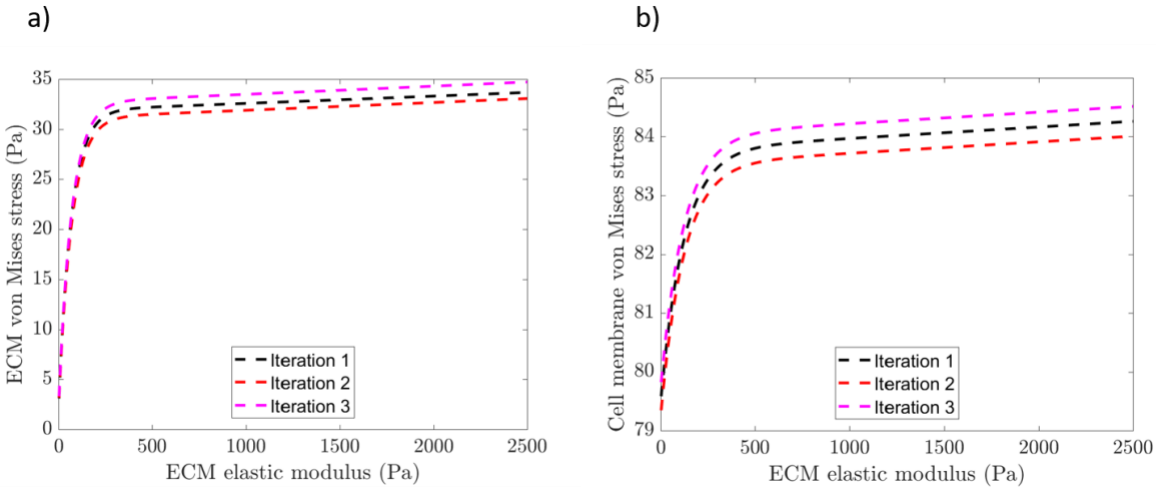


**Fig S3:** Shows the effect of randomness in the distribution of receptors on the von Mises stress of a) ECM and b) cell membrane as a function of ECM stiffness. It indicates that by changing the receptor position von Mises stress varies by ±10 % compared with the base model (iteration 1).

**Supplementary Material: Neuronal cell shape dependence on ECM stiffness and topography**

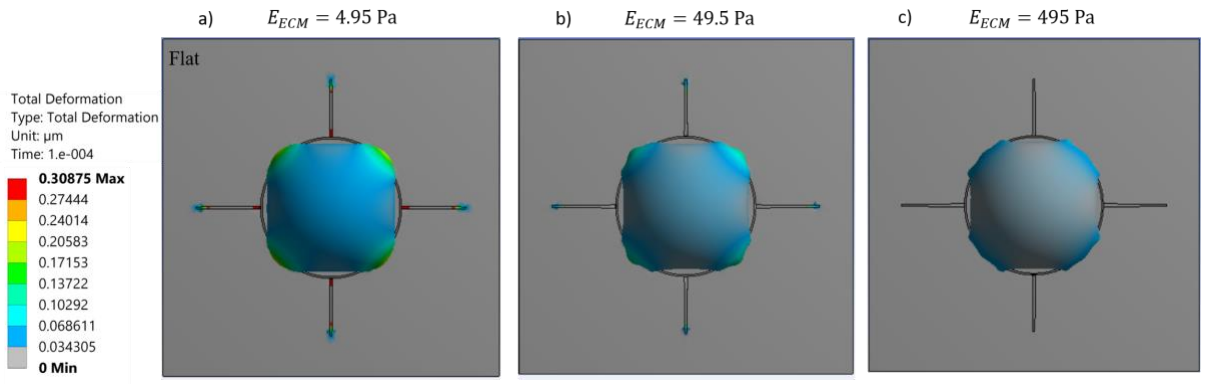


**Figure S4:** Effect of change in ECM stiffness on neuronal cell shape remodeling. Flat ECM shows the symmetric change in cell shape; however, by increasing the $E_{ECM}$ stiffness from 4.95 to 495 Pa (a-c), we observe minor change in cell shape.

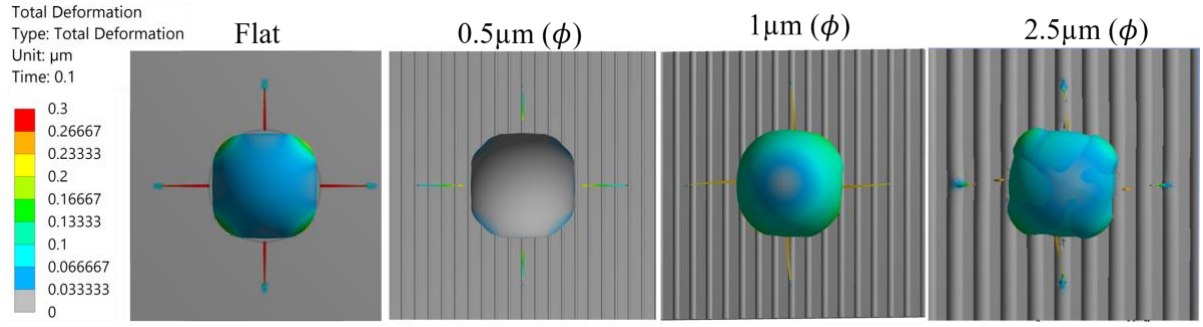


**Figure S5:** Effect of change in ECM topography on neuronal cell shape remodeling. a) Flat ECM shows the symmetric change in cell shape, however by increasing the asperity diameter ($\phi$=0.5, 1 and 2.5 $\mu m$, as shown in b-d), we observe an increase in the asymmetry in cell shape.

**References:**

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3. Zaman, M.H. The role of engineering approaches in analysing cancer invasion and metastasis. *Nat Rev Cancer* 13:596–603, 2013. Available from: https://www.ncbi.nlm.nih.gov/pubmed/23864050.