Mechanotransduction in Stretched Engineered Cardiac Tissue: A Mechanical Bidomain Approach

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Abstract

It has been observed in experiments that when a sheet of cardiac tissue is stretched, cells differentiate favorably on the free edge compared to the interior. A previous analysis has described this behavior using the mechanical bidomain model. This model is a mathematical representation that predicts where mechanotransduction occurs by looking at where forces, acting on integrin proteins, caused by differences in displacements between the intra- and extracellular matrices, are largest. The earlier analysis, however, only considered the mechanical bidomain model using plane strain, which assumes zero displacement in the z-direction. This analysis considers a sheet of tissue being stretched using the mechanical bidomain model under plane stress, which assumes zero stress in the z-direction. The mechanical bidomain model is solved numerically using finite differences. This analysis compares the results of the mechanical bidomain model on a stretched sheet of cardiac tissue found by plane strain and plane stress. The plane strain model predicted differentiation on the tissue’s free edge, similar to previous experiments, whereas plane stress did not predict any differentiation on the free edge. Thus, plane strain seems to be the better model in predicting where cell differentiation occurs in relation to the examined experiment.

Introduction

Mechanotransduction is the process through which mechanical forces, such as stretching, are converted to biological reactions. Mechanotransduction occurs when integrin proteins, connecting the intra- and extracellular spaces, have experienced a force caused by differences in
displacements between the intra- and extracellular spaces (Dabiri, Lee, & Parker, 2012). As a result of mechanotransduction, cells are hypothesized to differentiate and grow (Kresh, & Chopra, 2011). To qualitatively model the effects that mechanical forces have on growing cells, a numerical analysis of the mechanical bidomain model is used (Roth, 2016). Previous models predict stress and strain as causing mechanotransduction. The mechanical bidomain is a two-dimensional model which assumes that differences in displacements between the intra- and extracellular spaces of tissue subject integrin proteins to a force that results in mechanotransduction.

Previously, an experiment performed by Fink et al. (2000) in which they simulated a sheet of cardiac tissue being stretched was qualitatively analyzed by Sharma (2018) using the mechanical bidomain model. He conducted the model under plane strain which assumes that there are no displacements in the z-direction. Our paper performs the same analysis except plane stress is used, which assumes no stresses in the z-direction (Scribner, & Roth, 2017; Fee & Roth, 2019). Thus, the main difference between these two approaches, with the mechanical bidomain model, is that with plane stress the tissue can displace in the z-direction while with plane strain the tissue is limited to displacement in only the x and y directions. Plane stress is useful when examining thin monolayers of tissue because effects in the z-direction can still be modeled while only using a two-dimensional model. In this analysis, the extracellular matrix is stretched parallel to the tissue fibers while the rest of the tissue is free. The purpose of this paper is to simulate tissue stretching with plane stress and compare the results to an analysis performed under plane strain to determine which is a better model in predicting where mechanotransduction occurs.

**Methods**
The mechanical bidomain model defines stress (\( \tau \)) and strain (\( \varepsilon \)) relationships between the intra- and extracellular spaces through the following:

\[
\begin{align*}
\tau_{ixx} &= -p + 2v\varepsilon_{ixx} \\
\tau_{iyy} &= -p + 2v\varepsilon_{iyy} \\
\tau_{izz} &= -p + 2v\varepsilon_{izz} \\
\tau_{ixy} &= 2v\varepsilon_{ixy} \\
\tau_{ixz} &= 2v\varepsilon_{ixz} \\
\tau_{iyz} &= 2v\varepsilon_{iyz}.
\end{align*}
\] (1)

The variable \( v \) is the intracellular shear modulus and \( p \) is the intracellular hydrostatic pressure.

The intracellular strains are associated in terms of intracellular tissue displacement, \( \mathbf{u} \), by

\[
\begin{align*}
\varepsilon_{ixx} &= \frac{\partial u_x}{\partial x} \\
\varepsilon_{iyy} &= \frac{\partial u_y}{\partial y} \\
\varepsilon_{izz} &= \frac{\partial u_z}{\partial z} \\
\varepsilon_{ixy} &= \frac{1}{2}\left(\frac{\partial u_x}{\partial y} + \frac{\partial u_y}{\partial x}\right) \\
\varepsilon_{ixz} &= \frac{1}{2}\left(\frac{\partial u_x}{\partial z} + \frac{\partial u_z}{\partial x}\right) \\
\varepsilon_{iyz} &= \frac{1}{2}\left(\frac{\partial u_y}{\partial z} + \frac{\partial u_z}{\partial y}\right).
\end{align*}
\] (2)

The extracellular matrix is also defined in similar terms,

\[
\begin{align*}
\tau_{exx} &= -q + 2\mu \varepsilon_{exx} \\
\tau_{eyy} &= -q + 2\mu \varepsilon_{eyy} \\
\tau_{ezz} &= -q + 2\mu \varepsilon_{ezz} \\
\tau_{exy} &= 2\mu \varepsilon_{exy} \\
\tau_{exz} &= 2\mu \varepsilon_{exz} \\
\tau_{eyz} &= 2\mu \varepsilon_{eyz}.
\end{align*}
\] (3)

with the extracellular strains defined in terms of the extracellular displacement, \( \mathbf{w} \),

\[
\begin{align*}
\varepsilon_{exx} &= \frac{\partial w_x}{\partial x} \\
\varepsilon_{eyy} &= \frac{\partial w_y}{\partial y} \\
\varepsilon_{ezz} &= \frac{\partial w_z}{\partial z} \\
\varepsilon_{exy} &= \frac{1}{2}\left(\frac{\partial w_x}{\partial y} + \frac{\partial w_y}{\partial x}\right) \\
\varepsilon_{exz} &= \frac{1}{2}\left(\frac{\partial w_x}{\partial z} + \frac{\partial w_z}{\partial x}\right) \\
\varepsilon_{eyz} &= \frac{1}{2}\left(\frac{\partial w_y}{\partial z} + \frac{\partial w_z}{\partial y}\right).
\end{align*}
\] (4)

The variable \( q \) is the extracellular hydrostatic pressure and \( \mu \) is the extracellular shear modulus.

No active tension is present in the intracellular space because only the effects from a mechanical stretch on the tissue are intended to be analyzed. Tissue incompressibility implies that

\[
\varepsilon_{ixx} + \varepsilon_{iyy} + \varepsilon_{izz} = 0
\] (5)
\[ \varepsilon_{exx} + \varepsilon_{eyy} + \varepsilon_{ezz} = 0. \] (6)

Plane stress implies no stress in the z-direction so \( \tau_{izz} = \tau_{ixz} = \tau_{iyz} = \tau_{ezz} = \tau_{exz} = \tau_{eyz} = 0. \)

These conditions were used to solve for the intra- and extracellular hydrostatic pressures, such that, \( p = -2v(\varepsilon_{ixx} + \varepsilon_{iyy}) \) and \( q = -2\mu(\varepsilon_{exx} + \varepsilon_{eyy}) \). Plugging \( p \) and \( q \) into Eqs. 1 and 3, the intra- and extracellular stresses become

\[ \tau_{ixx} = 2v \left[ 2 \frac{\partial u_x}{\partial x} + \frac{\partial u_y}{\partial y} \right] \quad \tau_{iyy} = 2v \left[ 2 \frac{\partial u_y}{\partial y} + \frac{\partial u_x}{\partial x} \right] \quad \tau_{ixy} = v \left[ \frac{\partial u_x}{\partial y} + \frac{\partial u_y}{\partial x} \right] \] (7)

\[ \tau_{exx} = 2\mu \left[ 2 \frac{\partial w_x}{\partial x} + \frac{\partial w_y}{\partial y} \right] \quad \tau_{eyy} = 2\mu \left[ 2 \frac{\partial w_y}{\partial y} + \frac{\partial w_x}{\partial x} \right] \quad \tau_{exy} = \mu \left[ \frac{\partial w_x}{\partial y} + \frac{\partial w_y}{\partial x} \right]. \] (8)

Eqs. 7 and 8 can be differentiated and substituted into the static equilibrium equations

\[ \frac{\partial \tau_{ixx}}{\partial x} + \frac{\partial \tau_{ixy}}{\partial y} = K(u_x - w_x) \] (9)

\[ \frac{\partial \tau_{iyy}}{\partial y} + \frac{\partial \tau_{ixy}}{\partial x} = K(u_y - w_y) \] (10)

\[ \frac{\partial \tau_{exx}}{\partial x} + \frac{\partial \tau_{exy}}{\partial y} = -K(u_x - w_x) \] (11)

\[ \frac{\partial \tau_{eyy}}{\partial y} + \frac{\partial \tau_{exy}}{\partial x} = -K(u_y - w_y) \] (12)

to produce

\[ v \left[ 4 \frac{\partial^2 u_x}{\partial x^2} + 3 \frac{\partial^2 u_y}{\partial x \partial y} + \frac{\partial^2 u_x}{\partial y^2} \right] = K(u_x - w_x) \] (13)

\[ v \left[ 4 \frac{\partial^2 u_y}{\partial y^2} + 3 \frac{\partial^2 u_x}{\partial x \partial y} + \frac{\partial^2 u_y}{\partial x^2} \right] = K(u_y - w_y) \] (14)

\[ \mu \left[ 4 \frac{\partial^2 w_x}{\partial x^2} + 3 \frac{\partial^2 w_y}{\partial x \partial y} + \frac{\partial^2 w_x}{\partial y^2} \right] = -K(u_x - w_x) \] (15)
\[
\mu \left[ 4 \frac{\partial^2 w_y}{\partial y^2} + 3 \frac{\partial^2 w_x}{\partial x \partial y} + \frac{\partial^2 w_y}{\partial x^2} \right] = -K(u_y - w_y). \tag{16}
\]

The variable \(K\) is a Hookean spring constant that accounts for the elasticity of the integrins that connect the intra- and extracellular spaces (Roth, 2016).

Eqs. 13-16 were solved numerically by replacing the derivatives with finite differences and solving iteratively using overrelaxation for \(M\) iterations (Gandhi, & Roth, 2015). To describe the tissue sheet, it was most convenient to model it as a grid of points with \(i=1,2,\ldots,N_x\) in the \(x\)-direction and \(j=1,2,\ldots,N_y\) in the \(y\)-direction. Solving Eqs. 13-16 for the intra- and extracellular displacements in the \(x\) and \(y\) directions, \(u_x(i,j)\), \(u_y(i,j)\), \(w_x(i,j)\), and \(w_y(i,j)\) in terms of their nearest grid points, we obtained:

\[
\begin{align*}
    u_x(i,j) & = \frac{\lambda \omega_x \left( \frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)} + 10 \right) + \theta_x \left( \frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)} + 10 \right)}{\frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)} + 10} - \frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)^2} \\
    w_x(i,j) & = \frac{\lambda \omega_x \left( \frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)} + 10 \right) + \theta_x \left( \frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)} + 10 \right)}{\frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)} + 10} - \frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)^2} \\
    u_y(i,j) & = \frac{\lambda \omega_y \left( \frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)} + 10 \right) + \theta_y \left( \frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)} + 10 \right)}{\frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)} + 10} - \frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)^2} \\
    w_y(i,j) & = \frac{\lambda \omega_y \left( \frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)} + 10 \right) + \theta_y \left( \frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)} + 10 \right)}{\frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)} + 10} - \frac{\Delta^2 \lambda}{\sigma^2(1+\lambda)^2}
\end{align*}
\]

where,

\[
\theta_x = 4[u_x(i+1,j) + u_x(i-1,j)] + \frac{3}{4}[u_y(i+1,j+1) + u_y(i-1,j-1) - u_y(i-1,j+1) - u_y(i+1,j-1)] + u_x(i,j+1) + u_x(i,j-1) \tag{21}
\]
\[ \omega_x = 4[w_x(i + 1, j) + w_x(i - 1, j)] + \frac{3}{4}[w_y(i + 1, j + 1) + w_y(i - 1, j - 1) - w_y(i + 1, j - 1) + w_x(i, j + 1) + w_x(i, j - 1)] \]

\[ \theta_y = 4[u_y(i, j + 1) + u_y(i, j - 1)] + \frac{3}{4}[u_x(i + 1, j + 1) + u_x(i - 1, j - 1) - u_x(i - 1, j + 1) - u_x(i + 1, j - 1)] + u_y(i + 1, j) + u_y(i - 1, j) \]

\[ \omega_y = 4[w_y(i, j + 1) + w_y(i, j - 1)] + \frac{3}{4}[w_x(i + 1, j + 1) + w_x(i - 1, j - 1) - w_x(i - 1, j + 1) - w_x(i + 1, j - 1)] + w_y(i + 1, j) + w_y(i - 1, j) \]

The extra- and intracellular shear moduli are represented as a dimensionless ratio, \( \lambda \), defined as

\[ \lambda = \frac{\mu}{\nu} \]

\( \sigma \) is a length constant characteristic of the bidomain model (Roth, 2016)

\[ \sigma = \sqrt{\frac{\nu \mu}{E(\nu + \mu)}} \]

and \( \Delta \) is the space step between adjacent grid points.

The top and bottom edges of the intra- and the extracellular spaces were free, such that, \( \tau_{iyy} = \tau_{ixy} = \tau_{eyy} = \tau_{exy} = 0 \). Additionally, the stress is zero on the left and right edges of the intracellular space. The stretching was modeled on the extracellular right and left edges by a displacement \( A \) that was modeled by setting \( w_x = \pm A \). The stretching was only modeled in the \( x \)-direction so \( w_y = 0 \). These conditions were used to set the boundary conditions using fictitious nodes.

**Results**
Both the intra- and extracellular spaces experience a displacement greatest throughout the tissue with little to no effect in the center. As the left and right edges are stretched and expand outward, the top and bottom free edges compress inward similar to Sharma’s (2018) results except his model seemed to predict a larger inward displacement from the free edges than ours.

Figure 1: (a) The intracellular displacement, $u$, and (b) the extracellular displacement, $w$. 
The difference in displacement between the intra- and extracellular spaces is localized near the corners and along the left and right edges. The magnitude of $|u - w|$ falls off with length constant $\sigma$. Figure 2a is scaled differently than Figure 1. The magnitude of the membrane force in Fig. 2a is much smaller than the displacements in Fig. 1.
Unlike the magnitude of the membrane force in Fig. 2, the normal strain is largest in the center with the shear strain localized near the corners. The difference in distribution between Fig. 2 and Fig. 3 indicates that the mechanical bidomain model predicts cell differentiation to occur in different locations depending on if strain or membrane force is the cause of mechanotransduction.

**Discussion**

Plane stress predicted similar results as plane strain with a few notable differences. Both models predicted the same distribution of $u$ and $w$, but plane strain seemed to predict a larger displacement inward on the tissue’s free edges (Sharma, 2018). Additionally, Sharma’s (2018)
bidomain displacement was outward along the free edges whereas ours was zero along the free edges (see Fig. 2a). These differences indicate that plane strain predicts a larger and wider distribution of displacements and membrane force along a tissue’s free edge while plane stress predicts a smaller distribution of displacements and membrane force that is strictly along the vertical edges. According to the results, when the tissue was subjected to a mechanical stretch, the tissue expanded in the x-direction, indicated by a positive normal strain, and compressed in the y and z directions, indicated by negative y and z strain. This tissue behavior is similar to what Fink et al. qualitatively observed in their experiments and what Sharma (2018) mathematically described in his research. Looking at Fig. 2, the membrane force is largest at the corners and vertical edges so this is where mechanotransduction would occur according to the mechanical bidomain model under plane stress. Sharma (2018) found that the membrane force is largest at the corners, similar to our results, but he also found a large membrane force along the free edges where we found a membrane force of zero in this region. Our membrane force is also restricted to the vertical edges by a small length constant whereas Sharma’s (2018) membrane force distribution is larger throughout the tissue from the edges and corners. This indicates that plain strain predicts a larger magnitude and distribution of membrane force and individual displacements, especially along the free edge, than plane stress. Thus, when analyzing the tissue stretching performed by Fink et al. (2000), plain strain seems to be a better model than plane stress because plane strain showed a membrane force on the free edges like Fink et al. (2000) where plane stress did not. The reason plane stress did not show a membrane force on the free edges could be because the force that was seen there under plane strain could have been eliminated by the tissue compressing in the z-direction, whereas plane strain could not have had this effect. Our normal strain was largest in the center and our shear strain was largest near the
corners, similar to Sharma’s (2018) strains. If strain was responsible for mechanotransduction then cell differentiation would occur towards the middle of a tissue sample (Fig. 3), but this cannot be accurate in predicting cell growth because Fink et al. (2000) found differentiation localized on the free edges of their sample, not the center.

Engineered heart tissue (EHT) could be used to clinically repair damage to the heart (Zimmermann, Melnychenko, & Eschenhagen, 2004). Patients with ischemic heart disease, cardiomyopathy, or congenital heart diseases are potential targets for EHT therapies (Weinberger, Mannhardt, & Eschenhagen, 2017). EHT has improved functional and morphological properties when subjected to mechanical forces, e.g., stretching (Fink, et al., 2000). This research could be important in understanding how to better grow and engineer heart tissue by providing a mathematical model that can simulate various mechanical conditions on a sheet of tissue with varying physiological states. For example, three dimensional cardiac tissues can be made by stacking cell monolayers on each other (Weinberger, et al. 2017). Our plane stress model (or Sharma’s plane strain model) could then be used to simulate the effects of mechanical forces on the growth of such monolayers used for in vitro or in vivo applications.

This calculation has several limitations:

1. The ratio between the intra- and extracellular shear moduli is modeled as one.

When modeled as less than one, the membrane force was larger on the left and right edges whereas the magnitude of the intracellular strains were smaller. The intra- and extracellular displacements were very similar. When modeled as greater than one, the membrane forces was smaller on the left and right edges whereas the magnitude of the intracellular strains were larger. The intra- and extracellular displacements were very similar.
2. Linear stress-strain and strain-displacement relationships are assumed when nonlinearities may exist.

3. The mechanical bidomain model is limited to two dimensions. A three-dimensional model may be better in analyzing actual tissue especially that with a greater wall thickness.

4. The tissue is modeled as isotropic, but anisotropy could have significant results.

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**References**


