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# VISCOELASTIC MATERIAL PROPERTIES OF THE HAMBURGER-HAMILTON STAGE 12 CHICK HEART

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### INTRODUCTION

Mechanical force is believed to play a significant role in regulating the morphogenetic process of cardiac looping. To better understand this process, it is crucial to determine the material properties of the early chick heart. It is well known that biological tissues are viscoelastic, however previous data on early stage embryonic heart tissue shows a hyperelastic behavior only [1] and currently, only late stage heart tissues have been quantified using viscoelastic properties [2]. The objective of this study is to use microindentation and nonlinear finite element method (FEM) to characterize the viscoelastic material properties of stage 12 chick heart during cardiac looping.

#### **METHODS**

**Microindentation Experiment.** White Leghorn Chicken eggs are incubated at 38°C for two days, when Hamburger-Hamilton (HH) stage twelve is reached [1]. The embryo is extracted and placed into a dish with PBS solution at room temperature. The inner curvature of the heart is fixed by a vacuum micropipette, while the outer curvature is indented by a microforged glass tip attached to a cantilevered glass beam. Indentation is driven by a piezoelectric motor. As the glass tip indents the heart surface, the deflection of the cantilevered beam and the indentation depth are recorded. The deflection of the beam is calibrated to the indentation force.

**FEM Model of Indentation.** The heart is modeled as a bi-layered axisymmetric cylindrical structure comprised of a thin outer-most layer representing the muscle-like myocardium (MY), and a thick inner layer representing the extracellular matrix structure known as cardiac jelly (CJ) [1]. Both MY and CJ are modeled as isotropic, incompressible, viscoelastic materials. To simulate the residual stress within the MY, a uniform stretch is applied ( $\lambda_r = \lambda_{\theta} = \lambda$ ) that satisfies

the incompressibility condition  $(\lambda_r \lambda_0 \lambda_z = 1)$ . Finally, a rigid indenter surface is displaced into the tissue model and is then removed, to simulate indentation of the heart tissues.

**Viscoelastic Constitutive Modeling.** Simo and Hughes' [3] viscoelastic theory is implemented into *ABAQUS* through a user subroutine UMAT [4]. The basic concept is described using a generalized 1D model with an arbitrary number of Maxwell elements arranged in parallel with a spring element. The stress *S* is described by a convolution integral

$$S = \int_{t_0}^t g(t-\tau) \frac{dS^0(\tau)}{d\tau} d\tau, \qquad g(t) = \gamma_\infty + \sum_{i=1}^N \gamma_i \exp(-t/\tau_i), \qquad (1)$$

where,  $S^0$  is the instantaneous second Piola-Kirchoff (2PK) stress, g(t) is the relaxation function with  $\gamma_i$ ,  $\gamma_{\infty}$ , and  $\tau_i$  defining the nodimensional moduli and relaxation times ( $\gamma_i = E_i/E_0$ ,  $\gamma_{\infty} = E_{\infty}/E_0$ , and  $\tau_i = \eta_i/E_i$ ), respectively. Eq. (1) is solved by transforming the convolution integral into a recurrence relationship over the interval  $[t_n, t_{n+1}]$ , requiring the storage of internal variables  $h^i(t_n)$  at each quadrature point, giving

$$h^{i}(t_{n+1}) = \int_{t_{0}}^{t_{n+1}} \exp[-(t-\tau)/\tau_{i}] \frac{dS^{0}(\tau)}{d\tau} d\tau$$

$$= \exp[-\Delta t_{n}/\tau_{i}]h^{i}(t_{n}) + \exp(-\Delta t_{n}/2\tau_{i})(S_{n+1}^{0} - S_{n}^{0}).$$
(2)

The 1D model is then generalized into 3D finite strain elasticity by separating the strain energy into an elastic volumetric response and a viscoelastic deviatoric response. The material is defined by an incompressible exponential strain energy density function [1],

$$W^{0} = \frac{A}{B} \{ \exp[B(\bar{I}_{1} - 3) - 1] \} + \frac{1}{D} (J - 1)^{2},$$
(3)

where  $\overline{I}_1$  is the deviatoric first strain invariant.

The *ABAQUS* UMAT requires the definition of the Cauchy stress and the fourth-order material Jacobian matrix, which represents the derivative of stress increments with respect to the strain increments. The relationship between the viscoelastic Jacobian matrix and the instantaneous Jacobian matrix is given by Eq. (4), where  $C_0$ ,  $C_v$ , and and  $K_0$ , and  $K_v$  are the effective deviatoric elasticity and the bulk modulus for the instantaneous hyperelastic and viscoelastic materials, respectively [4] as

$$C_{\nu} = \left\{ \gamma_{\infty} + \sum_{i=1}^{N} \gamma_i \left[ \frac{1 - \exp(-\Delta \tau / \tau_i)}{\Delta \tau / \tau_i} \right] \right\} C_0 \quad and \quad K_{\nu} = K_0.$$
<sup>(4)</sup>

**Verification of Viscoelastic Modeling.** In order to verify the implementation of the viscoelastic model into UMAT, the theoretical solution of a uniaxial tensile test for quasi-linear viscoelastic (QLV) materials is derived. Consider the deformation obtained by ramping the strain to *a* at  $t_0$ , and holding it at the same strain value for  $t > t_0$  [6]. An expression of stress  $\sigma(t)$  can be obtained using Eq. (1), as

$$\sigma(t) = 6C_1(a/t_0) \{ H(t) \int_0^t g(\tau) [a(t-\tau)/t_0 + 1]^{-4} d\tau - (5)$$
  
$$H(t-t_0) \int_0^{t-t_0} g(\tau) [a(t-\tau)/t_0 + 1]^{-4} d\tau \}.$$

A single linear element undergoing a uniaxial extension test with the same ramped deformation history is analyzed in *ABAQUS* with UMAT and the FEM results are compared to Eq. (5).

**Inverse Computational Method.** The hyperelastic and viscoelastic material parameters of MY ( $A_{MY}$ ,  $B_{MY}$ ,  $\lambda_{MY}$ ,  $\gamma_{1MY}$  and  $\tau_{1MY}$ ) and CJ ( $A_{CJ}$ ,  $B_{CJ}$ ,  $\gamma_{1CJ}$  and  $\tau_{1CJ}$ ) are estimated to match the experimental force-displacement (FD) curve. Only one viscoelastic component is used for this preliminary study. The hyperelastic parameters are varied within the range in of values previously calculated [1], and the viscoelastic parameters are set to satisfy the physical requirements  $0 < \gamma_1 < 1.0$  and  $\tau_1 > 0$ . An approach taken from the design of experiments is used to vary the parameters concurrently [6].

# RESULTS

Good agreement with error less than 5% at the peak stress is found between 2PK stress predicted by QLV theory and FEM for different cases of loading and material parameters (Fig. 1).

The FD curve recorded in the microindentation experiments of the heart tissue clearly exhibits the major characteristic of viscoelastic material properties: hysteresis with energy loss during a loading and unloading cycle (Fig. 2).

The FEM with the parameters shown in (Fig. 2) matches the experimental FD curve with errors in maximal force of 18%, average curvatures of loading and unloading curves 29% and 11% and energy loss of 34%.

# DISCUSSION

Hysteretic force-displacement response is observed during microindentation experiment which clear indicated the viscoelastic material properties of the early chick embryonic heart. The successful implementation of a viscoelastic constitutive relationship in the commercial FEM package *ABAQUS* provides a framework for modeling the custom strain energy density and growth/contraction behavior of the active heart tissues.

The integration of microindentation and FEM modeling presented here provides an effective approach to characterize the viscoelastic material parameters of early chick heart for future study of the mechanics of cardiac looping. However, due to the large amount of unknown parameters, future experiments such as creep and stress relaxation tests, and experiments on the separated MY and CJ tissues are needed to ensure that a unique solution to the material parameters has been achieved.

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Fig 1. Comparison of 2PK stress predicted by QLV theory and FEM.



Fig 2. FD curve for one heart measured in a microindentation experiment and a finite element analysis.