Effects of Specimen Geometry and Boundary Conditions on Fiber Engagement and Mechanical Properties

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INTRODUCTION: The collagen structure of fiber-reinforced tissues allows tissues to withstand large tensile stresses [1, 2]. Early work by Adams *et al.* demonstrated that the linear-region modulus of the annulus fibrosus (AF) that tested along spinal column was highly dependent on the specimen width (58% variation in modulus) [3]. Moreover, the linear-region modulus was significantly larger when the vertebral bodies remain attached (~25 MPa with vertebral bodies vs. < 1 MPa without) [2-6]. We assumed that the wide variability in mechanical properties reported in the literature may be due, in part, to differences in specimen dimensions and applied boundary conditions (B.C.). Therefore, the objective of this study was to evaluate the effects of specimen dimension and grip boundary conditions on fiber engagement in the AF. For this study, we developed a structurally based finite element model with fibers described as a separate material from the extrafibrillar matrix. While the separate model is computationally expensive, we hypothesize that homogenized models neglect the structural differences that are important for accurately describing fiber engagement [7]. We developed the tissue-level models based on the collagen fiber architecture of the outer AF; however, the findings reported here are applicable to any fiber-reinforced material with off-axis fiber orientations.

METHODS: A multi-step approach was used to create the 'separate model' (Sep.), where the fibers and extrafibrillar matrix (EFM) are described as two distinct materials that occupy separate space (~$2x10^6$ tetrahedral elements). The model consisted of 4 lamellae (0.2 mm/lamella) with fibers modeled as cylinders oriented at ± 30º to the transverse plane (Fig. 1A; dimensions: 4.8, 2, and 0.8 mm for length, width, and thickness) [8]. A total of 13 models were developed with a modified dog-bone geometry to ensure mid-length (ML) failure (Fig. 1A) [9]. The model with the geometry of 2 mm x 4.8 mm (width x length; (2, 4.8)) served as the baseline model. The remaining 12 models were categorized into 3 groups: **long**: fixed specimen width (2 mm) with varied length (6.4, 7.2, 8.0, and 9.6 mm), **wide**: fixed specimen length (4.8 mm) with varied width (2.5, 3.0, 3.5, 4.0 mm), and **AR**: fixed specimen aspect ratio (AR = 2.4; widths = 2.5, 3.0, 3.5, and 4.0 mm). Two matching models were developed to identify limitations in the homogenization (Hom.) assumption for numeric analysis of fiber engagement (dimensions: 2 x 4.8 mm & 4 x 9.6 mm (width x length)). To investigate fiber engagement, fibers were categorized into three groups: **gripped** (Fig. 1B - blue), **ungripped** (Fig. 1B - green), and **through ML** (Fig. 1B - pink). The EFM was modeled as a compressible hyperelastic material (Neo-Hookean material) with parameters curve fit to data presented in [11]. Collagen fibers were modeled as a compressible hyperelastic material (Holmes-Mow material with exponential-linear fiber description) with parameters obtained by curve fitting to data presented in [12].

Two boundary conditions were investigated. **Traditional B.C.** represented the common B.C. for uniaxial tensile tests (vertebra attached); the bottom surface was fixed in all degrees of freedom (DoFs) and the top surface was fixed, allowing only along-loading displacement [3-5]. **Compressed B.C.** mimicked the grip setup in multi-lamella tissue-only tests, by applying 10% compressive strain at the grips with fixed DoFs to represent no-slip [2, 6]. A 20% stretch was applied to all models, and fiber elements were considered 'engaged' if the stress exceeded 2.35 MPa [10]. For one group, its fiber engagement was calculated by dividing its engaged element numbers by the model's total engaged elements. Multiple linear regressions with two predictors (length, width) were performed to determine the importance of tissue geometry on tensile mechanics (averaging over orderings metric, R, package "relaimpo") [13].

RESULTS: The bulk tissue mechanics of Hom. models were almost identical, regardless of specimen size and applied B.C. (Fig. 1C - diagonal bars). However, there were large differences between Sep. and matching Hom. models, where the linear-region modulus ($E_{lin.}$) was 29 MPa for the Hom. model and only 6 MPa for the Sep. model (Fig. 1C - solid vs. diagonal bars). The $E_{lin.}$ was higher for tissues modeled with the traditional B.C. rather than the compressed B.C. (Fig. 1D). Increasing either specimen length or width resulted in an increase in $E_{lin.}$ (Fig. 1D). In the baseline model, fiber engagement by gripped and through ML fibers was similar for both B.C.s (Fig. 1E & F - the leftmost bar). Of the three fiber groups, fiber engagement from ungripped fibers was the lowest, but increased with specimen length (Fig. 1E, F - long). Increasing specimen width increased fiber engagement of through ML fibers (> 80%, Fig. 1E, F - wide). Finally, changes in fiber engagement, due to changes in AR, were highly dependent on the applied B.C. (Fig. 1E, F - AR). Changes in specimen length and width significantly influenced AF tensile moduli under both B.C.s ($p < 0.05$). While the influence of specimen length on tensile modulus was stronger for compressed B.C. (length: 84%, width: 16%), its importance was small for traditional B.C. (length: 6%, width: 94%).

DISCUSSION: We investigated the effects of AF specimen geometry and boundary condition on fiber engagement and tensile modulus. Our findings show that homogeneous models are not capable of describing experimentally observed differences in material properties with various specimen dimensions and boundary conditions, justifying the use of a more computationally expensive separate model [2-6]. The higher linear-region modulus with the traditional boundary conditions agrees with differences in the literature between vertebrate-AF-vertebrate and AF-only specimens, emphasizing the importance of the applied boundary condition during testing and model simulations. ASTM standards suggest a 4:1 aspect ratio for uniaxial tension to reduce grip effects; however, specimen dimensions are often limited by tissue availability. Based on our results, specimens with the traditional B.C. were more sensitive to changes in specimen width, while specimens with the compressed B.C. were more sensitive to changes in specimen length. This agrees with the reported experimentally-proposed changes in specimen geometry that aim to increase engagement of fibers that run through the mid-substance (i.e., through ML fibers) [14, 15]. The importance of increasing through ML fiber engagement, compared to gripped fibers, also agrees well with our previous work that showed a notched geometry was important for ML tissue failure [8]. Future work will evaluate the effects of fiber orientation on fiber engagement with a larger model pool. In conclusion, homogeneous models are not sufficient for evaluating fiber-matrix interactions, and tensile mechanics of fiber-reinforced soft tissues are highly sensitive to the boundary conditions, and fiber engagement of the fibers that run continuously through the specimen mid-length.

SIGNIFICANCE: The findings from this study demonstrate sensitivity of bulk tissue mechanics to applied testing boundary conditions, specimen geometry, and relative fiber engagement, which is important for quantifying mechanical properties of fiber-reinforced soft tissues.

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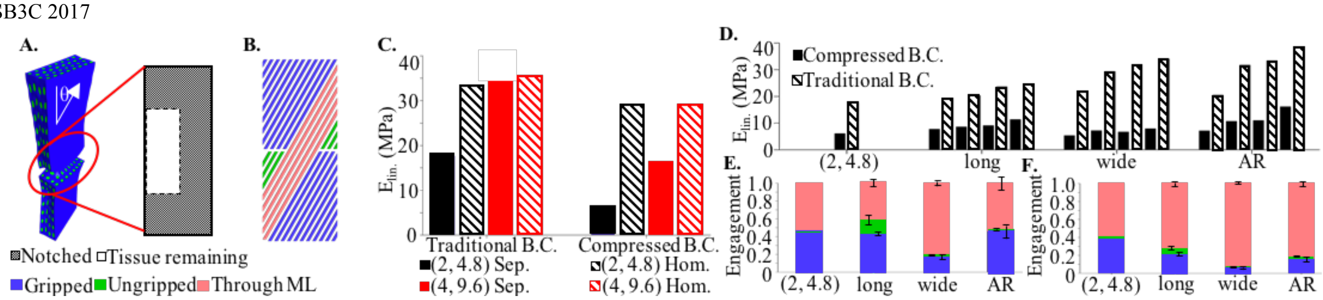

Fig. 1 (A) Schematic of the separate model, with mid-length notch geometry. (B) Three fiber groups (same colors used in E & F). (C) $E_{lin.}$ of Hom. and Sep. models. (D) $E_{lin.}$ for Sep. models (dimensions increase from left to right within each group). Fiber engagement with (E) traditional and (F) compressed B.C.s.