EPITENON CONNECTIVE TISSUE SHEATHS IN TENDON INFLUENCE THE RELAXATION RESPONSE

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Introduction: Predicting and measuring the mechanical response of tendon is important in the development and assessment of various orthopaedic reconstruction techniques. Understanding the mechanical implications of the collagen structures and extracellular matrix in tendon may provide insight into tendon healing and optimal graft choice for Anterior Cruciate Ligament (ACL) reconstruction. At this time, relatively little is known relative to the interaction of the collagen microstructure and interstitial fluid. The collagen microstructure of tendon has been described as being composed of fascicles which in turn are composed of subfascicles (1,2). The subfascicles are the smallest repeating structural element of the tissue and their structure in the patellar tendon has been documented by Yahia and Drouin (2). Atkinson et al. (3) suggested that the collagen subfascicle might be the fundamental structural unit within the tendon. They devised a finite element method (FEM) model of the structure, based on descriptions by Yahia and Drouin (2), where a band of collagen was helically oriented about a central core of matrix. This model suggested that the helical orientation causes the collagen to compress the interfibrillar matrix causing fluid motion and relaxation. The response of the model compared well with whole tendon and ligament responses, however no data was available to describe the mechanical response of the subfascicle itself.

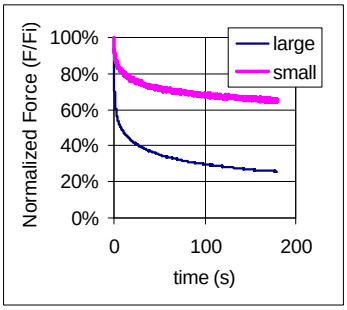
In the current study tendon specimens with both large and small cross sectional areas were tested to provide a comparison between mechanical responses at a subfascicular and whole tendon level. Finite element models of a subfascicle and a fascicle were constructed to examine how the collagen structures and matrix portions of tendon might interact to produce the mechanical responses observed in the experimental study.

Methods: The subfascicle finite element model utilized in the current study was a modification of the previously described subfascicle model (3). Briefly, the model represents Yahia and Drouin's (2) description of a subfascicle in patellar tendon using a representative 3-D section with a 50 μm radius . Helically oriented collagen fibers were wrapped around the periphery of the model and the matrix within the subfascicle was collected in the center. The top and bottom surfaces of the model were sealed (as the subfascicle is a long and thin structure) and the outer boundaries were assumed to be perfectly draining. The bottom plane of the model was constrained to in plane motions with 4 nodes, 90° apart, additionally constrained to radial motion. The top plane was assumed to deform uniformly in the z direction with r and θ free. The matrix portion, in the center of the model, was assumed to be linear isotropic poroelastic material. An orthotropic poroelastic material simulated the helically oriented fibers within the fibrous rings, where the E_2 direction represented the fiber modulus. The properties of the orthotropic material were selected to achieve a nearly incompressible material which was weak in shear. These properties allowed the fiber portion to helically twist in a nearly rigid body fashion. The fiber direction of the orthotropic outer ring was a 20^o declination from vertical, the approximate fiber orientation scaled from SEM images presented by Yahia and Drouin (2) and the crimp angle exhibited in young rat tail tendon (4). Fluid flow was assumed to obey Darcy's law and the permeability was assumed to be constant.

A simple fascicle model was constructed from two subfascicle models. In ligament, fascicles are covered by a connective tissue sheath termed epitenon (5,6). These areolar tissues bind the fasciculi into functionally independent units (5). The collagen in the epitenon is "randomly situated in a coiled manner along the long axis of the fasciculi" (5). In the fascicle model a thin epitenon layer surrounded the subfascicles and was assumed to be perfectly attached to the subfascicles, based on earlier observations of binding fibers between the structures (6,2). The thickness of the epitenon layer (1/12 of the fascicle major axis) was taken from average thicknesses measured in coronal Transmission Electron Micrographs of human anterior cruciate ligament (7). Danylchuk et al. (5) suggested that only the orientation of the collagen fibers in the epitenon distinguished it from the fasciculi. The epitenon was therefore simulated using the collagen fiber material model. Fiber orientations from 0° (horizontal, transverse to the length of the tendon) to -60° from horizontal were investigated in the fascicle model. The relaxation response of the fascicle was compared to that of two subfascicles without epitenon.

The experimental portion of the study involved four pairs of human cadaver knees. The patellar tendons were separated into quarters with bone blocks maintained at each end. One quarter sized specimen was selected from each cadaver to serve as a "large" sized specimen. The contralateral quarter specimen was then subdivided to create two "small" specimens. This protocol helped reduce the influence of spacial variations in tendon. A total of 4 "large" and 8 "small" specimens were tested. The specimens were inspected under a dissecting microscope and damaged portions were removed. At all times the specimens were kept moist with a spray of 0.1M PBS. Specimens were potted in grips using room temperature curing epoxy. The cross sectional area of each specimen was measured at 3 locations using a constant pressure area micrometer. The specimens were equilibrated at least 60 minutes at room temperature in distilled water. This bath was selected to increase the tendon's hydration and thereby enhance the tissue's hydration dependent response. The tissues were mounted for tests in a servo-hydraulic test machine in a vertical orientation. The specimen was immersed in a 37° C distilled water bath and allowed to equilibrate, while slack, for 5 minutes. A small preload was applied (2N for "large", 0.2N for "small") and the specimen alignment was visually verified. A constant strain (2%) relaxation experiment was then conducted. The peak strain was achieved at a cross head displacement rate of 123 mm/s (the maximum displacement rate of the equipment) and was held constant for 180s while force data was gathered at 15 Hz. Immediately following relaxation, the specimen was returned to slack for 2s, then subjected to a subfailure tensile test (peak strain of 5%) at a grip-to-grip strain rate of 1%/s (sample rate 100 Hz).

Results: The cross sectional areas of the larger portions of tendon ranged from 14.5 to 21.7 mm^2, while those of the small portions ranged from 0.1 to 2.6 mm^2. The small specimens relaxed at a rate which was significantly slower than that of the larger specimens. The large specimens relaxed significantly more than the smaller specimens (Figure 1). The small specimens exhibited greater elastic moduli than the large specimens.



The subfascicle FEM model exhibited mechanical responses which were similar to those of the small specimens in the experimental study. In simulated relaxation tests using the fascicle model, the pressure in each subfascicle was positive and continuous for all epitenon fiber directions. The fascicle model indicated that the rate and amount of relaxation increased when the collagen direction in the epitenon was oriented approximately transverse to its axis. At transverse orientations the epitenon tended to push the subfascicles together and increase the amount of twisting in each subfascicle, resulting in higher internal pressures in the model. When the epitenon fibers were declined 5 degrees from horizontal, the rate of relaxation was 18% faster and the model relaxed 39% more than when no epitenon layer was present.

Discussion: The fascicle model suggested that transversely oriented fibers in the epitenon can increase subfascicle deformations, thereby increasing the pressurization of the matrix. This results in an increase in the rate and amount of relaxation. The model described a single, simple fascicle, but it suggests that a transversely oriented epitenon might cause the relaxation response to continue to increase as greater numbers of subfascicles and fascicles are grouped together to represent the large specimen. The influence of the epitenon on relaxation in a large section of tendon therefore appears to be significant, and may explain the marked increase in relaxation observed in the experiments. These results suggest that differential responses may result when grafts of various cross sectional areas and varied microstructure are considered.

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