

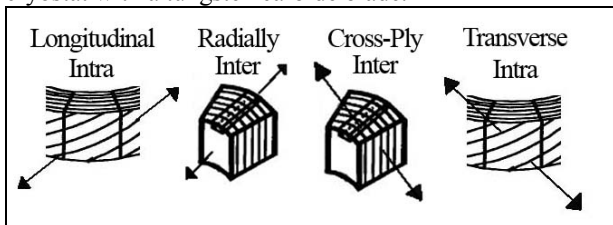
## Micromechanical characterization of Inter- and Intra-Lamellar Annulus Fibrosus Specimens

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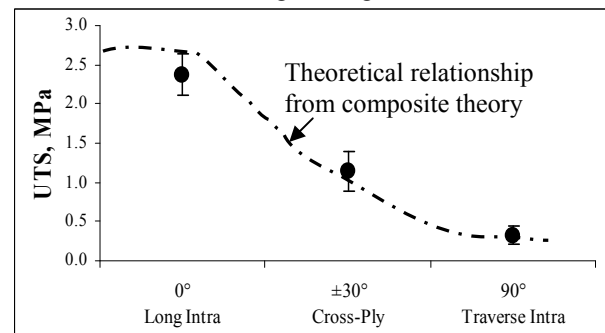
**Statement of Purpose:** Intervertebral discs (IVDs) stabilize and maintain alignment of the spine, allow movement between vertebrae and are responsible for load distribution and energy absorption under loading. IVDs are composed of three distinct parts: cartilaginous endplate, annulus fibrosus (AF), and nucleus pulposus (NP). The AF is composed of 25 unidirectional collagen fiber lamellae. Consecutive lamellae run in opposite directions with collagen fibers at an angle of 60° to the long axis of the spine. Collagen fibers may also bind the lamellae together as elastin fibers pass radially from one lamella to the next [1]. Elastic and collagenous elements are embedded in a hydrated glycosaminoglycan matrix. At lamellar interfaces, the arrangement of elastic fibers forms discrete connections between collagen bundles in consecutive lamellae. There is also evidence that elastic fibers form cross-bridges to connect collagen bundles in consecutive lamellae [2]. To understand the orientation attributions of AF mechanics, we have previously [3] developed a single lamellar model under different conditions to test annulus lamellar biomechanics in plane to the collagen and elastin fiber directions using a micro-mechanical test protocol. In this work, using the same technique, we investigate the degenerative mechanics that occur inter and intra-lamellar both in and out of the plane of the fibers.

**Methods:** Whole IVDs were removed at the endplates using a scalpel. Discs were hemi-sectioned mid-sagittally and intra-lamellar (150 μm thick) human lumbar annulus fibrosus (AF) samples were cut using a Leica 3050S cryostat with a tungsten carbide blade.

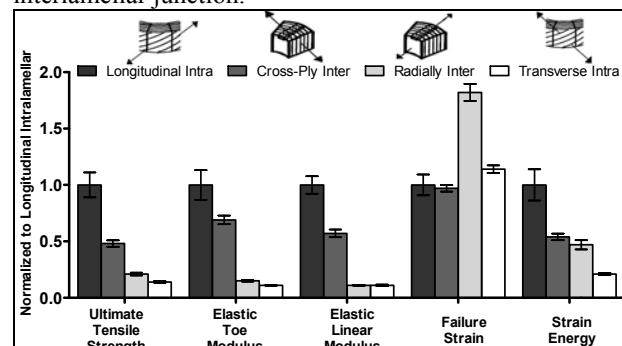


Samples were pre-strained to  $\epsilon = 0.05$  then with an average strain rate of 0.02 mm/sec were tested until failure. Micromechanical stress-strain curves were compiled and concurrent video streams captured morphological damage mechanics. Engineering stress (force divided by undeformed cross-sectional area at equilibrium hydration) vs. strain (the instantaneous length divided by the starting length) was plotted for each specimen. Initial modulus (tangent to the toe region of the response), ultimate modulus (tangent to the linear region of the response) and failure strain were calculated from the raw data.

**Results:** The cross-ply calculations follow the trend of fiber reinforced composites, and make a good approximation to off angle testing.



Data (UTS,  $\epsilon_f$ , E) for the in plane intralamellar samples were in the range from literature. Key findings showed reduction of properties between 0°, 90°, radial cross-ply, and between layers. Properties were reduced compared to longitudinal intralamellar except for failure strain. Failure strain across layers doubled, likely due to elastin at the interlamellar junction.



**Conclusions:** Elastin dominated interlamellar strain was double that of in ply (0° intralamellar). Ultimate tensile strength decreases intralamellarly with fiber angle (0° to 90°), with cross-ply properties falling in the mid point. A limitation is that AF mechanical properties in the radial orientation have been shown to be radially heterogeneous [4]. The fact that radial position was not strictly controlled may have therefore contributed to the variances. The properties described here provide insights into the intra- and inter-lamellar mechanical behavior of the AF. This may be important in diagnosis, prevention and repair of debilitating IVD disorders and manufacturing of tissue-engineered AF.

**References:** [1] Yu, J; CP Winlove; S Roberts; JP Urban. *J Anat.* 2002; 201:465-475. [2] Yu, J; U Tirlapur; J Fairbank; P Handford; S Roberts; CP Winlove; Z Cui; JP Urban. *J Anat.* 210:460-471, 2007. [3] Isaacs, J.L.; E Vresilovic; M Marcolongo. ORS Poster 2010. [4] Fujita, Y; NA Duncan; JC Lotz. *J Orth Res.* 15:814-819, 1997.

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