Visualization of collagen crimp in tendon using high-field MRI

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Tendon exhibits periodic banding especially with polarized light illumination [1,2]. Crimp refers to a periodic molecular arrangement of collagen fibers that, to date, have only been observed using electron and polarized light microscopy. T_2 MRI is sensitive to collagen fiber orientation [3]. We performed high-field MRI on a tendon to determine if parameters could be identified that would allow for visualization of crimp. Validation of crimp was done using polarized light microscopy.

Methods: A digital extensor tendon was obtained from a canine euthanized for other experiments. The tendon was bent in the bottom of a 10 mm (i.d.) round bottom test tube in PBS. The specimen was placed along B_0 in a 9.4T Magnex magnet for imaging with a Bruker Avance console using spin-echo MRI (TE/TR= 9/3000 ms, matrix 256×256, FOV=15×15mm voxel = 60x60x1000μm). After MRI, the tendon was fixed in formalin, and a 1-mm-thick strip of tendon was mounted on a glass slide in PBS. The tendon was placed between a circular polarizer and a liquid crystal analyser (CRI, Boston). This system measures the intensity and orientation of collagen birefringence as retardance (nm) and azimuth (degrees and color wedge).

Results: MR revealed periodic banding throughout the tendon (arrows), particularly in areas not parallel to B_0 (Fig. 1B). T_2 relaxation was rapid along the axis of the tendon when aligned with B_0, and slower when aligned away from B_0, reaching a maximum near 55° (Fig. 1A). T_2 intensity in the rectangle (Fig. 1A) is plotted in Fig 1E (top). Tendon collagen was highly birefringent along its axis, with a local zigzag characteristic of collagen crimp. The retardance and azimuth for the rectangle (Fig. 1C) are plotted in Fig 1E.

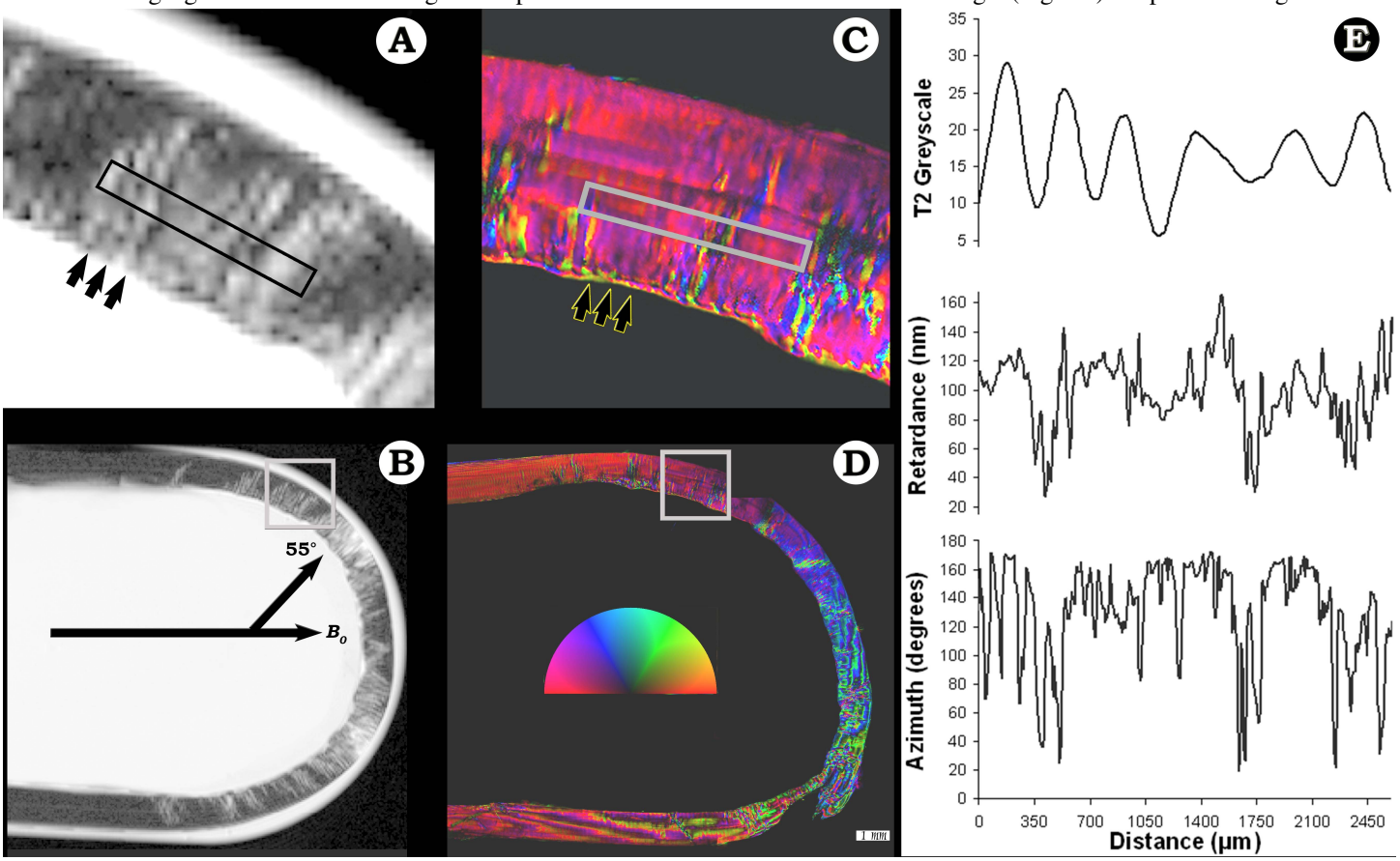

Figure 1: Collagen crimp MRI (A,B), Polarized light microscopy (C,D). A and C correspond to squares in B and D. Rectangles in A and C show ROI's for data in line graphs (E). Arrows point to "crimp" bands. B and D show whole sample (1mm scale bar in D). Long arrows in B show B_0 and 55° (tendon lightens due to magic angle effects on T_2). Semicircle in D is a map of birefringence angle.

Discussion: The similarity of banding on both MRI and polarized images implies that high field MR using T_2 weighted imaging can be used to visualize collagen crimp in unloaded tendon, though care must be used to interpret signal intensity as a function of orientation with respect to B_0. High-resolution T_2 MRI demonstrates the molecular arrangement of collagen, suggesting that MRI might be used as an in situ strain gauge to assess the functional state of the tendon as crimp is seen under low loads, yet disappears under high loads [4,5].

References: [1]Rigby BJ et al. *J.Gen.Physiol.* 1959;43:265-83. [2] Viidik A, Ekholm R . *Z Anat Entw.* 1968;127:154–164. [3] Goodwin DW et al., 1998. *Acad. Radiol.* 5:790-798. [4] Kastelic et al. *J. Biomech*: 1980;13:887-893 [5] Diament J et al. *Proc. R. Soc. Lond. B.* 1972;180:293-315.