

Dipolar Anisotropy Reveals Sub-structures in Achilles Tendon at 11.7 Tesla

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Introduction

The NMR behavior of water imbedded in collagen was first investigated fifty years ago in bovine Achilles tendon and described in a seminal paper by Berendsen (1). NMR spectra were obtained at various degrees of hydration and at multiple fiber to field orientations. At certain hydration levels resolvable broad lines were observed with a splitting that depended on the fiber to static magnetic field orientation, providing direct evidence for residual proton-proton dipolar coupling. These residual dipolar effects influence T2 relaxation times and are responsible for the magic angle effects known in MR imaging. In this study we manipulated the orientation of a human Achilles specimen in an 11.7 T system producing high contrast images which at certain orientations revealed fine sub-structures that were not otherwise observed.

Methods

A section of fresh (frozen once) human Achilles tendon was trimmed to a length of 14 mm and placed in a 10 ml plastic syringe, oriented with the specimen fiber axis perpendicular to the syringe barrel. The syringe was filled with perfluoro polyether (Fomblin) to reduce the specimen's bulk susceptibility effects. 3D FLASH images were obtained at 11.7 Tesla using a Bruker BioSpec 117/16USR system with parameters: TR=10, TE=2.8, FA=6°, voxel size= 50x100x100 μm^3 , bandwidth=250 hz/pixel, and NEX=2. A custom 18 mm diameter solenoid transmit/receive coil was used to optimize SNR. The fiber to B_0 field direction was varied in 30° increments by rotating the syringe barrel. Images were registered using FLIRT software (FSL, Oxford) and intensity fluctuations (coefficient of variation, CV) images were computed using macros written in ImageJ.

Results

High B_0 field strength and a small coil with good filling factor produced detailed images having good SNR (~40) in short imaging times (<13 min). There was a surprising amount of structure observed with high contrast on these images, especially at particular fiber to field orientations. Fig. 1 shows a slice from the 3D datasets as the orientation of the sample was rotated relative to the B_0 field. The FLIRT registration performed adequately in spite of the large differences in intensity between data sets. Signal intensity of the most fibrous tissue voxels (diamond symbols on Fig. 2) increased by a factor of 17 as the specimen was rotated from 0° (parallel to the field) to 60° (close to the magic angle). A much smaller effect was observed in endotenon (squares) and loose connective tissue on the distal end of the specimen (triangles). Fig. 3 is a coefficient of variation image which was computed from the first six co-registered datasets. The ROI boxes in Fig. 3 indicate the locations from where the intensity values were extracted to generate the plots seen in Fig. 2. High CV values on Fig. 3 arise from voxels having large intensity variations with sample rotation (B_0 orientations).

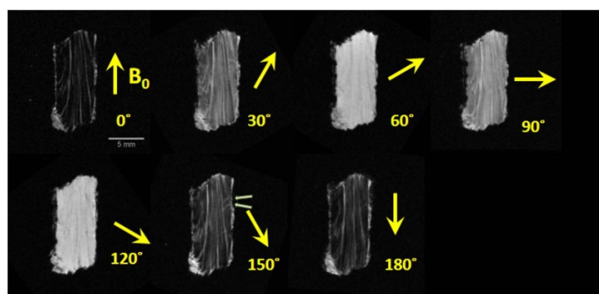


Figure 1. Achilles tendon images with various specimen to field orientations. The solid arrows indicate the B_0 direction. The open arrows on the 150° panel point to transverse structures which show up with high contrast at this orientation.

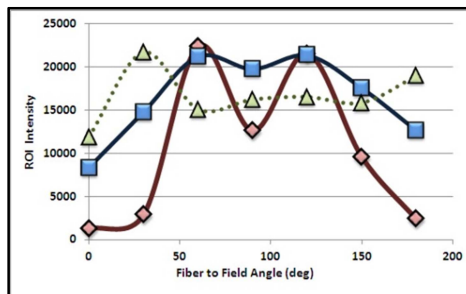


Figure 2. Plot of signal intensity from 3 ROIs as a function of specimen to field angle. Diamonds are fibrous tissue, squares are endotenon, and triangles are connective tissue.

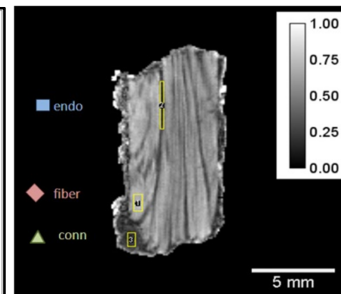


Figure 3. A coefficient of variation image (CV). High values indicate large signal intensity changes with orientation. The small boxes indicate the various ROIs used in Fig. 2.

Discussion

Through analysis of signal intensity as the sample to field direction is changed, it is possible to extract fiber structure information, i.e. direction and anisotropy values (2,3). This dipolar anisotropy fiber imaging (DAFI) approach can be abbreviated when one has prior knowledge of the structures making up the tissue of interest. A few select orientations of a tissue with respect to the magnetic field in some cases can produce intense contrast between structures that are otherwise difficult to identify. This approach has been applied to human Achilles tendon in this study to allow visualization of fine structures that otherwise are not readily seen. The fine oblique transverse structures seen in Fig. 1 (open arrows, 150° orientation) have been demonstrated on histology and polarized light microscopy, but have not previously been characterized using MRI.

References

1. Berendsen HJC. Nuclear magnetic resonance study of collagen hydration. *J. Chem. Phys.* 1962;36(12):3297-3305.
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