Effects of Repetitive Freeze-Thawing Cycles on Quantitative Magnetic Resonance Imaging of the Achilles Tendon

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Introduction: Magnetic resonance (MR) imaging allows quantitative characterization of whole tissues comparable to histological methods. However, cadaveric studies often involve tissues that undergo multiple freeze-thaw cycles. While the effects of this on material properties of tendon have been studied, there remain mixed conclusions as some authors have shown detrimental effects[1,2], while others have found little to no change[3,4]. Additionally, quantitative MR studies have been performed on fish meat[5] and equine soft tissues[6] after freeze-thaw cycles, but to our knowledge none have been performed on human tendons. The goal of this pilot study was to evaluate these changes.

Methods: Four legs from three donors (3 females, mean age 78 years old) were harvested below the knee within 8 hours of death. The Achilles tendon was dissected free from surrounding tissues in one specimen and left intact within the ankle in the remaining three. Tendons were placed parallel to the B0 field and imaged on a clinical 3T MR system (parameters as shown in Table 1). Axial MR images were performed at the same location within the tensile portion of the tendon in the fresh specimens and after 1, 2, 4, and 5 freeze-thaw cycles. Each cycle consisted of a 24-36 hour freeze period at -80°C followed by a 20-24 hour thaw period at room temperature. Specimens were wrapped in moist gauze during freezing to prevent dehydration. Regions of interest were manually drawn over the entire Achilles tendon and mono-exponential fitting was performed. Magnetization transfer ratio was calculated (Baseline signal-Signal after MT pulse/Baseline signal). A mixed-effects linear regression model was fit for each parameter as a function of freeze-thaw cycle with a random (subject-specific) intercept. Overall slope of parameters over cycles was estimated and adjusted for within-specimen dependence. P-values <0.05 were deemed significant.

Results: Quantitative measures are shown in Table 2. Spiral chopped magnetization prepared T1rho measurements could not be accurately fitted to a curve and therefore values could not be calculated with confidence. There was no significant trend of quantitative measures between fresh and subsequent freeze-thaw cycles.

Conclusions: In this pilot study, we have not found a significant difference in quantitative MR values on Achilles tendons imaged fresh and up to 5 freeze-thaw cycles. Ongoing work is being performed to validate these preliminary results utilizing larger sample sizes.

Table 1

Sequence	TR [ms]	TE [ms]	FOV [cm]	Matrix
T1	10,20,50,100,200,400,800,1200	0.01	6	192x192
T2	2000	10,20,30,40,50,60,70,80	6	320x256
UTE T2*	100	0.01,0.1,0.2,0.4,0.6,0.8,2,4,10,15,20,30	6	256x256
T1 rho	1500	10 (spin lock time 0.02,1,5,10)	6	320x256
UTE T1 rho	400	0.01 (spin lock time 0.02,1,5,10)	6	192x192
MT_0	300	0.01 without sat pulse	6	256x256
MT	300	0.01 after 3kHz off-resonance sat pulse	6	256x256

Table 2

	Fresh mean (SD)	Post 1 cycle mean (SD)	Post 2 cycles mean (SD)	Post 4 cycles mean (SD)	Post 5 cycles mean (SD)
T1	535.49 (98.75)	518.84 (74.41)	559.64 (75.66)	585.58 (45.68)	548.73 (75.34)
T2	17.23 (6.49)	15.09 (4.42)	16.98 (4.13)	19.00 (3.43)	17.09 (1.93)
UTE T2*	1.18 (0.45)	1.05 (0.30)	1.26 (0.44)	1.20 (0.34)	1.11 (0.37)
UTE T1rho	2.34 (0.87)	1.87 (0.67)	2.51 (0.90)	2.93 (0.89)	2.51 (0.73)
MTR	0.17 (0.02)	0.13 (0.02)	0.13 (0.01)	0.14 (0.01)	0.16 (0.03)

References:

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