Validation of Quantitative MRI Measures of Intervertebral Disc Health in Client-Owned Dogs

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INTRODUCTION: Intervertebral disc degeneration (IVDD) is a leading cause of low back pain. There is a need for clinically relevant animal models with a pathogenesis similar to human IVDD to better understand the disease and test new treatments [1]. While most animal models of IVDD artificially induce disc injury (e.g., by puncturing the disc), a model of spontaneous disease would better reflect the natural course of IVDD [1]. Dogs are one of the few species to develop spontaneous IVDD with a pathogenesis similar to human IVDD [2-4]. Client-owned companion dogs that are being imaged and treated for IVDD provide a unique population within which to study naturally occurring IVDD and trial new treatments. However, the given limited access to tissue samples in client-owned dogs, noninvasive imaging tools are needed that can characterize disc health, monitor IVDD progression, and evaluate the efficacy of treatments *in vivo*. Thus, the purpose of this study was to assess quantitative magnetic resonance imaging (qMRI) measures of disc health (relaxation time mapping and diffusion imaging) in vertebral columns obtained from client-owned dogs. We hypothesized that randomly selected dogs without clinical spine disease have a range of disc degeneration severity and that the qMRI measures correlate with radiological, histological, and biochemical measures of disc health. This study expands on our preliminary report of 5 dogs [5] to include more qMRI measures and analyses across 17 dogs.

METHODS: ***Animals:*** Thoracolumbar vertebral column specimens were collected from 17 client-owned dogs whose bodies were donated for research following clinical euthanasia. The dogs had a variety of signalments: 15 were nonchondrodystrophic breeds, 1 was a chondrodystrophic breed, and 1 was a mixed breed; 7 were female and 10 were male; ages ranged from 5.0 to 15.5 years (avg=11.1); and weight ranged from 8 to 46 kg (avg=26.4). No dogs had a known history of IVDD or other spinal disease. ***Magnetic Resonance Imaging:*** The fresh vertebral columns were imaged at 3T MRI to collect data on 10 discs (T11/T12 to L7/S1). T2-weighted TSE images in the sagittal and coronal planes were acquired for Pfirrmann grading. Seven qMRI measures ($T2_{MSME}$, T2*, T2, T1ρ, aT1ρ, aT2ρ, and ADC) were acquired axially for each of the 10 discs: $T2_{MSME}$ mapping using a multi-slice multi-echo (MSME) spin echo sequence; T2* mapping using a MSME gradient echo sequence; T2, T1ρ, aT1ρ, and aT2ρ mapping using a magnetization-prepared TSE sequence; and ADC mapping using a readout-segmented diffusion-weighted imaging (RESOLVE) sequence. The specimens were frozen immediately after the MRI exam. ***Histology:*** The frozen vertebral columns specimens were bisected along the sagittal midline of the spine using a bandsaw. One half of each bisected vertebral segment was fixed in 10% NBF, decalcified in 10% EDTA, and then routinely processed for H&E and alcian blue / picrosirius red (AB/PSR) staining. ***Biochemical Assays:*** The other half of each bisected vertebral segment was used to collect tissue samples of the nucleus pulposus (NP) and ventral annulus fibrosus (vAF). The samples were weighed before and after dehydration to calculate percent water weight (%water), and then glycosaminoglycan concentration ([GAG]) was measured using an s-GAG assay kit. ***Data Analysis:*** The 7 qMRI maps were generated using a mono-exponential signal decay model implemented in MATLAB. T2-weighted axial disc images were manually segmented to define NP and vAF regions of interest (ROIs). The median qMRI measures within each ROI were calculated. The radiological Pfirrmann grade of each disc was assessed by a board-certified veterinary radiologist [6]. The histology score of each disc was determined by a veterinary pathology resident according to the Bergknut scheme for canine discs [7]. The relationships between the 7 qMRI measures and dog, disc level, Pfirrmann grade, histology score, %water, and [GAG] were then assessed using R statistical software.

RESULTS: Most dogs had both healthy and degenerated discs as assessed by Pfirrmann grade and histology score (Figure 1). NP and vAF qMRI values in the healthy discs (Pfirrmann grade ≤ 2) varied greatly across the dogs but were similar across disc levels (Figure 2). The strength of the relationships (conditional R) between each pair of qMRI and disc health measures are shown in Figure 3 using a linear mixed effects model (lme4) with dog as a random effect. In the NP, $T2_{MSME}$, T2, T1ρ, aT1ρ, and aT2ρ were all strongly correlated, whereas T2* and ADC provided more unique responses. Similar relationships between the qMRI measures were seen in the vAF. In the NP, the qMRI values were negatively correlated with Pfirrmann grade and histology score and positively correlated with %water and [GAG]. In the vAF, the qMRI measures were generally positively correlated with Pfirrmann grade, histology score, and %water. There were no clear differences between the sensitivities of the relaxation times to the various measures of disc health.

DISCUSSION: Our study revealed several key findings. First, most elderly dogs have a range of disc health, with both healthy and degenerated discs. While IVDD may be more prevalent at some disc levels (e.g., L7/S1), we observed disc degeneration across the thoracolumbar spine (T11/T12 to L7/S1). Second, the qMRI values in the NP and vAF of healthy discs vary greatly between dogs (potentially due to breed differences), and thus qMRI values would be best compared across discs within a given dog and/or tracked longitudinally. Third, the qMRI measures are strongly related to disc health measures in the same way expected from human qMRI studies [8]: the qMRI values decrease in the NP as it dehydrates, loses [GAG], and becomes more collagenous and increase in the AF as it loses its structural integrity. Lastly, the 7 studied qMRI measures had similar sensitivities to the disc health measures in the NP and (with the exception of T2* and ADC) in the AF. The relatively decreased sensitivity of T2* and ADC in the AF may be due in part to their low signal in the AF.

SIGNIFICANCE/CLINICAL RELEVANCE: Client-owned dogs have spontaneous disc degeneration that can be detected using quantitative MRI techniques. qMRI methods may be particularly useful to quantify subtle and early changes in disc health in longitudinal and treatment efficacy studies.

REFERENCES: [1] Lee NN, et al. JOR Spine 2021; 4(2):e1162. **[2]** Lee NN, et al. JOR Spine 2020; JOR Spine 2020; 3(4):e1109. **[3]** Bergknut N, et al. Spine 2012; 37(5):351-8. **[4]** Bach FC, et al. BMC Vet Res 2014; 10:3. **[5]** Johnson CP, et al. Proc ORS 2023; No. 0256. **[6]** Bergknut N, et al. Am J Vet Res 2011; 72(7):893-8. **[7]** Bergknut N, et al. Vet J 2013; 195(2):156-63. **[8]** Russo F, et al. Spine J 2023; S1529-9430(23)00214-0.

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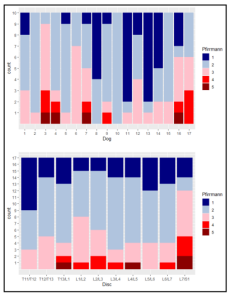

Figure 1. Distribution of Pfirrmann grades across dogs and disc levels.

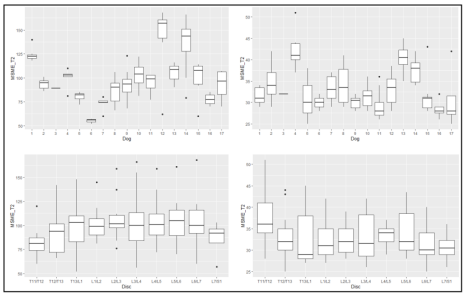

Figure 2. Average NP (left) and vAF (right) $T2_{MSME}$ relaxation times for relatively healthy discs (Pfirrmann ≤ 2) across dogs and disc levels.

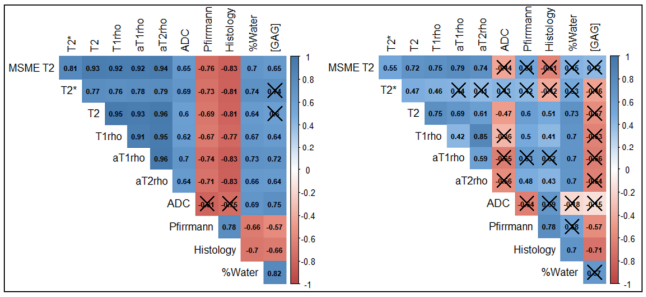

Figure 3. Correlation matrices of the quantitative MRI and disc health measures in the NP (left) and vAF (right) using linear mixed effects models with dog as a random effect. X = non-significant (p≥0.05).