Published in final edited form as:
Spine (Phila Pa 1976). 2006 May 15; 31(11): 1253–1257. doi:10.1097/01.brs.0000217708.54880.51.

Assessment of Human Disc Degeneration and Proteoglycan Content Using $T_{1\rho}$-weighted Magnetic Resonance Imaging

Wade Johannessen, BS[*], **Joshua D. Auerbach, MD**[*], **Andrew J. Wheaton, PhD**[†], **Alykhan Kurji, BS**[*], **Arijitt Borthakur, PhD**[†], **Ravinder Reddy, PhD**[†], and **Dawn M. Elliott, PhD**[*]

[*]Department of Orthopaedic Surgery, McKay Orthopaedic Research Laboratory, University of Pennsylvania, Philadelphia PA.

[†]Department of Radiology, University of Pennsylvania, Philadelphia PA.

Abstract

Study Design—$T_{1\rho}$ relaxation was quantified and correlated with intervertebral disc degeneration and proteoglycan content in cadaveric human lumbar spine tissue.

Objective—To show the use of $T_{1\rho}$-weighted magnetic resonance imaging (MRI) for the assessment of degeneration and proteoglycan content in the human intervertebral disc.

Summary of Background Data—Loss of proteoglycan in the nucleus pulposus occurs during early degeneration. Conventional MRI techniques cannot detect these early changes in the extracellular matrix content of the disc. $T_{1\rho}$ MRI is sensitive to changes in proteoglycan content of articular cartilage and may, therefore, be sensitive to proteoglycan content in the intervertebral disc.

Methods—Intact human cadaveric lumbar spines were imaged on a clinical MR scanner. Average $T_{1\rho}$ in the nucleus pulposus was calculated from quantitative $T_{1\rho}$ maps. After MRI, the spines were dissected, and proteoglycan content of the nucleus pulposus was measured. Finally, the stage of degeneration was graded using conventional T_2 images.

Results—$T_{1\rho}$ decreased linearly with increasing degeneration ($r=-0.76$, $P < 0.01$) and age ($r=-0.76$, $P < 0.01$). Biochemical analysis revealed a strong linear correlation between $T_{1\rho}$ and sulfated-glycosaminoglycan content. $T_{1\rho}$ was moderately correlated with water content.

Conclusions—Results from this study suggest that $T_{1\rho}$ may provide a tool for the diagnosis of early degenerative changes in the disc. $T_{1\rho}$-weighted MRI is a noninvasive technique that may provide higher dynamic range than T_2 and does not require a high static field or exogenous contrast agents.

Keywords

disc degeneration; quantitative magnetic resonance imaging; $T_{1\rho}$; matrix; proteoglycan

Degenerative disc disease afflicts nearly 12 million people in the United States. Although a single initiating cause of degeneration has not been identified, early degenerative changes occur in the nucleus pulposus.[1,2] Breakdown of the large aggregating proteoglycans reduces the capacity of the nucleus pulposus to attract and bind water, leading to a loss of disc

Address correspondence and reprint requests to Dawn M. Elliott, PhD, University of Pennsylvania School of Medicine, Department of Orthopaedic Surgery, McKay Orthopaedic Research Laboratory, 424 Stemmler Hall, Philadelphia, PA 19104-6081; delliott@mail.med.upenn.edu.

hydration and decreased hydrostatic pressure.[3,4] Ultimately, degeneration progresses to decreased disc height, structural changes in the lamellar architecture of the anulus fibrosus, anular tears and rim lesions, and the formation of osteophytes.[5-7] The success of treatment strategies aimed at halting the progression of disc degeneration will require detection of the early stages of the disease, particularly changes in the extracellular matrix content of the nucleus pulposus.

Conventional magnetic resonance imaging (MRI) techniques provide excellent detection of late-stage degenerative changes (*i.e*., changes in disc morphology, height, hydration, bulge, and herniation).[8,9] However, these methods are not sensitive to early degenerative changes in the matrix content of the disc.[10] Delayed gadolinium-enhanced MRI has been used to quantify proteoglycan in articular cartilage.[11,12] However, the contrast agent must be administered intravenously, and diffusion into the cartilage requires a long time.[13] This is a significant limitation in the avascular intervertebral disc, in which the negative fixed charged density of the nucleus pulposus hinders diffusion of ionic contrast agents.[14,15] Sodium MRI has also been used to measure proteoglycan in articular cartilage.[16-18] However, its clinical use is somewhat limited by a low spatial resolution and the need for instrumentation modifications for use on a clinical scanner.

Spin-lock MRI techniques have been used to provide noninvasive measures of degeneration in articular cartilage and may potentially be used to assess degeneration in the intervertebral disc. Spin-lock pulses are low power rf pulses applied directly on-resonance with the Larmor precession frequency, locking the magnetization vector into a rotated frame. The relaxation that occurs after the application of a spin-lock pulse is referred to as spin-lattice relaxation in the rotating frame, or $T_{1\rho}$ relaxation. Spin-lock allows the coupling of spins to frequencies that are generally lower than the Larmor frequency. Therefore, slow motion regimes can be studied, such as low frequency physicochemical interactions between water and extracellular matrix molecules. Thus, matrix changes, such as loss of proteoglycan, will be reflected in the $T_{1\rho}$ parameter. $T_{1\rho}$-weighting provides T_2-like images with the advantage of increased dynamic range to degenerative changes compared to conventional T_2-weighting.[19]

In articular cartilage, $T_{1\rho}$ is strongly correlated with proteoglycan content and, thus, has been shown to detect early osteoarthritic changes.[20-22] Recently, $T_{1\rho}$-weighted images of bovine intervertebral disc tissue have been acquired.[23] However, a relationship between $T_{1\rho}$ and intervertebral disc degeneration has not been established, nor has it been shown that $T_{1\rho}$ is sensitive to proteoglycan content in the disc. Thus, the objective of the present study was to demonstrate the use of $T_{1\rho}$ MRI for the assessment of degeneration and proteoglycan content in the human intervertebral disc. Quantitative $T_{1\rho}$ measurements were obtained from cadaveric human intervertebral disc tissue. Degenerative grade was assessed from standard T_2-weighted images, and tissue was subsequently analyzed for total sulfated-glycosaminoglycan content, a measure of proteoglycan content.

Materials and Methods

Seven fresh-frozen cadaveric human lumbar spine sections (mean age 51.6 years, range 15–81) were imaged on a 1.5 T whole-body clinical MR scanner (Sonata; Siemens Medical Solutions). A series of $T_{1\rho}$-weighted images was acquired using a self-compensating turbo spin-echo sequence with parameters: FOV = 28 × 28 cm; slice thickness = 4 mm; acquisition matrix = 512 × 512; and TE/TR = 3000 milliseconds/12 milliseconds. There were 5 evenly distributed spin-lock pulse durations from 15 to 75 milliseconds. The spin-lock pulse amplitude was set to 500 Hz.

$T_{1\rho}$ values were calculated on a pixel-by-pixel basis by a linear regression of intensity data to an exponential decay function: $S(TSL) = S_0 e^{-TSL/T1\rho}$. Values were used to create spatial maps of $T_{1\rho}$. A circular region (5-mm diameter) was manually segmented from the center of the nucleus pulposus, and mean $T_{1\rho}$ was computed within that region.

Diagnostic clinical T_2-weighted images were acquired and used for assessment of degenerative grade for each disc (n = 35). Two orthopedic surgeons and a radiologist independently performed the grading according to the classification scale described by Pfirrmann *et al*.[24] Briefly, discs were graded on a 1–5 integer scale, with grade 1 corresponding to nondegenerate, healthy tissue and grade 5 corresponding to severely degenerate tissue (Table 1).

Discs were isolated *via* sharp dissection, and 1.5-mm punches were harvested from the center of the nucleus pulposus for biochemical analysis. Water content was determined by weighing samples before and after 5 days of incubation at 65°C. Dried samples were then digested in proteinase-K solution, and determination of sulfated-glycosaminoglycan content was performed using 1,9-dimethylmethylene blue in a micro-plate reader assay.[25]

Linear regressions among degenerative grade, age, $T_{1\rho}$,water content, and sulfated-glycosaminoglycan content were performed using GraphPad Prism software (GraphPad Software, San Diego CA). Significance was set at $P < 0.05$, and correlations were considered strong for $r > 0.7$, moderate for $0.5 < r \leq 0.7$, and weak for $r \leq 0.5$.[26]

Results

Representative $T_{1\rho}$-weighted images and corresponding quantitative $T_{1\rho}$ maps are shown for 2 spine sections (donor ages 25 and 51 years) (Figure 1). The $T_{1\rho}$-weighted image is a standard MRI acquired from a single spin-lock pulse. In contrast, the $T_{1\rho}$ map is a graphical representation of the quantitative $T_{1\rho}$ parameter calculated at each pixel location. In general, $T_{1\rho}$ values were higher in the younger, nondegenerate discs. Based on assessment of T_2 images, degenerative grades ranged from 1 to 5. Severely degenerate discs (*i.e*., grades 4 and 5) were excluded from the study because late-stage degenerative changes can be reliably detected using conventional MRI methods, and insufficient tissue was available for biochemical analysis at this late stage.

Therefore, $T_{1\rho}$ measurements were only calculated from discs with grades ≤3.5 (n = 17). $T_{1\rho}$ values ranged from 45 to 173 milliseconds. There was a strong correlation between $T_{1\rho}$ and degenerative grade ($r = -0.76$, $P < 0.01$) (Figure 2). $T_{1\rho}$ was strongly correlated with sulfated-glycosaminoglycan per wet weight ($r = 0.70$, $P < 0.01$), and was moderately correlated with sulfated-glycosaminoglycan per dry weight ($r = 0.67$, $P < 0.01$) and water content ($r = 0.58$, $P < 0.05$) (Figure 3). There was a strong correlation between $T_{1\rho}$ and age ($r = -0.76$, $P < 0.01$).

Discussion

Early degenerative changes in the disc occur in the nucleus pulposus, where proteoglycan content decreases. Spin-lock MRI techniques, such as quantitative $T_{1\rho}$ measurements, may provide a noninvasive method to detect proteoglycan content, allowing detection of early stages of degeneration (Figure 4). As more disc treatment options are developed, including biologic treatments,[27,28] nucleus pulposus replacements,[29] and total disc replacement,[30] better and more sensitive imaging methods will be required to direct the surgeon toward the appropriate treatment. The most important finding of this study was that $T_{1\rho}$ is linearly related to sulfated-glycosaminoglycan content, suggesting that $T_{1\rho}$ may be sensitive to proteoglycan content in the intervertebral disc. We also found that $T_{1\rho}$ MRI was correlated

to degenerative grade. Current grading schemes are susceptible to observer bias and are limited in their ability to detect subtle changes because they are based on a limited 5-level integer scale. In addition, these schemes lack the ability to localize degenerative changes within the disc substructures. In contrast, $T_{1\rho}$ provides a spatial, quantitative measurement that may be more sensitive to early degenerative changes.

The use of quantitative imaging of the intervertebral disc has been investigated using T_1 and T_2 relaxation, magnetization transfer, spectroscopy, and diffusion measurements.[31-37] Boos *et al*[38] found that asymptomatic disc herniations showed shorter T_1 and T_2 relaxation times than symptomatic herniations, although the differences were considered small with respect to the reproducibility of measurement *in vivo*. In the nucleus pulposus, noncollagenous proteins and granular tissue increase with age and degeneration.[1] This fibrosus of the tissue may dampen the signal, shortening T_2 substantially. Because $T_{1\rho}$ is always higher than T_2, the increased dynamic range provided by $T_{1\rho}$ MRI may be particularly beneficial in the disc.

We found that $T_{1\rho}$ in the nucleus pulposus is directly correlated to proteoglycan content. However, in articular cartilage, $T_{1\rho}$ is inversely correlated to proteoglycan content and fixed charge density. Although both disc nucleus pulposus and articular cartilage are primarily comprised of water, proteoglycan, and type II collagen, the relative composition and degenerative processes of these tissues differ.[39] Specifically, other matrix constituents (*i.e.*, collagen, water, degree and type of collagen cross-linking, and proteoglycan aggregation) may influence $T_{1\rho}$ values in the disc. Therefore, future work will be directed at identifying the contributions of individual matrix components to $T_{1\rho}$. This initial study was limited to a relatively small number of cadaveric samples. However, results from this study highlight the potential use of $T_{1\rho}$-weighted imaging of the intervertebral disc, and we recently completed *in vivo* imaging studies.[39a] We found that $T_{1\rho}$ was strongly correlated with age in the present study; future application of this technique to a large patient population of both symptomatic and nonsymptomatic individuals may enable us to separate degeneration from age-related changes in the disc. Ultimately, the content and organization of the extracellular matrix determine the mechanical function of the disc,[40-42] thus, $T_{1\rho}$ may potentially be used to assess disc mechanics.

Conclusions

$T_{1\rho}$ is a promising technique for the *in vivo* diagnosis of intervertebral disc degeneration. It may be particularly useful in detecting the early stages of the disease and directing the appropriate treatment option. Conventional MRI is well suited for detecting the more dramatic changes in disc morphology and hydration that occur in late-stage degeneration. Correlations between $T_{1\rho}$ and sulfated-glycosaminoglycan suggest that $T_{1\rho}$ maybesensitive to proteoglycan changes in the intervertebral disc. Future *in vivo* imaging studies will seek to determine the diagnostic capabilities of $T_{1\rho}$ for early disc degeneration and subsequent low back pain.

Key points

- $T_{1\rho}$ is correlated to degenerative grade and sulfated-glycosaminoglycan content in the nucleus pulposus.
- The technique may provide a noninvasive, quantitative measure of disc degeneration that is particularly sensitive to loss of proteoglycan.
- $T_{1\rho}$ MRI is a potential tool for noninvasive diagnosis of early disc degeneration.

Acknowledgments

The authors thank Drs. Miltiadis Zgonis and Carol Dolinskas for their assistance with disc grading.

Federal funds were received in support of this work. No benefits in any form have been or will be received from a commercial party related directly or indirectly to the subject of this manuscript.

References

1. Buckwalter JA. Aging and degeneration of the human intervertebral disc. Spine 1995;20:1307–14. [PubMed: 7660243]
2. Pearce RH, Grimmer BJ, Adams ME. Degeneration and the chemical composition of the human lumbar intervertebral disc. J Orthop Res 1987;5:198–205. [PubMed: 3572589]
3. Antoniou J, Steffen T, Nelson F, et al. The human lumbar intervertebral disc: Evidence for changes in the biosynthesis and denaturation of the extracellular matrix with growth, maturation, ageing, and degeneration. J Clin Invest 1996;98:996–1003. [PubMed: 8770872]
4. Urban JP, McMullin JF. Swelling pressure of the lumbar intervertebral discs: Influence of age, spinal level, composition, and degeneration. Spine 1988;13:179–87. [PubMed: 3406838]
5. Marchand F, Ahmed AM. Investigation of the laminate structure of lumbar disc anulus fibrosus. Spine 1990;15:402–10. [PubMed: 2363068]
6. Andersson GB. What are the age-related changes in the spine? Baillieres Clin Rheumatol 1998;12:161–73. [PubMed: 9668961]
7. Kirkaldy-Willis WH, Wedge JH, Yong-Hing K, et al. Pathology and pathogenesis of lumbar spondylosis and stenosis. Spine 1978;3:319–28. [PubMed: 741238]
8. Morgan S, Saifuddin A. MRI of the lumbar intervertebral disc. Clin Radiol 1999;54:703–23. [PubMed: 10580761]
9. Gunzburg R, Fraser RD, Moore R, et al. An experimental study comparing percutaneous discectomy with chemonucleolysis. Spine 1993;18:218–26. [PubMed: 8441937]
10. Luoma K, Vehmas T, Riihimaki H, et al. Disc height and signal intensity of the nucleus pulposus on magnetic resonance imaging as indicators of lumbar disc degeneration. Spine 2001;26:680–6. [PubMed: 11246386]
11. Bashir A, Gray ML, Burstein D. Gd-DTPA2- as a measure of cartilage degradation. Magn Reson Med 1996;36:665–73. [PubMed: 8916016]
12. Bashir A, Gray ML, Hartke J, et al. Nondestructive imaging of human cartilage glycosaminoglycan concentration by MRI. Magn Reson Med 1999;41:857–65. [PubMed: 10332865]
13. Burstein D, Velyvis J, Scott KT, et al. Protocol issues for delayed Gd-(DTPA)(2-)-enhanced MRI (dGEMRIC) for clinical evaluation of articular cartilage. Magn Reson Med 2001;45:36–41. [PubMed: 11146483]
14. Ibrahim MA, Haughton VM, Hyde JS. Enhancement of intervertebral disks with gadolinium complexes: Comparison of an ionic and a nonionic medium in an animal model. AJNR Am J Neuroradiol 1994;15:1907–10. [PubMed: 7863940]
15. Ibrahim MA, Jesmanowicz A, Hyde JS, et al. Contrast enhancement of normal intervertebral disks: time and dose dependence. AJNR Am J Neuroradiol 1994;15:419–23. [PubMed: 8197936]
16. Shapiro EM, Borthakur A, Gougoutas A, et al. 23Na MRI accurately measures fixed charge density in articular cartilage. Magn Reson Med 2002;47:284–91. [PubMed: 11810671]
17. Wheaton AJ, Borthakur A, Shapiro EM, et al. Proteoglycan loss in human knee cartilage: Quantitation with sodium MR imaging–Feasibility study. Radiology 2004;231:900–5. [PubMed: 15163825]
18. Reddy R, Insko EK, Noyszewski EA, et al. Sodium MRI of human articular cartilage in vivo. Magn Reson Med 1998;39:697–701. [PubMed: 9581599]
19. Regatte RR, Akella SV, Borthakur A, et al. Proteoglycan depletion-induced changes in transverse relaxation maps of cartilage: Comparison of T2 and T1rho. Acad Radiol 2002;9:1388–94. [PubMed: 12553350]

20. Duvvuri U, Reddy R, Patel SD, et al. T1rho-relaxation in articular cartilage: Effects of enzymatic degradation. Magn Reson Med 1997;38:863–7. [PubMed: 9402184]
21. Akella SV, Regatte RR, Gougoutas AJ, et al. Proteoglycan-induced changes in T1rho-relaxation of articular cartilage at 4T. Magn Reson Med 2001;46:419–23. [PubMed: 11550230]
22. Wheaton AJ, Casey FL, Gougoutas AJ, et al. Correlation of T1rho with fixed charge density in cartilage. J Magn Reson Imaging 2004;20:519–25. [PubMed: 15332262]
23. Regatte, RR.; Akella, SV.; Borthakur, A., et al. High Resolution T1rho Relaxation and Dispersion Imaging of Intervertebral Disc. Proceedings of the International Society for Magnetic Resonance in Medicine; Kyoto, Japan. May 14–16, 2004; p. 1544
24. Pfirrmann CW, Metzdorf A, Zanetti M, et al. Magnetic resonance classification of lumbar intervertebral disc degeneration. Spine 2001;26:1873–8. [PubMed: 11568697]
25. Farndale RW, Buttle DJ, Barrett AJ. Improved quantitation and discrimination of sulphated glycosaminoglycans by use of dimethylmethylene blue. Biochim Biophys Acta 1986;883:173–7. [PubMed: 3091074]
26. Devore, JL. Probability and Statistics for Engineering and the Sciences. 6th ed. Thomson-Brooks/Cole; Belmont, CA: 2004.
27. Masuda K, Oegema TR Jr, An HS. Growth factors and treatment of inter-vertebral disc degeneration. Spine 2004;29:2757–69. [PubMed: 15564925]
28. Shimer AL, Chadderdon RC, Gilbertson LG, et al. Gene therapy approaches for intervertebral disc degeneration. Spine 2004;29:2770–8. [PubMed: 15564926]
29. Guyer RD, Ohnmeiss DD. Intervertebral disc prostheses. Spine 2003;28:S15–23. [PubMed: 12897469]
30. Shuff C, An HS. Artificial disc replacement: The new solution for discogenic low back pain? Am J Orthop 2005;34:8–12. [PubMed: 15707133]
31. Chiu EJ, Newitt DC, Segal MR, et al. Magnetic resonance imaging measurement of relaxation and water diffusion in the human lumbar intervertebral disc under compression in vitro. Spine 2001;26:E437–44. [PubMed: 11698903]
32. Benneker LM, Heini PF, Anderson SE, et al. Correlation of radiographic and MRI parameters to morphological and biochemical assessment of intervertebral disc degeneration. Eur Spine J 2005;14:27–35. [PubMed: 15723249]
33. Antoniou J, Demers CN, Beaudoin G, et al. Apparent diffusion coefficient of intervertebral discs related to matrix composition and integrity. J Magn Reson Imaging 2004;22:963–72.
34. Keshari KR, Zektzer AS, Swanson MG, et al. Characterization of intervertebral disc degeneration by high-resolution magic angle spinning (HR-MAS) spectroscopy. Magn Reson Med 2005;53:519–27. [PubMed: 15723415]
35. Paajanen H, Komu M, Lehto I, et al. Magnetization transfer imaging of lumbar disc degeneration. Correlation of relaxation parameters with biochemistry. Spine 1994;19:2833–7. [PubMed: 7899987]
36. Hsu EW, Setton LA. Diffusion tensor microscopy of the intervertebral disc anulus fibrosus. Magn Reson Med 1999;41:992–9. [PubMed: 10332883]
37. Antoniou J, Pike GB, Steffen T, et al. Quantitative magnetic resonance imaging in the assessment of degenerative disc disease. Magn Reson Med 1998;40:900–7. [PubMed: 9840835]
38. Boos N, Dreier D, Hilfiker E, et al. Tissue characterization of symptomatic and asymptomatic disc herniations by quantitative magnetic resonance imaging. J Orthop Res 1997;15:141–9. [PubMed: 9066539]
39. Mwale F, Roughley P, Antoniou J. Distinction between the extracellular matrix of the nucleus pulposus and hyaline cartilage: A requisite for tissue engineering of intervertebral disc. Eur Cell Mater 2004;8:58–63. [PubMed: 15602703]
39a. Auerbach JD, Johannessen W, Borthakur A, et al. In vivo quantification of human lumbar disc degeneration using $t_{1\rho}$-weighted magnetic resonance imaging. Eur Spine J. 2006 In press.
40. Urban JP, McMullin JF. Swelling pressure of the intervertebral disc: Influence of proteoglycan and collagen contents. Biorheology 1985;22:145–57. [PubMed: 3986322]

41. Johannessen W, Elliott DM. Effects of degeneration on the biphasic material properties of human nucleus pulposus in confined compression. Spine 2005;30:E724–9. [PubMed: 16371889]

42. Best BA, Guilak F, Setton LA, et al. Compressive mechanical properties of the human anulus fibrosus and their relationship to biochemical composition. Spine 1994;19:212–21. [PubMed: 8153833]

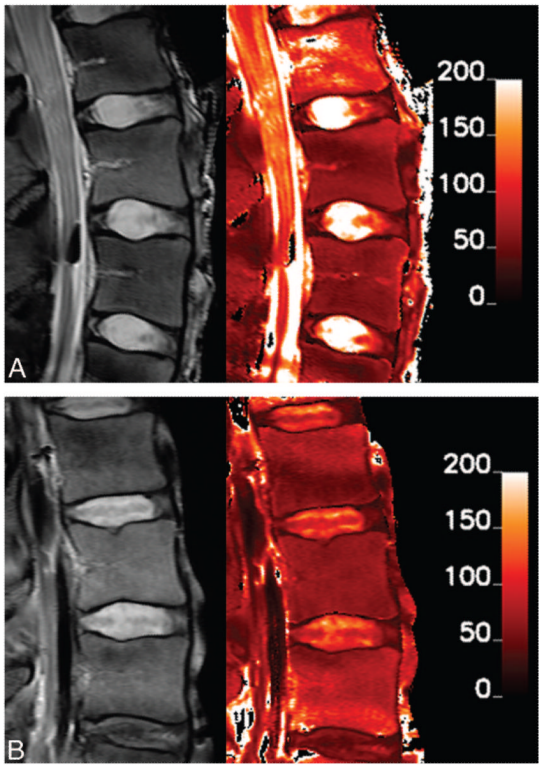

Figure 1.

Representative $T_{1\rho}$-weighted images (**Left**) and quantitative $T_{1\rho}$ maps (**Right**) for 2 lumbar spine sections. Each pixel in the maps represents the quantitative measurement of $T_{1\rho}$ at that spatial location. Age 25, average degenerative grade 1.25 (**Top**). Age 51, average degenerative grade 2.25 (**Bottom**).

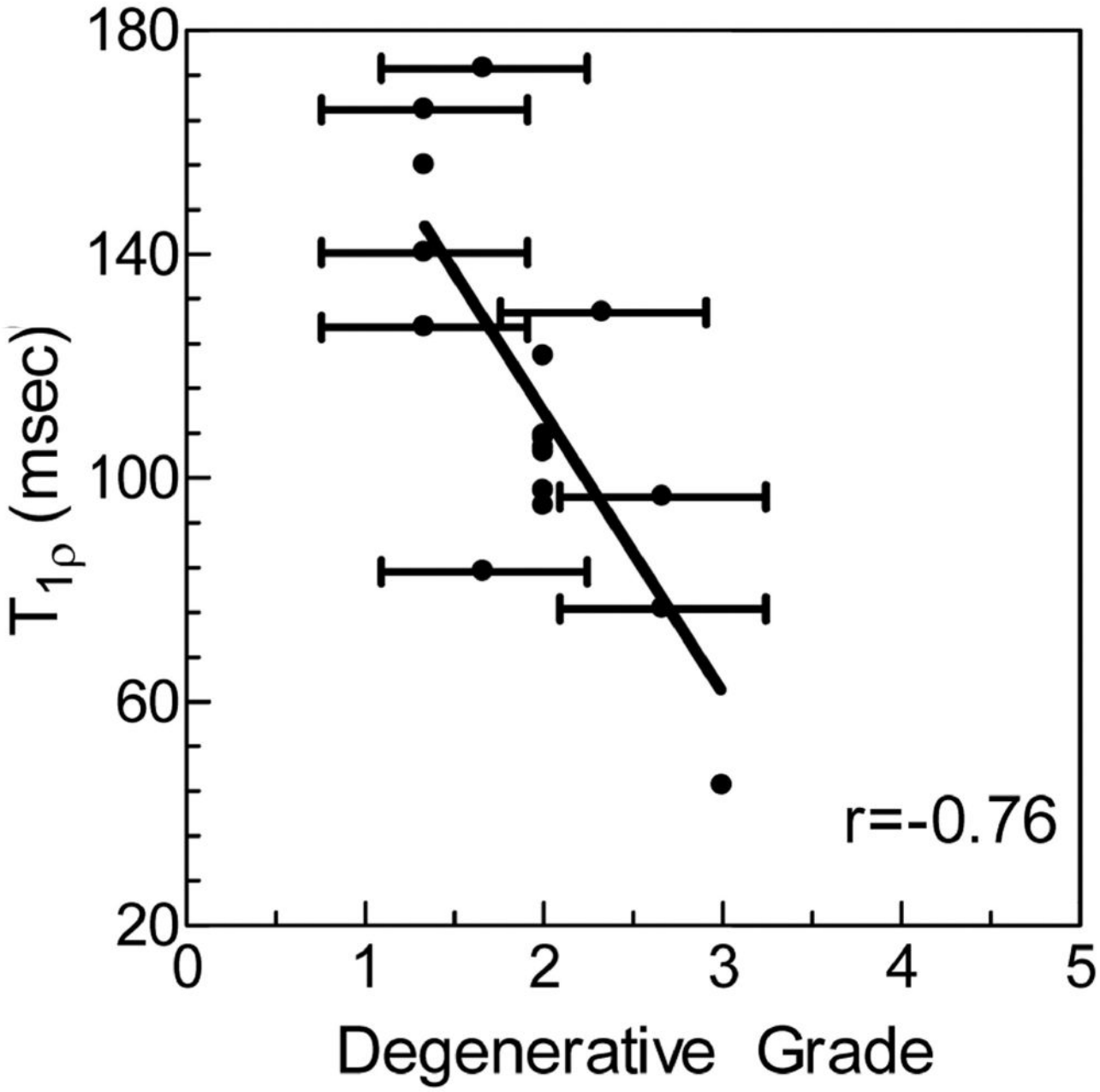

Figure 2.
Correlation between $T_{1\rho}$ values and degenerative grade.

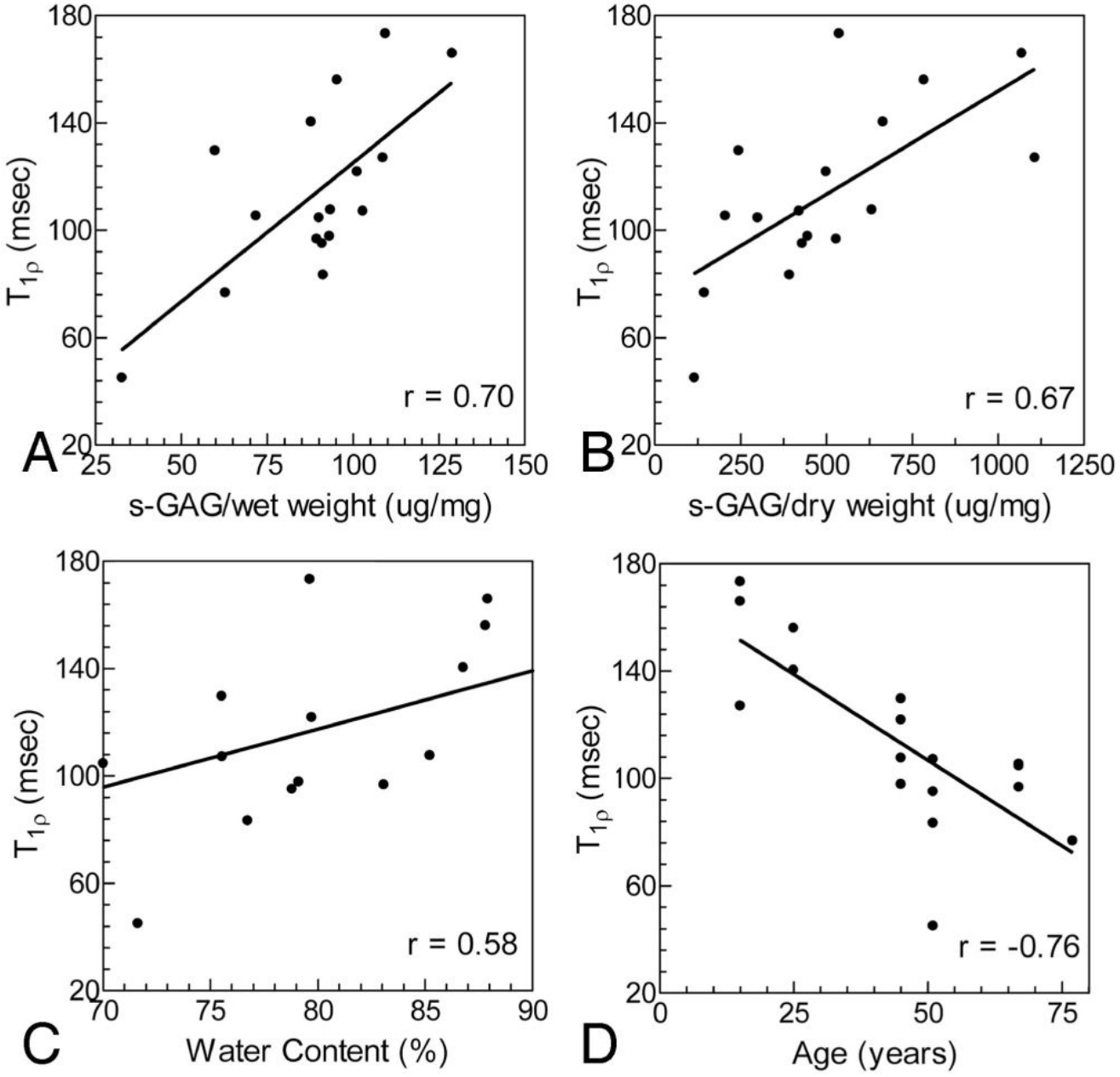

Figure 3.
A, Correlation between $T_{1\rho}$ and sulfated-glycosaminoglycan/wet weight (**B**) $T_{1\rho}$ and sulfated-glycosaminoglycan/dry weight (**C**) $T_{1\rho}$ and water content, and age (**D**).

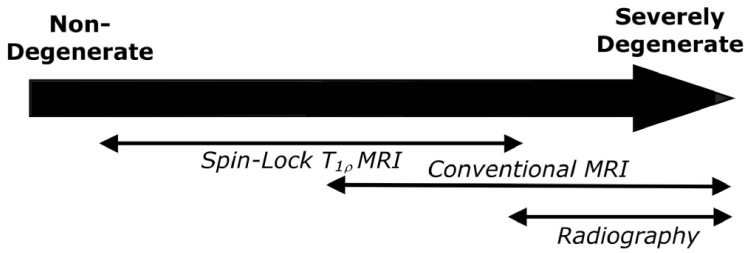

Figure 4.
Spin-lock MRI techniques, such as $T_{1\rho}$-weighted imaging, may detect degenerative changes earlier than conventional MRI or radiography.

Table 1

Grading System for the Assessment of Lumbar Disc Degeneration

Grade	Description
1	The structure is homogeneous, with a bright hyperintense white signal intensity and a normal disc height.
2	The structure is inhomogeneous, with a hyperintense white signal. The distinction between nucleus and anulus is clear, and the disc height is normal, with or without horizontal grey bands.
3	The structure is inhomogeneous, with an intermediate grey signal intensity. The distinction between the nucleus and anulus is unclear, and the disc height is normal or slightly decreased.
4	The structure of the disc is inhomogeneous, with a hypointense dark gray signal intensity. The distinction between nucleus and anulus is lost, and the disc height is normal or moderately decreased.
5	The structure of the disc is inhomogeneous, with a hypointense black signal intensity. The distinction between nucleus and anulus is lost, and the disc space is collapsed.

Adapted with permission from *Spine* 2001;26:1873–8.[24]