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# Evaluation of Water Retention in Lumbar Intervertebral Disks Before and After Exercise Stress with T2 Mapping

Kou Chokan, MD, Hideki Murakami, MD, Hirooki Endo, MD, Yoshikuni Mimata, MD, Daisuke Yamabe, MD, Itsuko Tsukimura, MD, Ryosuke Oikawa, MD, Minoru Doita, MD

Department of Orthopedic Surgery, School of Medicine, Iwate Medical University, Morioka, Japan

Kou Chokan, MD. Department of Orthopaedic Surgery, School of Medicine, Iwate Medical University 19-1 Uchimaru Morioka, Iwate 0208505 Japan Tel. +81-19-651-5111(ext.6404), Fax. +81-19-626-3699 Email: lupin\_punch520@yahoo.co.jp corresponding author

Hideki Murakami,MD. Department of Orthopaedic Surgery, School of Medicine, Iwate Medical University 19-1 Uchimaru Morioka, Iwate 0208505 Japan Tel. +81-19-651-5111(ext.6404), Fax. +81-19-626-3699 Email: hmura@iwate-med.ac.jp

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### Abstract

**Study Design.** T2 mapping was used to quantify moisture content of the lumbar spinal disk nucleus pulposus and annulus fibrosus before and after exercise stress, and after rest, in order to evaluate the intervertebral disk function.

**Objective.** To clarify water retention in intervertebral disks of the lumbar vertebrae by performing magnetic resonance imaging (MRI) before and after exercise stress and quantitatively measuring changes in moisture content of intervertebral disks with T2 mapping.

**Summary of Background Data.** To date, a few case reports describe functional evaluation of articular cartilage with T2 mapping; however, T2 mapping to the functional evaluation of intervertebral disks has rarely been applied. Using T2 mapping may help detect changes in the moisture content of intervertebral disks, including articular cartilage, before and after exercise stress, thus enabling the evaluation of changes in water retention shock absorber function.

**Methods.** Subjects, comprising 40 healthy individuals (males: 26, females: 14), underwent MRI T2 mapping before and after exercise stress and after rest. Image J image analysis software was then used to set regions of interest in the obtained images of the anterior anulus fibrosus, posterior anulus fibrosus, and nucleus pulposus. T2 values were measured and compared according to upper vertebrae position and degeneration grade.

**Results.** T2 values significantly decreased in the nucleus pulposus after exercise stress and significantly increased after rest. According to upper vertebrae position, in all of the upper vertebrae positions, T2 values for the nucleus pulposus significantly decreased after exercise stress and significantly increased after rest. According to the degeneration grade, in the nucleus pulposus of grade 1 and 2 cases, T2 values significantly decreased after exercise stress and significantly increased after rest.

**Conclusion.** T2 mapping could be used to not only diagnose the degree of degeneration but also evaluate intervertebral disk function.

Key words: intervertebral disc, T2 mapping, shock absorber, function of water retention

Level of Evidence: 3

### Introduction

As lumbar intervertebral disk degeneration (IDD) progresses, the deformity of the facet joints and the thickening of ligamentum flavum jointly causes stricture of the spinal canal and neural foramen. These are factors that cause lower back pain and symptoms of the lower limbs. An intervertebral disk (IVD) connects the vertebral bodies juxtaposed above and below it, and is involved in the support and movement of the spine. The nucleus pulposus (NP) is particularly composed of the cells, extracellular matrix, and water. Its 70%–80% water content acts as a shock absorber in response to weight and movement by maintaining elasticity in the IVD. The anulus fibrosus (AF), however, is composed of a concentric circular layered structure and it maintains elasticity in IVDs by resisting outward pressure from the NP. IDD begins from the NP, with decreased water and proteoglycan content and breakdown in collagen structure causing lowered IVD function.

Imaging evaluation of articular cartilage function has previously been attempted with magnetic resonance imaging (MRI)<sup>1</sup> and, in recent years, the use of T2 mapping has enabled quantitatively evaluation of cartilage degeneration from early stages, which could not be rendered on conventional T1 and T2 weighted images.<sup>2</sup> T2 mapping is also effective for evaluating articular cartilage after exercise stress. Liess et al. used T2 mapping to evaluate changes in T2 values and patellar cartilage thickness directly after exercise and after a 45-min rest and reported that cartilage thickness and T2 values increased after rest.<sup>3</sup> Nishii et al. used loading *in situ* MRI to evaluate the knee joints of healthy volunteers by performing T2 mapping with legs subjected to continuous pressure and reported that articular cartilage thickness and T2 values decreased during pressure loading compared to non-pressure loading.<sup>4</sup>

Recently T2 mapping is also becoming useful for the qualitative and quantitative evaluation of early-stage IDD<sup>5,6</sup>.T2 values on T2 mapping correlate with the cartilaginous tissue component of type II collagen, aggrecan core protein, and biochemical quantitative values for water. Thus, although there have been some reports regarding T2 mapping for IDD, very few have involved imaging evaluation before and after exercise stress, as that used for articular cartilage. T2 mapping can be used to quantify IVD water content before and after exercise stress, followed by the determination of water retention or shock absorber function. Here we aim to clarify water retention in IVDs of the lumbar vertebrae by performing MRI before and after exercise stress and quantitatively measuring changes in moisture content of IVDs with T2 mapping.

### MATERIALS AND METHODS

## Subjects

This study was approved by the ethics committee and was conducted in accordance with the ethical rules. Subjects comprised 40 healthy volunteers (males: 26, females: 14) with 200 IVDs

of the lumbar vertebrae (L1/2, L2/3, L3/4, L4/5, and L5/S1). The mean age was 25.6 years (21–31 years). The subjects were medical students with no low back pain or sciatic pain in the past 3 months and with no history of spinal surgery.

### **MRI T2 mapping**

MRI was performed on all subjects at a fixed time in the afternoon considering the influence of diurnal variation in load placed on IVD during the day.<sup>7</sup> All subjects underwent imaging with 1.5-Tesla MRI (Signa HDxt, Ver15, M4A, GE, JAPAN) and imaging conditions were repetition time (TR): 2000 ms, echo time (TE): 13, 26, 39, 51, 64, 77, 90, 103 ms, field of view (FOV): 24 cm, section thickness: 5.0 mm, space: 1.2 mm, matrix: 256×256, number of excitation (NEX): 1. MRI was performed three times (before exercise stress, after exercise stress, and after rest). Exercise stress involved performing 15 repetitions each of just over 30° extension, 45° flexion, and 40° left and right rotation, the normal range of motion of the lumbar spine. After exercise stress, subjects immediately underwent imaging and rested on the MRI table for 30 min in the dorsal position before undergoing MRI again.

Images were color mapped with horizontal sections of the IVDs in the center using image analysis software, Image J (National Institutes of Health, USA), and regions of interest (ROIs) were set in the anterior AF, posterior AF, and NP of each lumbar IVD. ROIs were set in the ventral, central, and dorsal sides of the anterior AF, posterior AF, and NP in horizontal sections mean values for the ROIs were set as T2 values.

## Assessment items and statistical analysis

NP, anterior AF, and posterior AF T2 values for the horizontal sections of IVDs obtained via T2 mapping were summarized and then classified into four grades according to T2 mapping IDD classifications by Watanabe et al.<sup>8</sup> (Figure 1-a,b,c,d) T2 values before exercise stress, after exercise stress, and after rest were compared, according to IVD level and IDD grade. Measurement values were shown as mean  $\pm$  standard deviation. The Student's t-test was used for statistical analysis and a *P*-value of <5% was considered to indicate a statistically significant difference.

#### RESULTS

### T2 values of the NP, anterior AF, and posterior AF

In the NP, T2 values significantly decreased after exercise stress (p<0.01) and significantly increased after rest (p<0.01). No significant differences were observed between values before exercise stress and those after rest. For the anterior AF, no significant differences were noted with exercise stress or rest. For the posterior AF, rest after exercise stress resulted in a significant

increase (p=0.03). (Table 1, Figure 2).

#### T2 values for each IVD level

L1/2: In the NP, values significantly decreased after exercise stress (p<0.01) and significantly increased after rest (p<0.01). In the anterior AF, no significant differences were noted for any of these changes. In the posterior AF, values significantly increased with rest after exercise stress (p=0.04). L2/3: In the NP, values significantly decreased after exercise stress (p<0.01) and significantly increased after rest (p<0.01). No significant difference was observed between before exercise stress and after rest. No significant differences for either of these changes were noted in the anterior AF. In the posterior AF, the values significantly increased with rest after exercise stress (p=0.01). L3/4: the values in the NP significantly decreased after exercise stress (p<0.01) and significantly increased after rest (p<0.01). No significant difference was observed between before exercise stress and after rest. No significant differences for either of these changes were noted in the anterior or posterior AF. L4/5: The values in the NP significantly decreased after exercise stress (p < 0.01) and significantly increased after rest (p < 0.01). No significant difference was observed between before exercise stress and after rest. No significant differences for either of these changes were noted in the anterior or posterior AF L5/S1: The values in the NP significantly decreased after exercise stress (p=0.04) and significantly increased after rest (p<0.01). No significant differences for either of these changes were noted in the anterior or posterior AF. (Table 2, Figure 3-a,b,c)

### T2 values for each IDD grade

There were 140 grade 1, 39 grade 2, 11 grade 3, and 4 grade 4. Grade 1: Values in the NP significantly decreased after exercise stress (p<0.01) and significantly increased after rest (p<0.01). No significant differences for either of these changes were noted in the anterior AF. In the posterior AF, values increased significantly with rest after exercise stress (p=0.02). Gade 2: Values in the NP significantly decreased after exercise stress (p<0.01) and significantly increased after rest (p<0.01). No significant difference was observed between values before exercise stress and after rest (p<0.01). No significant differences for either of these changes were noted in the anterior or posterior AF. Grade 3: No significant changes were observed for the NP, anterior AF, or posterior AF. Grade 4: No significant changes were observed for the NP, anterior AF, or posterior AF. (Table 3, Figure 4-a,b,c)

#### DISCUSSION

In recent years, there have been attempts to quantify and diagnose early-stage IDD, which could not be evaluated with conventional MRI diagnosis, using new MRI technology.<sup>9-15</sup> In T2

mapping, one such MRI technology, the T2 values of tissue are dependent on the phase dispersion speed of hydrogen nucleus spin. Slow phase dispersion due to water molecule structure results in high T2 values.<sup>2,8,16</sup> Thus, T2 values are relatively high in tissue with high water retention, such as the NP, in normal IVDs in young individuals, and, when degeneration causes decrease in water content, T2 values also decrease. However, in the AF in normal IVD, regular collagenous sequences mean that proton mobility in water molecules is limited, causing T2 values to be relatively lowered. When degeneration occurs, this causes collagenous sequences to become irregular, increasing proton mobility and therefore causing T2 values to increase. Watanabe et al. reported that the T2 mapping could confirm IDD at an earlier stage than with conventional MRI imaging. They classified the image findings of IDD by the T2 mapping into 4 stages and stated that this method is useful in evaluating degeneration at an earlier stage as compared with other classification methods. In this study, we could also assess changes in water content of the lumbar IVDs before and after exercise load by using the T2 mapping method. The T2 values in the NP before and after exercise and after rest were all approximately 3 times higher than those in the AF. Our investigation, according to Watanabe's degeneration grade classifications, also indicated that as degeneration progressed, T2 values tended to decrease in the NP and increase in the anterior AF.

Some previous studies have investigated changes in MR images before and after exercise stress in articular cartilage. However, changes in MR images of IVDs before and after exercise stress have been rarely studied. Ellingson et al.<sup>17</sup> performed MRI with the T2 star method and evaluated the degree of degeneration in cadaver lumbar vertebrae after applying flexion, side flexion, and rotation stress. Therefore, we are the first to perform T2 mapping to quantity water content in IVDs before and after exercise stress on lumbar spine in vivo as far as we know. In this study, we compared T2 values before and after exercise stress and after rest. Results indicated that T2 values in the NP decreased after exercise stress and recovered to close to pre-exercise stress levels after rest. However, in the AF, changes were not as significant as those observed in the NP. In articular cartilage, water leaks out when stress is applied to the joint and relative increase in collagen and proteoglycan volume causes changes in MRI signals.<sup>18</sup> In particular, because T2 values directly reflect water and proteoglycan content in tissue, measuring changes in T2 values enables evaluation of water retention in the cartilage. The cartilage maintains elasticity and shape using water retention, so that it functions as a shock absorber. The MRI signal changes observed in the NP in this study appeared to be reflecting changes that occurred when the application of exercise stress to lumbar vertebrae caused water to leak from within the NP and the lost moisture was then once again absorbed during rest. Thus, we could prove by MRI that the NP exhibits a shock absorber function due to its water retention.

We also comparatively investigated T2 values in the NP and AF according to IVD level. In all IVD levels, T2 values in the NP significantly decreased after exercise stress and significantly increased after rest. No significant increases or decreases in T2 values in the AF were observed for any of the IVD levels. In addition, there was no significant change in the T2 value of the anterior AF or posterior AF before and after exercise load; however, the T2 value of the posterior AF significantly increased after rest. Divid et al. reported that a fluid shift occurs from the anterior to the posterior in IVDs with a high T2 value in the NP in a supine position. In this study, the T2 value in the posterior AF increased after rest only in grade 1 IVD in the evaluation by grade, which is consistent with their finding. White et al. investigated mobility in vertebrae in the spinal column and reported that anteroposterior flexion, lateral flexion, and lumbar vertebrae rotation were greatest in L5/S1, followed by between L4/5.<sup>19</sup> Due to compressive force during standing, etc., the degree of lumbar IDD is greater as caudal level.<sup>20-24</sup> In this study, the lumbar vertebrae that exhibited T2 values in the NP was lower in order of L5/S1, L4/5 and L3/4, although T2 values of the NP did not differ significantly at L1/2-L3/4. Since the subjects were young and asymptomatic, the difference in the degree of degeneration between the IVD levels at upper lumbar vertebrae might not have been marked.

Aging and mechanical stress results in degeneration of the IVDs, which gradually develops into functional impairment and IVD disorder. Because T2 mapping can detect earlier stage degeneration than conventional MRI techniques, it aid early diagnosis and treatment of early-stage cases of IDD possible and could be used for preventing this disease. In this study, we used T2 values to not only evaluate the degree of degeneration but also assess IVD water retention by quantifying water content in the NP and AF before and after exercise stress and after rest. T2 mapping method reported by Watanabe et al was employed to categorize IDD into grades of degeneration. Grade 1 and 2, in which a high T2 or mildly decreased T2 was observed (i.e., mild IDD), showed higher amounts of water in the IVD before an exercise test, and water lost in the exercise test showed good recovery through resting. We can therefore assume that in these cases, the shock absorption is maintained through proper water retention ability. This suggests that the T2 mapping method can be used not only for diagnosing the degree of degeneration, but also for functional evaluation of IVDs. This method has the potential of providing useful information in evaluating benefits or defects from conservative therapies such as physical therapy or exercise therapy, or the effects of surgical treatments such as fixation on the IVDs.

There are some limitations to this study. The first one is about the exercise test method. Although it is well known that the pressure in the lumbar IVDs is higher in extension, flexion or during rotation exercise in comparison to being in the upright position, the relationship between the angle during each exercise and the resulting load is unknown. It is also unknown how much

load is generated by each exercise repeated particular times. These are subjects of our future investigation. Meanwhile, for the present study, we considered the appropriate angle during the exercise test to be the upper limit of the normal range of motion in which daily activities can be carried out without pain; and the appropriate number of repetitions to be 15 times each of extension and flexions, and 15 times each of right and left rotation, which mean a total of 60 repetitions, as it can be done without strain. It would be also necessary to examine the relationship between the degree of exercise test load and observations in IVD T2 mapping images in future studies. The IVD volume, especially the volume of the NP, i.e., the water content, is known to show a circadian variation, because the diurnal cycle of erect and supine posture result in large changes in compressive load acting on the IVD. In order to eliminate the effects of difference between morning and evening, MRI imaging was conducted at a set time in the afternoon in this study. However, there are no previous reports examining this circadian variation using the T2 mapping method; therefore, it should be verified in the future. In the past, we have used rabbit and human lumbar IVDs to demonstrate that the degree of degeneration determined by the T2 mapping method correlate with the contents of water and proteoglycan measured biochemically in the NP. However, a significant correlation was not demonstrated in the AF. Similar results were observed using the immunohistological method. We are considering increasing the sample size to report on these results. In addition, the inconsistent results from this study on the AF resemble the results from previous studies, we think that further detailed evaluations are required in the future.

#### CONCLUSION

In IVDs with mild degeneration, T2 changes in the NP were observed but such changes were not noted IVDs with advanced degeneration. In conclusion, T2 mapping could be used for the functional evaluation of IVDs.

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Figure 1. Classification of Intervertebral Disk Degeneration Using Axial T2 Mapping as reported by Watanabe et al.<sup>8</sup>.

A:Grade I . T2 Value of NP is high T2 and homogeneous. T2 Value of AF is low T2 and homogeneous. Distinction of NP and AF is clear with regular border.

B:Grade II. T2 Value of NP is Mild decrease of T2,mildly inhomogeneous. T2 Value of AF is mild increase of T2,mildly inhomogeneous. Distinction of NP and AF is clear with irregular border.

C:GradeIII. T2 Value of NP is moderate decrease of T2 and moderately inhomogeneous. T2 Value of AF is moderate increase of T2 and moderately inhomogeneous. Distinction of NP and AF is Unclear.

D:GradeIV. T2 Value of NP is severe decrease of T2 and severely inhomogeneous. T2 Value of AF is severe increase of T2 and severely inhomogeneous. Distinction of NP and AF is lost.



Figure 2. T2 values of the nucleus pulposus, anterior anulus fibrosus, and posterior anulus fibrosus

In the nucleus pulposus, T2 values significantly decreased after exercise stress and significantly increased after rest. In the anterior anulus fibrosus, no significant differences were noted with exercise stress or rest. In the posterior anulus fibrosus, T2 values significantly significantly increased after rest.



Figure 3-a. T2 values of the NP for each IVD level.

The values significantly decreased after exercise stress and significantly increased after rest.





Figure 3-b. T2 values of the anterior AF for each IVD level. No significant differences for either of these changes were noted

Figure 3-c. T2 values of the posterior AF for each IVD level. The T2 values significantly increased with rest after exercise stress.



Figure 4-a. T2 values the NP for each IDD grade.

In grade1 and 2 T2 values significantly decreased after exercise stress and significantly increased after rest.



Figure 4-b. T2 values the anterior AF for each IDD grade. No significant differences for either of these changes were noted.







	Before exercise stress	After exercise stress	After rest
NP	$131.0 \pm 28.6$	$117.9 \pm 24.2$	$129.6 \pm 27.3$
Anterior AF	$49.8 \pm 13.0$	$48.5 \pm 14.7$	$48.9 \pm 11.1$
Posterior AF	$46.4 \pm 11.1$	$46.0\pm9.0$	$48.3 \pm 11.8$

Table 1. T2 values of the NP, anterior AF, and posterior AF.

			1	
	Level	Before exercise stress	After exercise stress	After rest
NP	L1/2	$124.1 \pm 28.3$	$112.4 \pm 26.1$	$120.1 \pm 29.6$
	L2/3	$135.4 \pm 36.0$	$118.6 \pm 32.1$	$135.1 \pm 33.2$
	L3/4	$147.6 \pm 26.7$	$129.3 \pm 23.3$	$144.9 \pm 25.7$
	L4/5	$134.9 \pm 23.0$	$120.2 \pm 17.1$	$130.3 \pm 20.1$
	L5/S1	$113.1 \pm 12.4$	$108.9 \pm 13.5$	$117.7 \pm 16.1$
Anterior AF	L1/2	$48.7 \pm 13.8$	47.5 ± 11.0	49.4 ± 14.2
	L2/3	$48.1 \pm 9.0$	$46.6 \pm 8.4$	$48.3 \pm 9.7$
	L3/4	$49.2 \pm 7.8$	$49.6 \pm 7.8$	49.3 ± 8.1
	L4/5	$54.5\pm20.0$	50.1 ± 18.2	$51.0 \pm 12.0$
	L5/S1	$44.0\pm9.9$	$48.5 \pm 22.4$	$46.5 \pm 10.6$
Posterior AF	L1/2	$48.12 \pm 8.22$	48.36 ± 10.06	$52.06 \pm 12.46$
	L2/3	43.1 ± 7.0	$42.6 \pm 9.9$	$47.3 \pm 10.2$
	L3/4	46.1 ± 8.7	$46.3 \pm 7.9$	$47.7 \pm 11.5$
	L4/5	49.2 ± 18.2	$45.2 \pm 8.1$	$48.0 \pm 10.6$
	L5/S1	45.3 ± 9.2	47.7 ± 7.8	$46.4 \pm 13.7$

Table 2. T2 values for each IVD level.

Table 2. T2 values for each IVD level.

In all of the vertebrae positions, T2 values for the NP significantly decreased after exercise stress and significantly increased after rest. In the L1/2 and L2/3 posterior AF, T2 values increased significantly after rest. No other significant changes were observed.



	Grade	Before exercise stress	After exercise stress	After rest
NP	1	$138.3 \pm 24.9$	$123.0 \pm 21.4$	$135.8 \pm 24.6$
	2	$122.1 \pm 25.6$	$113.4 \pm 21.7$	$124.63 \pm 21.7$
	3	92.5 ± 23.3	88.7±19.7	$124.6 \pm 21.7$
	4	$60.5 \pm 24.9$	55.2 ± 23.2	71.5 ± 34.1
Anterior AF	1	$49.2 \pm 14.1$	$48.0 \pm 16.2$	49.0 ± 11.7
	2	51.1 ± 11.6	47.7 ± 11.8	$48.2 \pm 9.8$
	3	$51.8 \pm 6.7$	53.6 ± 8.2	53.2 ± 9.3
	4	$52.3 \pm 1.2$	$50.3 \pm 4.8$	$47.9 \pm 4.1$
Posterior AF	1	$48.5 \pm 11.7$	$46.6 \pm 8.8$	$49.7 \pm 12.3$
	2	$48.4 \pm 11.0$	$46.6 \pm 9.3$	$48.4 \pm 10.1$
	3	$48.4 \pm 7.7$	$46.7 \pm 9.6$	$48.5 \pm 8.3$
	4	$48.3 \pm 6.0$	$46.5 \pm 1.5$	$47.9 \pm 20.7$

Table 3. T2 values for each IDD grade.

Table 3. T2 values for each IDD grade.

In the NP of grade 1 and 2, T2 values significantly decreased after exercise stress and significantly increased after rest. In grade 1, T2 values also significantly increased after rest in the posterior AF. No other significant changes were observed.

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