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Original research article

Comparison of the biochemical and radiological criteria for lumbar disc degeneration

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ABSTRACT

Background: The relationship between radiological degeneration criteria on lumbar magnetic resonance imaging (MRI) and both the keratan sulfate (KS) and chondroitin sulfate (ChS) levels was examined in disc material taken from patients undergoing lumbar disc herniation (LDH) surgery. To examine whether the biochemical and radiological degeneration criteria testing the reliability of radiological degeneration findings agreed and to evaluate the contribution of the KS/ChS ratio to disc form (protruding or extruding).

Methods: This was a prospective experimental cohort study. Using enzyme-linked immunosorbent assay, KS and ChS levels were measured in the degenerate nucleus pulposus taken from 71 patients with a diagnosis of LDH who underwent surgery. The degeneration levels and disc form (protruding or extruding) were determined according to the Pfirrmann five-stage grading system on preoperative T2-weighted lumbar MRIs. According to the Pfirrmann system, 28 patients were grade III and 43 were grade IV. The relationship between radiological criteria and the KS/ChS ratio was statistically evaluated.

Results: The KS levels ($p = 0.046$) and the KS/ChS ratio ($p = 0.001$) were significantly higher in grade IV patients than in grade III patients. However, there was no difference between the KS and ChS levels and the KS/ChS ratio when patients were classified as protruding or extruding according to their disc structure. Disc structure and biochemical degeneration indicators were not correlated.

Conclusions: The KS level and the KS/ChS ratio were high in patients with marked radiological degeneration on lumbar MRI, demonstrating the sensitivity and reliability of the Pfirrmann five-stage grading system for showing radiological degeneration.

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1. Introduction

The intervertebral disc (IVD) consists of two main structures: the gelatinous, water-binding nucleus pulposus and a layered circular structure called the annulus fibrosus. One of the first signs of disc degeneration is the loss of proteoglycans from the nucleus pulposus [1]. It has been hypothesized that the subsequent loss of water results in a loss of pressure, leading to the collapse of IVD and clinical signs, such as decreased disc height and decreased signal intensity on T2-weighted magnetic resonance imaging (MRI) [2]. Several morphologic grading systems for lumbar disc degeneration have been described. One of the most used grading systems is Pfirrmann et al. classification and grading system for degeneration of lumbar disc herniation [3]. Pfirrmann grading system is based on T2-weighted MRI features of IVD; disc structure, signal intensity, distinction between nucleus and anulus, and disc height. The latter feature is important for distinguishing between Grades IV and V discs. For Grades III and IV, the disc height is not a discriminative feature. The discriminative feature between Grades III and IV is distinction between nucleus and anulus. If it was lost, the radiologist of MRI can classify it as Grade IV. Homogeneous disc structure is the discriminative feature for Grade I discs [3].

The amount of degeneration in IVD can be biochemically quantified by determining the amount of collagen present in the nucleus pulposus, the number of collagen cross-links, the proportions of proteoglycan and collagen, and the keratan sulfate (KS)/chondroitin sulfate (ChS) ratio.

This study evaluated the reliability of the Pfirrmann five-stage classification by comparing the preoperative MRI with the KS/ChS ratio in disc material (degenerate nucleus pulposus) obtained perioperatively and investigated the effects of biochemical degeneration on the type of disc herniation (protruding or extruding).

2. Material and methods

This study was conducted at the Neurosurgery Clinic of our between March 2016 and September 2016. Seventy-one patients diagnosed with lumbar disc herniation (LDH) were included in this study after obtaining their verbal and written consent. Patients with cervical or thoracic disc degeneration, active malignancy, pregnancy, prior back surgery, spine fractures, sacroiliac arthritis, metabolic bone disease, spinal infection, or rheumatoid arthritis and patients having abdominal trauma within 7 days of admission were excluded.

The patients included in this study were classified as grade III ($n = 28$) or IV ($n = 43$) according to Pfirrmann's radiological classification system (Fig. 1a and b). According to Pfirrmann grading system for degeneration of lumbar discs; Grade III was described as inhomogeneous with gray colored discs, in which unclear distinction of nucleus and anulus with normal or slightly decreased disc height. For Grade IV, the distinction of nucleus and anulus was lost with normal or moderately decreased disc height [3]. According to the appearance of the disc herniation on MRI, 28 patients had the protruding form and 43 had the extruding form. Surgery was performed at levels L2-3, L3-4, L4-5, and L5-S1 in 5, 9, 33, and 24 patients, respectively (Table 1).

Disc material collected during surgery was stored at −80 °C until it was sent for biochemical analysis to analyze their KS and ChS content. The tissue samples were homogenized in phosphate-buffered saline (pH 7.4) (FastPrep 24; MP Biomedicals, Santa Ana, CA, USA). Total protein levels in all homogenized tissue samples were spectrophotometrically measured using the Bradford method [4]. The KS and ChS levels in the homogenates were measured with the sandwich enzyme-linked immunosorbent assay (ELISA) using commercial kits (lot no: E1459/Hu-E1895/Hu; Bioassay Technology Laboratory, Shanghai, China) and an ELISA reader (Thermo Fisher Scientific, Waltham, MA, USA). The ELISA result of each sample was divided by the amount of protein found. The results were expressed in ng/mg protein.

The radiological degeneration was evaluated by three independent radiologists using the five-stage Pfirrmann classification system based on T2-weighted mid-sagittal images obtained using contemporary MRI techniques. All three radiologists independently evaluated by three certified radiologists who were blinded to the classification groups of this study (radiologists did participate in any part of operation or biochemical analysis). The interobserver agreement was acceptable. Complete agreement was achieved in a range from

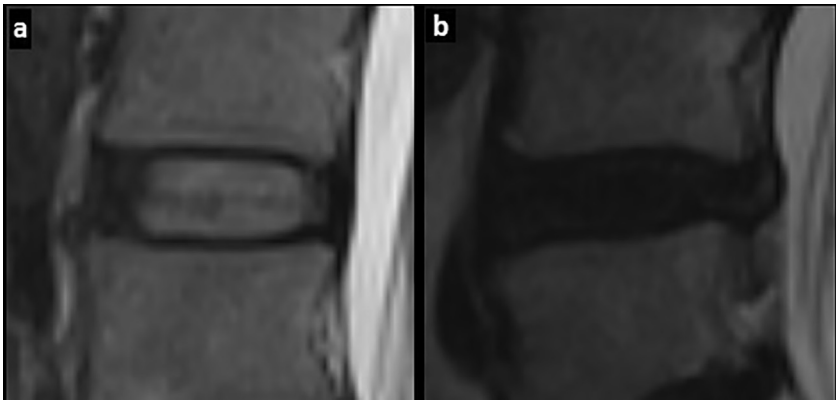

Fig. 1 – Sagittal T2-weighted MRI images. They are evaluated as Pfirrmann grade III (a) and grade IV (b). *MRI: magnetic resonance imaging.

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