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Microscopic Assessment of Degenerated Intervertebral Disc: Clinical Implications and Possible Therapeutic Challenge

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Abstract. *Aim: To investigate a possible correlation between the histological and morphometric properties of herniated intervertebral disc, clinical and magnetic resonance imaging (MRI) characteristics of patients with lumbar disc degeneration (LDD). Materials and Methods: Thirty six patients with LDD were clinically evaluated using Japanese Orthopaedic Association Score (JOAS), visual analogue scale (VAS) for pain in the lower back or in the pelvic limb; MRI-based classification according to Pfirrmann and Modic criteria. All patients underwent decompressive surgery and herniated intervertebral disc samples were histologically and morphometrically analyzed. Data obtained were statistically analyzed for bivariate and partial correlations. Results: The mean area size of chondron clusters correlated with age, JOAS (r=–0.385, p=0.032, tau=–0.279, rho=-0.380), Pfirrmann (r=0.505, p=0.002, tau=0.289, rho=0.365) and Modic (r=0.500, p=0.002, tau=0.331, rho=0.419) grading. There was a strong correlation between maximum area size of chondrons and JOAS (r=–0.427, p=0.009, tau=–0.299, rho=–0.430), Pfirrmann changes (r=0.432, p=0.008, tau=0.309, rho=0.388) and Modic endplate changes (r=0.444, p=0.007, tau=0.343, rho=0.434). JOAS correlated with both MRI classifications used for LDD. Conclusion: The intervertebral disc cells tend to aggregate in clusters and the size of the chondrons from LDD correlated with JOAS, Pfirrmann and Modic. JOAS correlates with the imagistic evaluation systems Pfirrmann and Modic.*

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Lumbar disc degeneration (LDD) including lumbar disc herniation (LDH) is a major cause for lower back pain with a high prevalence leading to very high associated costs (1-4). Still, there are certain "blurry" aspects regarding its etiology, risk factors, evolution and stage-related therapies. To date there are many studies trying to uncover risk factors associated with intervertebral disc disease (IDD), such as body mass index (BMI), mechanical loading, physical activities, smoking or familial predisposition (5-8). Research is also targeted towards elucidating the physiological molecular mechanisms that lead to, or can modify the, degeneration process that takes place in the intervertebral disc (9-12). To date it is known that in the normal intervertebral disc, cells have a scattered distribution, constituting only 1% of the adult disc tissue volume and are responsible for secretion and organization of the extracellular matrix, which forms the major component of the fibro-elastic cartilage. LDD is characterized by an alteration of cell functions leading to loss of proteoglycans and, due to the fact that intervertrebal disc (IVD) has a limited capacity to regenerate, until now, we consider this process naturally irreversible. Changes occurring during the degenerative process consist of shape and volume alterations of the disc, an increase in blood vessels number and matrix metalloproteinase activity, presence of clefts, alterations of tensile properties and dehydration (13-15). The cell population is also subjected to dramatic changes with age, due to apoptosis of the cells in the nucleus pulposus (NP), shifting to small sparse cells resembling chondrocytes. As these cells regulate the matrix composition, cell population changes also affect the concentration of large matrix proteoglycans, such as aggrecan, and the secretion of collagen. As the degenerative process continues, there is an increase in vascularization and innervation of the disc's peripheral tissue, changes in extracellular matrix composition and the appearance of chondron clusters. These, often large, aggregations of chondrocyte-like cells have a big overall volume and, thus, can dislocate important amounts of extracellular matrix leading to a further decrease in biomechanical characteristics. However, little is known about the relationship of these changes and the

actual clinical features of LDD, especially in patients who have indication for surgery and more likely to be subjected to key degeneration patterns. A link between IDD and its clinical features was created through magnetic resonance imaging (MRI) studies that managed to quantify the degeneration process within IVD and provided classification systems helping clinicians in the therapeutic management. Currently, there are two widely used classifications: the Pfirrmann grading system, providing morphologic and semi-quantitative evaluation of IVD (16, 17) and the Modic grading system based on endplate signal intensity modifications (18). Some consider the Pfirrmann grading system as being subjective and inadequate to evaluate severe disc degeneration (16) leading to a more complex modified Pfirrmann classification system (17). Several studies found strong correlations between Modic changes and lumbar disc degeneration and, therefore, regard endplate degeneration as a consequence of LDD (19, 20). However, these MRI studies have included mainly patients with mild disc degeneration processes and only few with severe degeneration. There is a lack of data regarding MRI findings and the degree of disc degeneration in patients that have an indication for surgery. Furthermore, to our knowledge, no data exist on the relation between MRI findings, clinical features and morphometric characteristics in LDD.

The aim of this clinicopathological study was to investigate the clinical relevance of a correlation between histological and, especially, the morphometric aspects of surgically obtained disc material with the MRI findings and symptomatology in patients with LBP undergoing surgical treatment.

Materials and Methods

The current study was undertaken in accordance with the local ethics committee guidelines and was approved by the Ethical board of Timis County Emergency and Clinical Hospital. Written informed consent for the experimental use of the surgical samples was obtained from each patient.

Patients and data collection. The present study was conducted on 36 patients, between October 2012 and January 2014 for LDD. Initially, all patients underwent 6 weeks of conservatory treatment prior to surgical treatment. Patients were evaluated using the criteria of the Japanese Orthopaedic Association evaluation system for low back pain syndrome (JOAS) and Visual Analogic Scale (VAS) for lumbar and pelvic limb pain. The JOAS ranges from a minimum of -6 to a maximum of 29. After the patients were investigated through MRI, they underwent surgical treatment (discectomy or sequestrectomy), performed by the same team of surgeons. Herniated disc fragments were obtained during surgery and further analyzed.

MRI assessment. Lumbar MRI examination was performed on a 1.5 T scanner (Siemens Magnetom Essenza, Siemens AG, Erlangen, Germany), using a dedicated spine coil. The imaging protocol included sagital T1-weighted spin-echo and T2-weighted FSE images that were analyzed and classified according to Pfirrmann and Modic grading systems by two of the authors (PI, MO).

Table I. *Descriptive data on clinical and imagistic parameters studied*.

	Minimum	Maximum	Mean	SD
J.O.A.S.	1	13	7.83	2.71
Lumbar VAS	2	10	7.14	1.78
Pelvic limb VAS	5	10	8.20	1.14
Pffirrmann	2	5	3.64	0.72
Modic	0	3	1.61	0.80

JOAS: Japanesse Orthopaedic Association Score. VAS: Visual analogic scale. SD: Standard deviation.

Tissue biopsies. Disc specimens were obtained during the surgical intervention from spinal levels L4-L5 or L5-S1. They were washed and fixed in 10% buffered formal saline immediately following surgery.

Histological preparation. Surgically-obtained herniated intervertebral disc specimens were fixed for up to 48 h and then paraffin embedded. Three micrometers serial sections were obtained from paraffin blocks. Sections were stained with hematoxylin and eosin (H&E) and toluidine blue (TB). The sections were viewed with a light microscope (Nikon Eclipse E600, Nikon Corporation, Tokyo, Japan).

Morphometry. Systematic examination of the toluidine blue-stained sections was carried out under the light microscope so that the area observed was clearly defined. Six random grid areas per section were viewed at ×10 objective magnification. The number of chondron cluster profiles was counted (defined as a group of nuclei forming a cluster with a distinct and dense surrounding matrix). Using a computer software, NIS Elements 4.2 (Nikon, Nikon Corporation, Tokyo, Japan), each chondron cluster area was measured in pixels.

Statistical analysis. Data obtained were subjected to standard statistical analysis using the SPSS 20.0 statistical software (IBM Corporation, Armonk, NY, USA). Both parametric and non-parametric bi-variate correlation analyses were performed. We noted the r coefficient, Kendall's tau-b (τ) and Spearman's rho (ϱ). A p-value of ≤ 0.05 was considered as statistically significant and a p-value of ≤ 0.001 was considered as highly significant.

Results

Thirty-six patients were evaluated (23 males and 13 females) by clinical and MRI methods. During surgery, 36 biopsy specimens were obtained that were evaluated through histological and morphometric methods. Thirty-six hematoxylin and eosin (HE)-stained sections and 36 toluidine blue (TB) sections were created. For the morphometric assessment, 216 fields of view from the 36 HE-stained sections were evaluated.

Descriptive data. The mean age of the patients was 39.3 years (ranging from 21 to 62 years). Mean JOAS was 7.8 (ranging from 1 to 13), mean lumbar VAS was 7.1 (ranging

Table II. *Morphometric characteristic of studied specimens from herniated intervertebral discs.*

Assessment	Minimum	Maximum	Mean	SD
Total no. of chondrons/field of view	8.00	51.00	30.30	9.08
Mean surface area of chondrons (pixels)	658.70	2,937.00	1,615.36	559.39
Minimum surface area of chondrons (pixels)	188.00	925.00	493.42	198.63
Maximum surface area of chondrons (pixels)	1,171.00	1,1811.00	5,250.94	2,785.34
Median surface area of chondrons (pixels)	247.50	1621.00	785.3944	324.79
Deviation of the area of chondrons (pixels)	592.00	2,419.50	1,305.6250	482.22
Chondrons/FOV	1.70	7.80	5.0750	1.57

FOV: Field of view. SD: Standard deviation.

from 2 to 10) and mean pelvic limb VAS was 8.2 (ranging from 5 to 10). The MRI study revealed 2 patients with grade II Pfirrmann degeneration, 12 with grade III degeneration, 19 with grade IV degeneration and 3 patients with grade V Pfirrmann degeneration (Table I). No type I Pfirrmann intervertebral discs were identified. When grading according to Modic degeneration scale (Table I) 4 patients had no endplate changes thus, we noted them as Modic 0 (11.1%), nine patients were with grade I degeneration (25%), 20 with grade II degeneration (55.6%) and 3 with grade III degeneration (8.3%). Modic endplate changes were observed in 32 cases (88.9%)

Histological characteristic of degenerated intervertebral discs. On the degenerated disc sections, highly fibrotic lamellae could be seen, as well as interlamellar cells both round and elongated. Layers were irregularly distributed with an increase in the interbundle spaces (optically-empty spaces) suggesting cartilage degeneration consecutive to a sustained load response (Figure 1a). Chondron clusters, aggregates of three or more rounded cells with a territorial matrix, seemed to be dependent on the grade of degeneration. Chondron clusters, with a pale stained pericellular matrix, were seen forming from the rounded cells rather than the elongated ones (Figure 1b). It seems that cells from the prolapsed tissue proliferate in small groups (Figure 1c) dynamicly trying to produce matrix, although insufficient and form a distinct chondron cluster in an attempt to adapt to changes in biomechanical loads in the tissue. This is similar to a regenerative process but it looks like being a faulty one producing insufficient extracellular matrix and chondrocyte-like cells in different stages of evolution. Normal fibrous cartilage was observed to be in mixed composition with fibrous cartilage tissue in either regenerative or degenerative phases. Chondrons in the regeneration areas are formed from groups of 3 or more chondrocyte-like cells (Figure 1d), compared to normal tissue areas where a lacuna is formed from only 1 or 2 chondrocytes (Figure 1d, inset). The morphology of the chondrocyte-like cells forming the chondrons inside the regeneration areas corresponds to all three types of chondrocytes. Around the lacunas (chondrons) in the regenerated cartilage there is no or insufficient territorial matrix. Cartilage matrix is disorganized, with many optically-empty spaces on HE staining suggestive for fissures or cracks. Around the chondrocyte areas that appear to be proliferative vascular structures lined by endothelium with a high tendency to invade the cartilage and to be distributed in a peri-lacunar arrangement can be observed (Figure 1d, arrows, Figure 1e, f). In contrast, around the lacunas from the remaining normal fibrous cartilage no vascularization can be seen. In some specimen, nervous structures suggesting terminal fillets could be observed.

Morphometric characteristic of degenerated intervertebral discs. For the morphometric evaluation of the intervertebral disc tissue (Table II) 6 fields of view/ specimen were evaluated. The mean total number of chondrons encountered per specimen was 30 (ranging from 8 to 51), the mean surface area of the chondrons was 1,615.4 pixels (ranging from 658.7 to 2,937 pixels), the mean minimum chondron area was 493.4 pixels (ranging from 188 to 925 pixels), the mean maximum chondron area was 5,250.9 pixels (ranging from 1,171 to 11,811 pixels) and a mean overall median of 785.4 (ranging from 247.5 to 1,621 pixels).

Statistical analysis. The mean surface area of the chondron was negatively correlated (Table III) with the JOA score (r=–0.385, p=0.032, Kendall's tau=–0.279, Spearman's rho=–0.380) and directly proportional with Pfirrmann grading scale (r=0.505, p=0.002, tau=0.289, rho=0.365) and with Modic degeneration scale (r=0.500, p=0.002, tau=0.331, rho=0.419). The mean surface area of the chondrons correlated strongly with age (r=0.456, p=0.005, tau=0.351, rho=0.463) and only with the lumbar VAS (r=0.376, p=0.024, tau=0.221, rho=0.304) and not with pelvic limb VAS. Age correlated with the Modic degeneration scale (r=0.370, p=0.026, tau=0.305, rho=0.395) and with Pfirrmann grading (r=0.354, p=0.034, tau=0.242, rho=0.321).

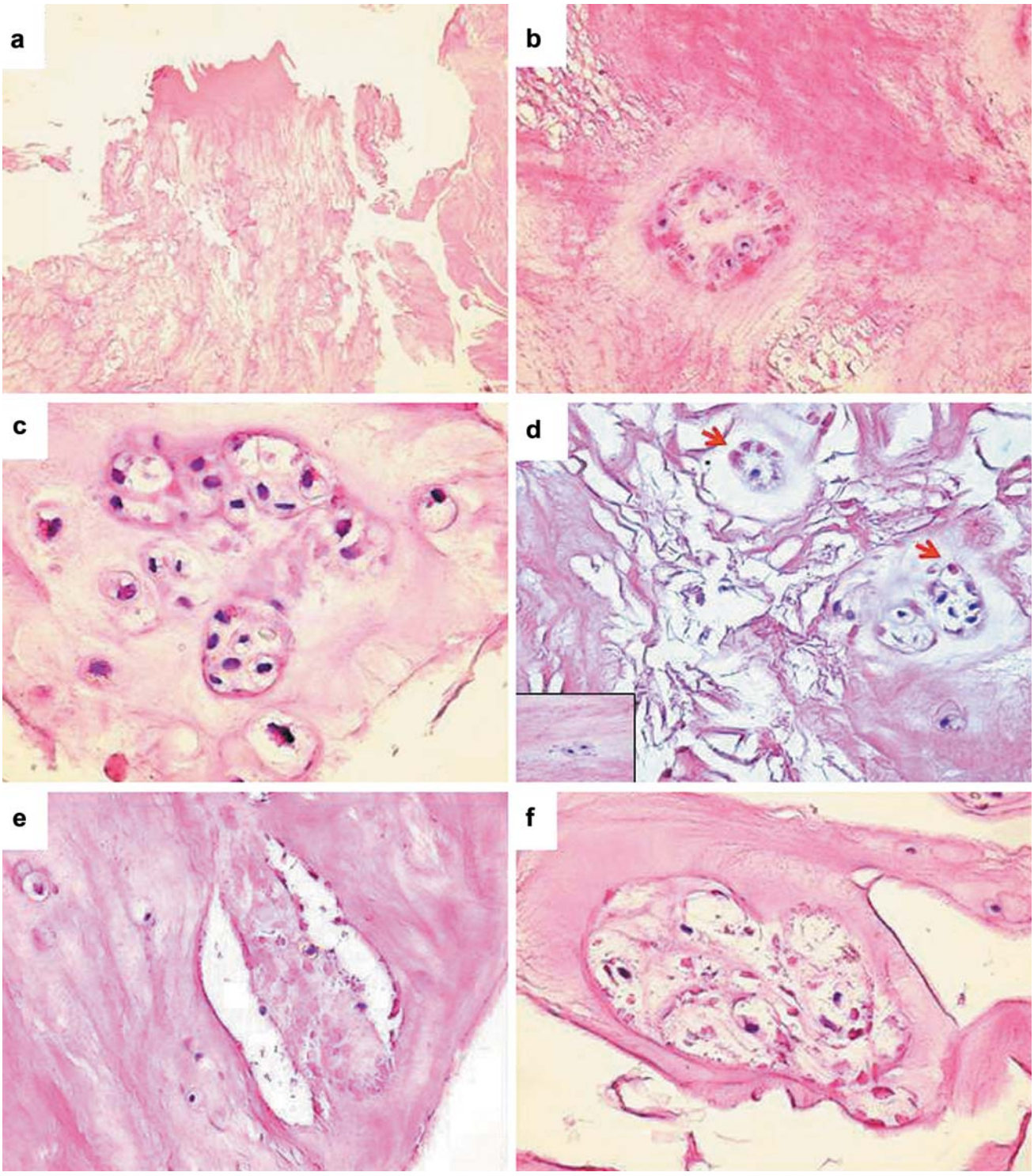

Figure 1. *Histological aspects of cartilage changes during lumbar disc herniation and degeneration. General overview of the modified fibrous cartilage with a loose matrix and increased number of lacunae (a). Lack of normal appearance of territorial matrix, which becomes looser than in normal condition (b). Chondrons composed of heterogeneous groups of chondrocyte- like cells (c) showing a high density inside the matrix. Degenerated matrix was characterized by several empty like spaces intermixed with fibrous matrix, including chondrons surrounded by a fine network of small capillaries (d, arrows). Cartilage matrix was invaded by blood vessels with a well- defined lumen (e) and cartilage canals filled with loose connective tissue and small capillaries (f).*

Table III. *Correlations between the collected data*.

Parameter		Mean surface area (M.S.A.)	Maximum M.S.A.	M.S.A. deviation	JOAS	Lumbar VAS	Pelvic limb VAS	Pffirrmann grading	Modic grading	Age
Mean surface	Pearson	1	0.808**	0.960**	–0.358*	0.376*	0.085	0.505**	0.500**	0.456**
area (M.S.A.)	Correlation r		*p*=0.000	*p*=0.000	*p*=0.032	*p*=0.024	*p*=0.623	*p*=0.002	*p*=0.002	*p*=0.005
	Kendall's tau-b	1	0.702**	0.781**	–0.279*	0.221	0.059	0.289*	0.331*	0.351**
	Spearman's rho	1	0.866**	0.934**	–0.380*	0.304	0.085	0.365*	0.419*	0.463**
Maximum	Pearson	0.808**	1	0.732**	–0.427**	0.266	0.142	0.432**	0.444**	0.270
M.S.A.	Correlation	*p*=0.000		*p*=0.000	*p*=0.009	*p*=0.117	*p*=0.409	*p*=0.008	*p*=0.007	*p*=0.111
	Kendall's tau-b	0.702**	1	0.735**	–0.299*	0.183	0.106	0.309*	0.343**	0.245*
	Spearman's rho	0.866**	1	0.741**	–0.430**	0.240	0.135	0.388*	0.434**	0.368*
M.S.A. deviation	Pearson	0.960**	0.732**	1	–0.295	0.457**	–0.006	0.501**	0.468**	0.463**
	Correlation	*p*=0.000	*p*=0.000		*p*=0.081	*p*=0.005	*p*=0.971	*p*=0.002	*p*=0.004	*p*=0.004
	Kendall's tau-b	0.781**	0.735**	1	–0.239*	0.297*	–0.004	0.253	0.327*	0.370**
	Spearman's rho	0.934**	0.741**	1	–0.321	0.406*	–0.009	0.313	0.397*	0.496**
JOAS	Pearson	–0.358*	–0.427**	–0.295	1	–0.411*	–0.220	–0.381*	–0.490**	–0.210
	Correlation	*p*=0.032	*p*=0.009	*p*=0.081		*p*=0.013	*p*=0.197	*p*=0.022	*p*=0.002	*p*=0.220
	Kendall's tau-b	–0.279*	–0.299*	–0.239*	1	–0.242	–0.236	–0.275*	–0.365**	–0.157
	Spearman's rho	–0.380*	–0.430**	–0.321	1	–0.304	–0.289	–0.330*	–0.440**	–0.182
Lumbar VAS	Pearson	0.376*	0.266	0.457**	–0.411*	1	–0.112	0.396*	0.260	0.341*
	Correlation	*p*=0.024	*p*=0.117	*p*=0.005	*p*=0.013		*p*=0.514	*p*=0.017	*p*=0.126	*p*=0.042
	Kendall's tau-b	0.221	0.183	0.297*	–0.242	1	–0.125	0.208	0.124	0.269*
	Spearman's rho	0.304	0.240	0.406*	–0.304	1	–0.165	0.245	0.146	0.342*
Pelvic limb VAS	Pearson	0.085	0.142	–0.006	–0.220	–0.112	1	0.191	0.116	–0.410*
	Correlation	*p*=0.623	*p*=0.409	*p*=0.971	*p*=0.197	*p*=0.514		*p*=0.264	*p*=0.500	*p*=0.013
	Kendall's tau-b	0.059	0.106	–0.004	–0.236	–0.125	1	0.152	0.118	–0.275*
	Spearman's rho	0.085	0.135	–0.009	–0.289	–0.165	1	0.175	0.130	–0.354*
Pffirrmann grading	Pearson	0.505**	0.432**	0.501**	–0.381*	0.396*	0.191	1	0.686**	0.354*
	Correlation	*p*=0.002	*p*=0.008	*p*=0.002	*p*=0.022	*p*=0.017	*p*=0.264		*p*=0.000	*p*=0.034
	Kendall's tau-b	0.289*	0.309*	0.253	–0.275*	0.208	0.152	1	0.615**	0.242
	Spearman's rho	0.365*	0.388*	0.313	–0.330*	0.245	0.175	1	0.658**	0.321
Modic grading	Pearson	0.500**	0.444** 7	0.468**	–0.490**	0.260	0.116	0.686**	1	0.370*
	Correlation	*p*=0.002	*p*=0.00	*p*=0.004	*p*=0.002	*p*=0.126	*p*=0.500	*p*=0.000		*p*=0.026
	Kendall's tau-b	0.331*	0.343**	0.327*	–0.365**	0.124	0.118	0.615**	1	0.305*
	Spearman's rho	0.419*	0.434**	0.397*	–0.440**	0.146	0.130	0.658**	1	0.395*
Age	Pearson	0.456**	0.270	0.463**	–0.210	0.341*	–0.410*	0.354*	0.370*	1
	Correlation	*p*=0.005	*p*=0.111	*p*=0.004	*p*=0.220	*p*=0.042	*p*=0.013	*p*=0.034	*p*=0.026	
	Kendall's tau-b	0.351**	0.245*	0.370**	–0.157	0.269*	–0.275*	0.242	0.305*	1
	Spearman's rho	0.463**	0.368*	0.496**	–0.182	0.342*	–0.354*	0.321	0.395*	1

**Correlation is significant at the 0.01 level (2-tailed). *Correlation is significant at the 0.05 level (2-tailed). VAS: Visual Analogic Scale. JOAS: Japanese Orthopaedic Association Score. M.S.A.: Mean surface area.

Table IV. *Partial correlation between Mean Surface Area, JOAS, Pfirrmann grading and Modic classification controlling for age*.

Control	Variables		M.S.A	Max. Area	JOAS	Pffirrmann	Modic
Age	Mean surface area (M.S.A.)	Correlation	1.000	0.800**	–0.302	0.413*	0.400*
		Significance (2-tailed) *p*	0.	0.000	0.078	0.014	0.017
	Maximum Area	Correlation	0.800**	1.000	–0.393*	0.374*	0.384*
		Significance (2-tailed) *p*	0.000	0.	0.019	0.027	0.023
	JOAS	Correlation	–0.302	–0.393*	1.000	–0.336*	–0.454**
		Significance (2-tailed) *p*	0.078	0.019	0.	0.048	0.006
	Pffirrmann grading	Correlation	0.413*	0.374*	–0.336*	1.000	0.639**
		Significance (2-tailed) *p*	0.014	0.027	0.048	0.	0.000
	Modic grading	Correlation	0.400*	0.384*	–0.454**	0.639**	1.000
		Significance (2-tailed) *p*	0.017	0.023	0.006	0.000	0

**Correlation is significant at the 0.01 level (2-tailed). *Correlation is significant at the 0.05 level (2-tailed). JOAS: Japanese Orthopaedic Association Score. M.S.A.: Mean surface area.

Results, after performing a partial correlation test controlling for age (Table IV) between mean surface area of chondrons and JOAS, showed no correlation (r=–0.302, p=0.078); in the same partial correlation test, mean surface area of chondrons correlated with Pfirrmann grading (r=0.413, p=0.014) and with Modic scale (r=0.400, p=0.017), but the correlation coefficient was weaker. There is a strong correlation between the maximum area size of the chondron cluster (Table III) and JOAS (r=–0.427, p=0.009, tau=–0.299, rho=–0.430), Pfirrmann grading (r=0.432, p=0.008, tau=0.309, rho=0.388) and Modic grading (r=0.444, p=0.007, tau=0.343, rho=0.434); however, after performing a partial correlation controlling for age all the correlations became weaker (Table IV).

Discussion

The purpose of the present study was to establish a relationship between the morphometric characteristics of disc cells from intervertebral disc tissue biopsies obtained during surgery and the clinical and imagistic features of patients with degenerative disc disease as, to our knowledge, this is the first study to approach these issues. Among genetic, biologic and mechanical factors involved in the process of disc degeneration, the loss of the gel-like consistency of the NP matrix, due to decreased proteoglycan content, is recognized as a main factor. This phenomenon progresses towards fibrosis of NP, altered transmission of the intervertebral forces, damage to the annulus fibrosus (AF) and various degenerative processes. These findings are consistent with our results from histological analysis of HE-stained specimens. In early degeneration, cell proliferation is up-regulated in an attempt to combat progressive extracellular matrix (ECM) loss. Cell clusters arranged in chondrons try to restore matrix synthesis and mechanical properties. In our specimens, we observed many chondrons with a small and pale territorial matrix, whereas normal territorial matrix was observed only surrounding normal chondrocyte lacunas. It is reported that the proportion of clustered cells is similar both in AF and NP in the degenerated disc samples either herniated or not (21). This aspect, however, was difficult to evaluate because our samples consisted of herniated intervertebral disc material providing limited control over the regions of analysis. The tissue samples from prolapsed discs analyzed in this study contained numerous chondron clusters consisting of nests of cells in different evolution phases and of different sizes, surrounded by a reduced quantity of territorial matrix with an overall aspect of degeneration due to an inefficient and incomplete regeneration process. The presence of large number of chondrons, insufficient and disorganized extracellular matrix with clefts and fissures, necrotic areas, vascular and nervous structures suggests a process of "degeneration through regeneration". This process might be a response to a mechanical stress that forces the intervertebral disc to reorganize in order to withstand the new biomechanical forces. Regarding the clinical characteristics of the study sample, we measured a mean preoperative JOA score of 7.83 (range=1-13), which is lower than data from literature (22, 23), suggesting that the patients included in our study had more severe symptoms and functional disabilities than the ones in other studies. Modic endplate changes were detected in 32 cases (88.9%), more than in other reported studies (19, 20, 24, 25). Based on the mean preoperative JOAS of 7.83% and on Modic changes found in 88.9% of cases, we can conclude that our group of patients presented more severe degenerative changes than reported in the literature. Results from this study show that the area size of chondron clusters correlates with both clinical and imagistic characteristics. There is a correlation between the area size of the chondrons and JOAS that was used to evaluate clinical symptoms of patients with a herniated lumbar disc. We have to note that the variables taken into consideration correspond only to a number of patients with LDD. This group is represented by patients who have a surgical indication, thus having more severe symptoms and a more obvious degeneration hallmark in the intervertebral disc. This category of patients is important as a starting point for future investigations because subtle symptoms and degeneration changes are more difficult to quantify objectively and may alter the results. The area size of chondron clusters correlated only with pain as evaluated through lumbar VAS and not with pelvic limb VAS that might be due to the more subjective nature of these evaluation tools. The lack of correlation between pelvic limb VAS and area size of chondrons can be explained from the fact that every patient that has to undertake surgery, often has intense pain in the lower limb and, thus, the VAS method cannot be properly used in this matter. However, in order to properly evaluate a candidate for surgery more objective evaluation tools such as JOAS or ODI (Oswestry Disability Index) are needed. In our study, we used JOAS, which is a reliable tool for evaluating clinical symptoms of patients with LDD (26). In this context, its correlation with morphometric changes of the intervertebral disc is of high clinical importance. Another finding is the correlation of the mean maximum area of chondrons with JOAS, Pfirrmann and Modic degeneration scales suggesting that the size of these chondrocyte-like cells aggregates definitely influence both biomechanics and symptoms. The correlation decreased when controlling for age indicating that age plays a lesser role in this relationship. This finding suggests that a possible hallmark of the pathological degeneration of the intervertebral disc is the appearance of large chondron clusters that have a double impact not only due to the incomplete matrix synthesis but also to the large volume of

optimal biomechanical matrix that this aggregation of cells dislocates. The strong correlation between imagistic classification scales utilized in this study has already been confirmed in the literature and, therefore, is not the subject of this study. Our results show a correlation between JOAS and Pfirrmann classification, although not as strong as the correlation between JOAS and Modic classification. JOAS is negatively correlated with Modic with a stronger coefficient (r=0.490, p=0.002) than that reported in the literature (27). This might be due to the limitations imposed by the small number of cases evaluated in this study.

Conclusion

Results from our current study demonstrate that our patient population had more severe degenerative changes, with Modic changes being present in 88.9% of cases and a mean JOAS of 7.83. Intervertebral disc cells tend to aggregate in clusters and their maximum size correlated with JOAS, Pfirrmann and Modic demonstrating a link between microscopic and clinical features of LDD. Further studies are required in order to relate the morphometric characteristic to an optimal time for surgical intervention.

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