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Prognostic Significance of Glutathione Peroxidase (GSH-Px) Levels in Lumbar Degenerative Disc Disease

Lomber Dejeneratif Disk Hastalığında Glutatyon Peroksidaz (GSH-Px) Düzeylerinin Prognostik Önemi

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ABSTRACT

Objective: In this study, the main aim was to research whether decreased antioxidant activity was manifesting the intervertebral disc degeneration in a population in which the degeneration could be compatible with their ages.

Material and Method: The study group consisted of 39 patients with lumbar disc degeneration (LDD) and 37 healthy controls. Patient data including age, symptoms, neurological examination findings, lumbar MRI findings, Oswestry Disability Index (ODI) scores, and Visual Analogue Scale of pain (VAS) were used. Human Glutathione peroxidase (GSH-Px) level was determined using the Enzyme-Linked Immunosorbent Assay (ELISA) method.

Results: Serum GSH-Px levels were significantly lower in the patients with LDD when compared to the healthy controls (p=0.011).

Conclusions: In the present study, we demonstrated that, in addition to environmental factors, there is a correlation between GSH-Px enzyme deficiency and lumbar disc degeneration.

Keywords: Glutathione peroxidase, lumbar degenerative disc disease, ELISA

ÖZ

Amaç: Bu çalışmanın temel amacı, yaş nedeniyle disk dejenerasyonu potansiyeli olan bir popülasyonda, azalmış antioksidan aktivitesinin intervertebral disk dejenerasyonu ile ilişkisini araştırmaktır.

Gereç ve Yöntem: Çalışmaya lomber disk dejenerasyonu (LDD) tanısı almış 39 hasta ve 37 sağlıklı gönüllü dahil edildi. Hasta verileri yaş, semptomlar, nörolojik muayene bulguları, lomber MRG bulguları, Oswestry Skala (ODI) skorları ve Visüel Analog Skala (VAS) kullanıldı. İnsan Glutatyon peroksidaz (GSH-Px) seviyeleri Enzime Bağlı İmmünosorbent Ölçüm (ELISA) yöntemi ile tespit edildi.

Bulgular: Serum GSH-Px düzeylerinin LDD tanısı almış hasta grubunda sağlıklı kontrollere göre anlamlı olarak düşük olduğu belirlendi (p=0,011).

Sonuç: Çalışmamızda çevresel faktörlere ek olarak GSH-Px enzim eksikliği ile lomber disk dejenerasyonu arasında bir korelasyon olduğu tespit edilmiştir.

Anahtar Kelimeler: Glutatyon peroksidaz, lomber dejeneratif disk hastalığı, ELISA

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INTRODUCTION

Intervertebral disc tissue is one of the earliest tissues in the human body to degenerate (1,2) and is one of the major pathological causes of chronic pain and debility, especially in the elderly population. The decrease of hydration in disc content, loss in the height of disc space, thinning, microfractures, ossification, and Modic changes in endplates and neighboring vertebral bodies are essential indicators of intervertebral disc degeneration (3-8). Such a degeneration evolves in every human being's intervertebral disc during the aging process, but, in some cases, it starts earlier and seems more like a pathological degenerative disease than an indication of aging (9-13).

Genetic predisposition, trauma, environmental factors, and smoking are other predisposing factors apart from aging in intervertebral disc degeneration (11,14-17). Glutathione, a substantial antioxidant peptide in the human cytoplasm, plays a crucial role in protecting cells from oxidative damage (18). Glutathione peroxidase (GSH-Px) belongs to an antioxidant enzyme family that has peroxidase properties. GSH-Px has the essential biological role of protecting the organism from oxidative damage. Lifelong reduction in GSH-Px was shown to increase lifespan and decrease age-related effects on pathology and increase tissues' sensitivity to oxidative stress-induced apoptosis (19).

In the present study, the main aim was to research whether the decreased antioxidant activity manifested the intervertebral disc degeneration in a population that the degeneration could be compatible with their ages.

MATERIAL AND METHOD

Study Population

In a total of 76 cases, our study group consisted of 39 lumbar disc degeneration (LDD) patients and 37 healthy controls. The study's patient population was composed of operated LDD patients who were admitted to the Neurosurgery Department of Yeditepe University. Patient data included: age, symptoms, neurological examination, lumbar MRI examination, Oswestry disability index (ODI) scores, and Visual Analogue Scale (VAS) of pain (20). The same neuroradiologist reported the lumber MRI results of patients and control groups. The Medical Faculty Ethics Committee approved the present study protocol at Yeditepe University. Informed consent was obtained from all participants included in the study.

The inclusion criteria for patients in the present study were having lower back pain and radiculopathy and identification of lumbar disc herniation in the lumbar MRI. The exclusion criteria for patients were history or radiodiagnosis of infection, vertebral fractures or spinal deformities, spondylolisthesis, oncologic diseases, congenital anomalies, and osteoporosis. Erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP) were checked to exclude inflammatory diseases. In the patient group, the severity of pain was analyzed using the Visual Analogue Scale (VAS) of pain scores and the Oswestry Disability Index (ODI) (20).

In the control group, the individuals who had lumbar MRI scans reported as normal for nonspecific low back pain. They also had laboratory analysis included CRP and ESR results as normal levels. The demographic data information of the patients was evaluated using the patient data system (Table 1).

In the patient group, the blood samples were collected before the surgery. After the centrifugation process, separated serum samples were frozen and stored at -80°C. The collected samples were analyzed once a week, for a total of 60 days.

Determination of Glutathione Peroxidase Enzyme Levels

GSH-Px was determined by the two-site sandwich Enzyme-Linked Immunosorbent Assay (ELISA) method. Serum GSH-Px level was measured by Human GSH-Px ELISA Kit (Abbkine Scientific Co., Ltd., Wuhan, Hubei, China). The serum GSH-Px levels were determined by spectrophotometrically.

Statistical Analyses

Statistical analyses were performed by the IBM SPSS version 23 Statistics program (SPSS Inc, Chicago, Illinois, USA). The statistical significance was determined at a p-value <0.05. A Chi-square analysis was performed, and the values were expressed in numbers and percentages. The student t-test was used to compare the quantitative parameters in the patient and control groups. The results were expressed in means±standard deviations.

RESULTS

The present study included a total of 76 individuals as 39 patients with LDD and 37 healthy controls. The demographic data of all cases are presented in Table 1. The mean age of patients was 43±10.34, and controls were 45.43±10.01, and there was no statistically significant difference between the groups (p=0.301). The examination of patients' gender distribution

Table 1. Demographic information of patient and control groups.

	Control Group (n= 37)	LDDD (n= 39)	P value
Age (years) (Mean±SD)	45.43±10.01	43±10.34	0.301
GENDER (Male /Female)	23(62.2%) /14 (37.8%)	14 (35.9%) / 25(64.1%)	0.022*

* statistically significant difference; LDDD: Lumbar disc degeneration disease, SD: Standart deviation

Table 2. The patients mean VAS and ODI scores.

Score	Minimum	Median	Maximum
Oswestry	26	66	94
VAS	60	70	100

showed that 35.9% were male, and 64.1% were female. The ratio of males and females in the control group were 62.2% and 37.8%, respectively. The gender distribution was significantly different between the groups. The risk of developing LDD was 2.9 times higher in females compared to males (p=0.022). Also, 12 had a lumbar MRI scanning before the diagnosis and a history of low back pain on at least one occasion. Of these 39 patients, 16 had a black disc, and 23 had a lumbar disc herniation. The patient's mean VAS and ODI scores are presented in Table 2.

The serum GSH-Px levels were statistically significantly lower in the patient group than the controls (p=0.011). The serum GSH-Px levels of the study groups are presented in Figure 1.

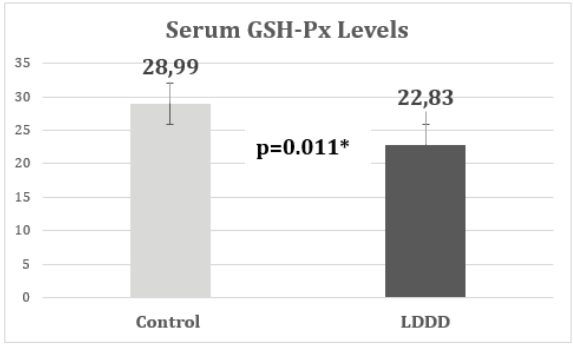

Figure 1. Serum GSH-Px levels.

DISCUSSION

Although intervertebral disc degeneration is a normal part of the aging process, in recent years, some indicators have shown that some genetic predispositions and environmental factors, such as smoking and chronic injuries, could also play a part (11, 14-16, 21). Typically, disc tissue, the counterpart to the intervertebral disc space, is mostly devoid of vascular tissue, so blood supply is mainly with diffusion from the outer layer of the annulus fibrosis and vertebral endplates (22). Even this reality is one of the critical factors, regeneration of such tissue would be difficult without an optimal vascular supply. This is such an irony that one of the most important suspension points of the body, the "intervertebral disc tissue" exposed to mobility, erected posture, and loading stresses, discloses a weak point against secondary insults of movement-related trauma. Working under traumatic stress cause limited tissue perfusion during whole life-time, so degeneration of vertebral disc space can be accepted as a normal consequence. However, modern underlying molecular mechanisms playing roles in this degeneration point outs multiple clues (21).

As in all aging processes, genomic instability, epigenetic alterations, mitochondrial dysfunction, disorders in intercellular communication, accumulation of damaged molecules, disorders in cellular response, and dysfunction of biological responses may play essential roles in degeneration (21,23). This molecular mechanism set thinking that some unknown disorders in this metabolic mechanism may aggravate disc degeneration, and these may intern leads to earlier pathologic clinically detected disorders in our clinical practice as lumbar disc herniation and degenerative spinal disorders with stenosis.

Programmed cell death and degeneration of extracellular matrix-induced oxidative stress participate in the pathophysiology of disc degeneration. Vo et al. reported that the healthy intervertebral disc's significant characteristic is an absence of vascularization. Reactive Oxygen Species (ROS) are produced as a result of oxidative phosphorylation caused by hypoxic conditions in disc cells (24). Dimozi et al. investigated that oxidative stress could trigger premature senescence and reduce cell proliferation in the aged intervertebral disc (25). Also, the catabolic processes of these senescent cells could contribute to the degeneration of the tissue.

The GSH-Px biochemical function reduces lipid hydroperoxides to their corresponding alcohols and hydrogen peroxide to water. The effect of GSH-Px on various diseases such as celiac disease, coronary heart disease, diabetic nephropathy, and vitiligo has been studied previously (26-29).

Yang et al. (27) investigated that GSH-Px plays an essential role in oxidative stress-induced disc degeneration in human nucleus pulposus cells. In this study, they claim that the antioxidative effect of GSH-Px could be a candidate marker in reduced oxidative stress in disc degeneration.

Our study is the first study to investigate the effect of GSH-Px on lumbar disc herniation patients. In comparison between patient and control groups in similar age groups, GSH-Px levels were decreased significantly in the lumbar disc degeneration group compared to the healthy controls (p=0.011). This suggests that GSH-Px enzyme deficiency might correlate with degeneration of lumbar disc tissue and lumbar disc herniation in addition to environmental factors. Anti-oxidant mechanisms seem to play a significant part in lumbar disc herniations. In light of these results, if a biomarker could be found for the deficiency of anti-oxidant mechanisms, diagnosis of lumbar degenerative diseases could be made earlier, and treatment with anti-oxidant therapies could prevent patients from surgery.

CONCLUSION

The present study showed that there is a correlation between GSH-Px enzyme deficiency and lumbar disc degeneration in addition to environmental factors. However, there are some limitations to our study. Namely, the number of individuals should be increased in order to confirm these results. Moreover, degenerative spinal diseases like spinal stenosis, spondylolisthesis, and spondylosis could be added and compared with our results to work out correlations between all lumbar degenerative spine diseases for enzyme levels.

Ethical Approval: All procedures in this study was accordance with the ethical standards of the 1975 Declaration of Helsinki guidelines and its later amendments. The present study protocol was approved by The Yeditepe University Medical Faculty Ethics Committee. Informed consent was obtained from all participants included in the study.

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Conflict of interest: The authors declare that they have no conflict of interest.

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