RESEARCH ARTICLE / *Araştırma Makalesi*

Total Antioxidant Levels in Patients with Spinal Herniation

Spinal Herniasyon Hastalarının Toplam Antioksidan Seviyeleri

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Abstract

Objective	Inflammatory response has been acknowledged to be an important factor in the process of disc degeneration and may play an important role in generation of pain, among patients with spinal herniation. The aim of this study is to investigate the total antioxidant status (TAS), total oxidant status (TOS), and their ratio, oxidative stress index (OSI) in patients with spinal herniations, and the relationship between these dependent variables and location of herniation, gender and age.
Materials and Methods	19 female (48,72%) and 20 male (51,28%) patients are included in the study. TAS and TOS levels are measured by commercially available kits in laboratory (Rel Assay Diagnostics, Turkey). OSI levels are calculated by their ratio.
Results	Results show that mean TAS level is 1,518 ± 0,176, mean TOS level is 3,168 ± 0,956, and mean OSI level is 0,222 ± 0,078. No correlations are found between age with the TAS, TOS and OSI levels.
Conclusion	Post-operative patients with spinal herniation have normal oxidative statuses and the location of herniation and gender seems to have no statistically significant effect on the TAS, TOS and OSI levels.
Keywords	Spinal herniation; antioxidant; free radical; oxidative stress.

Öz

Amaç	*İnflamatuar yanıt disk bozulmalarında görülen önemli bir faktör olarak kabul görmüştür ve spinal herniasyon hastalarında acı hissinin ortaya çıkmasında önemli bir rolü olabilir. Bu çalışmanın amacı, cerrahi tedavi görmüş spinal herniasyon hastalarındaki toplam antioksidan statüsü (TAS), toplam oksidan statüsü (TOS) ve oranlanmalarından ortaya çıkan oksidatif stress endeksinin (OSE) ölçülmesi ve bu değerlerin toplanan yaş, cinsiyet ve herniasyon bölgesi arasında ilişki olup olmadığının araştırılmasıdır.*
Gereç ve Yöntemler	*19 kadın (48,72%) ve 20 erkek (51,28%) hasta çalışmaya dahil edilmiştir. TAS ve TOS seviyeleri laboratuvarda bulunan kitlerle ölçülmüş ve OSE bu değerlerin oranlanmasıyla elde edilmiştir (Rel Assay Diagnostics, Türkiye).*
Bulgular	*Sonuçlar spinal herniasyonlu hastalarda ortalama TAS seviyesinin,1,518 ± 0,176, ortalama TOS seviyesinin 3,168 ± 0,956, ve ortalama OSE değerinin 0,222 ± 0,078 olduğunu göstermektedir. Demografik ve herniasyon bölgesi değişkenlerinin TAS, TOS ve OSE değerleri üzerinde bir etkisi bulunamamıştır.*
Sonuç	*Cerrahi tedavi görmüş spinal herniasyon hastalarının normal oksidatif durumda oldukları ve diğer değişkenlerin bu bulgu üstünde istatistiksel açıdan anlamlı bir etkisi olmadığı bulunmuştur.*
Anahtar Kelimeler	*Spinal herniasyon; antioksidan; serbest radikal; oksidatif stres.*

INTRODUCTION

Spinal disc herniations are the displacement of disc material (nucleus pulposus or annulus fibrosis) beyond the intervertebral disc space.[1] They are common diseases that affect all people around the world without any exception for gender, age, religion, country etc. For example, lumbar disc herniation affects around 9% of all people worldwide.[2] Nonetheless, there are risk factors known to be associated with spinal disc herniations such as obesity, working conditions, smoking, genetics and family history.[3-6] For example, Muramatsu and colleagues found the prevalence of lumbar disc herniation is high in civil servants in China (44.8%).[1]

Spinal herniations are highly associated with the inflammatory response and the inflammation is associated with adverse symptoms related to the stimulation of nerve fibers which in turn causes back pain generation.[2] Although the role of inflammation is not yet fully known, many inflammatory mediators are identified in relation with disc herniation associated radiculopathy.[7]

The treatment for spinal disc herniation and other degenerative disc diseases may be categorized into surgical versus conservative approaches.[2] The decision for surgery depends on the severity of the herniation. For less severe cases non-surgical approaches can be considered, but many non-surgical treatments for disc herniations are considered to be ineffective or safe. As an alternative Xiao and colleagues suggested the use of curcumin, which has exceptional anti-inflammatory profile, could alleviate lumbar radiculopathy by attenuating neuroinflammation, oxidative stress and nociceptive factors. Furthermore, Yang and colleagues reported antioxidant drugs as nanofullerol prevent the degeneration of the disc tissue.[8] The use of anti-inflammatory properties underlines the active role of inflammation in spinal disc herniation.[9]

Normal aerobic metabolism produces free radicals such as reactive oxygen species (ROS) and the reactive nitrogen species are generated various endogenous systems, and by exposure to different physiochemical conditions or pathological states.[10-11]

A balance between free radicals and antioxidants is a necessity for proper physiological functioning.[12] When free radicals overwhelm the body's ability to regulate them, a condition known as oxidative stress (OS) ensues.[13] OS is not considered to be a disease itself, but may be the cause or by-product of some diseases such as radiculopathy. Thus, the biomarkers of OS are useful for understanding the disease status. The ROS to total antioxidant capacity (TAC) has been used as a measure of oxidative stress in various fields of medicine, and becoming increasingly popular.[14-17]

The aim of this study is to investigate the total antioxidant status (TAS), total oxidant status (TOS), and their ratio, oxidative stress index (OSI) in patients with spinal herniations, and the relationship between these dependent variables and location of herniation, gender and age.

MATERIALS and METHODS

This study is a descriptive research based on a cross-sectional design. The ethics committee's approval was granted by T.C. Biruni University's ethics committee with the decision number 2020/40-13, on 28th of May, 2020.

Participants

Between 15th of September, 2018 and 15th of April, 2020, 42 patients were operated for spinal herniation in Neurosurgery Clinic of Medicana Bahçelievler Hospital. All patients' surgeries are performed by the same author. 3 patients' data are not available due to failure of communication. Thirty-nine patients, 19 female (48,72%) and 20 male (51,28%), are included in the study. The ages of the patients ranged between 27 and 71. All patients' blood samples are taken in Biochemistry Laboratory of Medicana Bahçelievler Hospital. Blood samples are kept in 0 degrees Celsius, and all tests are examined in Rel Assay

Diagnostics Laboratory.

Total Antioxidant Status (TAS)

TAS levels were measured using commercially available kits (Rel Assay Diagnostics, Turkey). The novel automated method is based on the bleaching of characteristic color of a more stable ABTS (2,2′ - Azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)) radical cation by antioxidants. The assay has excellent precision values, which are lower than 3%. The results were expressed as mmol Trolox equivalent / L.[18]

Total Oxidant Status (TOS)

TOS levels were measured using commercially available kits (Rel Assay Diagnostics, Turkey). In the new method, oxidants present in the sample oxidized the ferrous ion-o-dianisidine complex to ferric ion. The oxidation reaction was enhanced by glycerol molecules abundantly present in the reaction medium. The ferric ion produced a colored complex with xylenol orange in an acidic medium. The color intensity, which could be measured spectrophotometrically, was related to the total amount of oxidant molecules present in the sample. The assay was calibrated with hydrogen peroxide and the results were expressed in terms of micromolar hydrogen peroxide equivalent per liter (μol H2O2 equivalent / L).[19]

Oxidative Stress Index (OSI)

The ratio of TOS to TAS was accepted as the Oxidative Stress Index (OSI). For calculation, the resulting unit of TAS was converted to μol / L, and the OSI value was calculated according to the following Formula: OSI (arbitrary unit) = TOS (μol H2O2 equivalent / L) / TAC (μol Trolox equivalent / L).

RESULTS

Results show that mean TAS level is 1.518 ± 0.176, mean TOS level is 3.168 ± 0.956, and mean OSI level is 0.222 ± 0.078. The descriptive statistics for TAS, TOS and OSI are presented (Table 1).

Table 1. Descriptive Statictics for LoH, TAS, OSI

LoH	TAS-1 mmol/L	TAS-1 mmol/L	OSI
Lomber: 23	Min.: 1.033	Min.: 0.249	Min.: 0.059
Cervical: 16	1st Qu.: 1.253	1st Qu.: 2.443	1st Qu.: 0.151
	Median: 1.551	Median: 2.878	Median: 0.218
	Mean: 1.518	Mean: 3.168	Mean: 0.222
	3rd Qu.: 1.732	3rd Qu.: 3.923	3rd Qu.: 0.259
	Max.: 1.987	Max.: 8.815	Max.: 0.796

LoH: Location of Herniation, TAS: Total Antioxidant Status, TOS: Total Oxidant Status, OSI: Oxidative Stress Index, Min: Minimum, Max: Maximum, 1st Qu: 1st Quartile, 3rd Qu: 3rd Quartile.

The location of herniation differed as Lumbar and Cervical, 23 (58.97 %) and 16 (41.02 %), respectively. Below, the herniation of the patient number three is given (Figure 1).

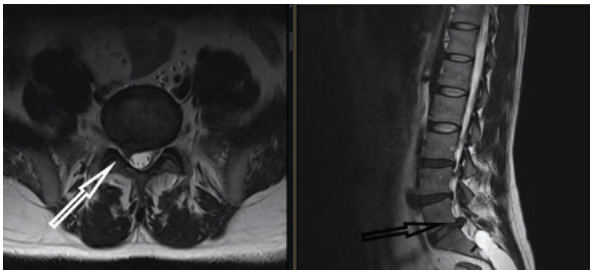

Figure 1. The herniation of the patient number three. Axial and Sagittal T2 sections of MRI. White and black arrows show right foraminal extruded herniated disc.

Pearson's product-moment correlation analyses are run in order to examine possible relation of age and the dependent variables. No correlation is found between age and OSI (r (37) = -0,115, p = 0,48). No correlation was found between age and TAS (r (37) = 0,001, p = 0,992). No correlation was found between age and TOS, (r (37) = -0,116, p = 0,48).

Two sample t-tests are performed to determine whether the Location of Herniation has any effect on the dependent variables. Patients with lumbar herniation (M = 1,54, SD = 0,295) and patients with cervical herniation (M = 1,47, SD = 0,237) had no difference in TAS levels (t (37) = 0,764, p

= 0,449). Patients with lumbar herniation (M = 3,28, SD = 1,543) and patients with cervical herniation (M = 2,99, SD = 1,402) had no difference in TOS levels (t (37) = 0,618, p = 0,540). Patients with lumbar herniation (M = 0,22, SD = 0,137) and patients with cervical herniation (M = 0,22, SD = 0,094) had no difference in OSI levels (t (37) = -0,087, p = 0,930).

Two sample t-tests are performed to determine whether the gender has any effect on the dependent variables. Female patients (M = 1,56, SD = 0,302) and male patients (M = 1,47, SD = 0,239) had no difference in TAS levels (t (37) = -0,954, p = 0,346). Female (M = 3,03, SD = 1,205) and male patients (M = 3,30, SD = 1,714) had no difference in TOS levels (t (37) = -0,582, p = 0,564). Female (M = 0,22, SD = 0,088) and male patients (M = 0,21, SD = 0,147) had no difference in OSI levels (t (37) = 0,254, p = 0,800).

DISCUSSION

Spinal disc herniations are the displacement of disc material (nucleus pulposus or annulus fibrosis) beyond the intervertebral disc space.[1] They are common diseases that affect many people, especially those who are working in physically demanding jobs. For example, lumbar disc herniation affects around 9% of all people worldwide.[2] Many risk factors are known to be at play.[3-6] The pain caused by spinal disc herniation may decrease the quality of life for the patients experiencing it.

The relationship between the spinal disc herniations and the inflammatory response is well-established. The inflammatory response is important in the process of disc degeneration and may play an important role in pain generation. Not only the inflammatory mediators are identified, but also the substances and drugs with anti-inflammatory properties are suggested as treatments for spinal disc herniations.

In this study, we present the post-operative TAS, TOS, and consequently OSI scores for patients who underwent spinal disc herniation surgery with gender, age and location of herniation variables. It is well-known that our body's aerobic metabolism produces free radicals such as reactive oxygen species (ROS) normally. However, diseases such as spinal disc herniations may increase the ROS production abnormally. This, in turn, increases the oxidative stress (OS) in the body. Inversely, after a surgical operation to treat the spinal disc herniations is performed, the OS should be in normal range. Our results are in accordance with this prediction. Our patients' post-operative OSI scores are ranging between 0,059 and 0,796 with a mean of 0,22. There are no signs of a heightened inflammatory response caused by the spinal disc herniations.

This study is valuable for the understanding of inflammatory responses caused by spinal disc herniations, and their post-operative results. However, this study is limited by the fact that we don't have the pre-operative measurements for TAS, TOS and OSI scores. Thus, the design of this study is primarily descriptive rather than causal. A controlled clinical trial with a control group would enable us to make comparisions and causal claims about whether the quantative change was really caused by the surgical operation. In our future studies, we plan to take pre-operative blood samples for those variables and include a control group.

CONCLUSION

The current study examines the post-operative patients with spinal disc herniations' oxidative and antioxidative statuses. These patients have normal antioxidant statuses and the location of herniation and gender seem to have no effect on this finding. However, we recommend future studies to also measure pre-operative oxidative and antioxidative scores in order to claim that this effect is caused by the surgical operation itself.

The ethics committee's approval was granted by T.C. Biruni University's ethics committee with the decision number 2020/40-13, on 28th of May, 2020.

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