Evaluation of Oxidants and Antioxidants in Scorpion Envenomation

Akrep Sokmalarında Oksidant ve Antioksidantların Değerlendirilmesi

Behçet AL,[a]
Pınar YARBİL,[b]
Suat ZENGİN,[a]
Seyithan TAYSI,[c]
Mustafa ÖRKMEZ,[c]
Cuma YILDIRIM,[a]
Seval KUL[d]

Departments of
[a]Emergency Medicine,
[c]Biochemistry,
[d]Biostatistic,
Gaziantep University
Faculty of Medicine,
[b]Clinic of Emergency,
Gaziantep Sehitkamil State Hospital,
Gaziantep

Geliş Tarihi/*Received:* 04.12.2012
Kabul Tarihi/*Accepted:* 25.03.2013

Yazışma Adresi/*Correspondence:*
Behçet AL
Gaziantep University
Faculty of Medicine,
Department of Emergency, Gaziantep,
TÜRKİYE/TURKEY
behcetal@gmail.com

ABSTRACT Objective: The purpose of this study is to evaluate the variations of pre- and post-treatment antioxidant status (TAS) and oxidant status (TOS) levels in scorpion envenomation, and of oxidative stress index (OSI) calculated with these levels. **Material and Methods:** Forty-four cases of scorpion envenomation who applied to Gaziantep University Medical Faculty Emergency Clinic between May 2009 and October 2010, and a control group of 20 volunteers, were enrolled to the study. Whole blood, biochemistry, coagulation parameters, and blood samples for toxin-antioxidant (TAS-TOS) study were obtained from patients (at the time of arrival and at the next control) and the control group. Erythrocyte packages were prepared for TAS-TOS study. Serum, plasma and erythrocyte packages were prepared for each patient at the first application and at the control after one month, and stored under -80o in the freezer. **Results:** No correlation was observed between age and gender and the levels of TAS, TOS, or OSI. TAS, TOS, and OSI levels in the first application were higher than the control levels taken one month later. TAS, TOS, and OSI arrival levels were higher than healthy control group levels. In patients who received scorpion serum, both arrival and control levels of TAS, TOS, and OSI were higher than those of patients who did not receive scorpion serum. TAS, TOS, and OSI levels in patients who were stung at more then one site were higher. **Conclusion:** Patients who received or did not receive scorpion antivenom were improved, and TAS, TOS, and OSI levels regressed. The significant increase in TOS and OSI levels in patients who received scorpion serum was not experienced in TAS levels.

Key Words: Scorpions; antioxidants

ÖZET Amaç: Bu çalışmada amacımız akrep sokmalarında tedavi öncesi ve sonrası antioksidan status (TAS), oksidan status (TOS) değerlerini ve bu değerler eşliğinde hesaplanan oksidatif stres indeksindeki (OSİ) değişimleri değerlendirmektir. **Gereç ve Yöntemler:** Mayıs 2009 ve Ekim 2010 tarihleri arasında Gaziantep Üniversitesi Tıp Fakültesi Acil Tıp Kliniğine başvuran 44 akrep sokması vakası ve gönüllü 20 kişilik kontrol grubu çalışmaya alındı. Hastalardan (geliş anı ve bir sonraki kontrolde) ve kontrol grubundan tam kan, biyokimya, koagülasyon parametreleri ile beraber toksin-antioksidan (TAS-TOS) çalışması için kan örnekleri alındı. TAS-TOS çalışması için eritrosit paketleri oluşturuldu. Serum, plazma, eritrosit paketleri her hasta için ilk başvuru ve bir ay sonra kontrolde olmak üzere hazırlanarak -80 derece dondurucuda saklandı. **Bulgular:** Yaş ve cinsiyet ile TAS, TOS, OSİ değerleri arasında korelasyon saptanmadı. İlk başvurudaki TAS, TOS, OSİ değerleri; bir ay sonraki kontrol değerlerinden daha yüksek idi. TAS, TOS, OSİ geliş değerleri; sağlıklı kontrol grubu değerlerinden yüksekti. Akrep serumu verilen hastalarda TAS, TOS ve OSI hem geliş hem kontrol değerleri akrep serumu verilmeyen hastalara göre daha yüksekti. Birden fazla yerden ısırılanlarda TAS, TOS ve OSİ değerleri daha yüksek saptandı. **Sonuç:** Akrep antivenomu kullanılan ve kullanılmayan hastalar iyileşmiş ve TAS, TOS, OSİ değerleri gerilemiştir. Akrep serumu verilen hastalarda TOS ve OSI değerlerinde sağlanan anlamlı artış TAS değerlerinde sağlanamamıştır.

Anahtar Kelimeler: Akrepler; antioksidanlar

doi: 10.5336/medsci.2012-32981

Turkiye Klinikleri J Med Sci 2013;33(4):1119-26

The scorpion is a invertebrate arthropod, and all types of scorpions are toxic. It is the hardest hunter to overwhelm among the animals that hunt at night. If the person stung is allergetic to this poison, a more dangerous conclusion is predicted.[1,2] Scorpions with powerful poison live in the Middle East, North Africa, India, Mexico **and Latin America**. The most lethal type known is the Buthidae family.[3] Annually over 100,000 cases of scorpion envenomation are recorded around the world, and about 800 deaths are reported, most of whom are children under six years of age.[4] The most lethal type of scorpion in Turkey and in the Gaziantep region in which we live is *Leiurus quinquestriatus* from the *Buthidae family*.[3] Excluding some transition metals (such as Fe^{3+}, Cu^{2+}, Mn^{2+} and Mo^{5+}) atoms, atom groups, or molecules that include some unpaired electrons are defined as free radicals.[5] Free radicals, although they have a very short life-cycle, are highly destructive to organisms due to their high activity. As long as there is a balance between the rate of occurrence and rate of annihilation for free radicals, the organism is not affected. When this balance is ruined in favor of oxidants, free radicals interact with biomolecules such as carbohydrates, lipids, proteins, and DNA and cause structural and metabolic alterations in cells. This results in tissue damage in organs that mostly have vital significance, primarily including heart, kidneys, liver, stomach, lungs, and brain.[6-8] The sources that form free radicals are classified into two groups, namely endogens (mostly mitochondrial electron transport chains) and exogens (such as cigarettes, pesticides, solvents, petrochemical products, drugs, alcohol, solar rays, stress, X-ray, and heavy physical activity).[9,10]

Antioxidants are classified into enzymmatic (superoxide dismutase, catalase, glutathione peroxidase, glutathione-s-transferase, etc.)[11] and non-enzymmatic (ascorbic acid, α-tocopherol, carotenoids, melatonin, etc.).[7,12] They impact by decreasing local oxygen concentration, preventing the initiation of lipid peroxidation by cleansing hydroxyl radicals, binding and deactivating transition metal ions, playing an active role in conversion of peroxides into non-radical products such as alcohol, and breaking the chain by reacting with all of the radicals that cause chain reactions. As a result, radical metabolite production is prevented, radicals produced are cleansed, cell damage is repaired, chain reactions that produce secondary radicals are stopped, and endogen antioxidant capacity is increased.[8]

In this study, our aim was to investigate the variations in pre- and post-treatment antioxidant status (TAS) and oxidant status (TOS) levels in scorpion stings, and oxidative stress index (OSI) levels calculated with these levels. As far as we have determined from the literature, this study is the first and most comprehensive clinical study in this area..

MATERIAL AND METHODS

This study complies with the Declaration of Helsinki Decisions, the Regulation of Patient Rights and ethical rules, and is approved by Gaziantep University Medical Faculty Ethical Committee no. 06–2009/257, dated June 06/2009. Forty-four cases of scorpion sting that applied to our emergency department between May 2009 and October 2010, with no history of cardiac failure, renal failure, chronic obstructive lung disease, diabetes mellitus, malignancy, or drug use, and a control group of 20 volunteers under the same conditions were enrolled in the study. All patients in the study group and in the control group were informed about the study, and their consents were taken in reply of their signs. The study was carried out prospectively. Arterial tension, pulse measurements, area of envenomation, consciousness, symptoms and signs at the first application and at control one month later, were recorded for each patient. Urea, creatinine, aspartate aminotansferase (AST), alanine aminotransferase (ALT), lactate dehydrogenase (LDH), creatine kinase (CK), MB fraction of creatine kinase (CKMB), electrolytes, white blood count, hemoglobin, platelets, coagulation parameters [activated partial thromboplastin time (aPTT), international normalized ratio (INR)], and also blood samples for toxin-antioxidant study, were taken from both patients and the control group. Two milliliter blood samples were taken into two

tubes, with heparin and without heparin, for the TAS-TOS study. Serum from the tube without heparin and plasma from the tube with heparin were resolved after the samples taken were centrifuged for five minutes at 5,000 cycles. Blood sample that remained in the tube with heparin was washed three times with 0.15 mL of 0.9% isotonic sodium chloride, and re-run at 10,000 cycles for 30 minutes. The plasma remaining in the tube was retaken and discarded. Thus, erythrocyte packages were formed. The same procedures were repeated on the patients one month later in the control. Serum, plasma, and erythrocyte packages were prepared for each patient at the first application and at the control after one month, and stored under -80° in the freezer. To measure TOS levels of the samples, we used the colorimetric method that depends on cumulative oxidation of ferrous ion to ferric ions for the oxidant molecules that the samples included.[13] The measurement method of the total antioxidant status level of the samples is based on the proportional decolorization of antioxidant molecules of the colorful radicals with the total concentrations of molecules as a result of the reduction of colorful 3-ethylbenzothiazoline-6-sulphonic acid (ABTS) cationic radicals by all antioxidant molecules of the sample, using Trolox as a calibrator, which is the water soluble analogue of vitamin E.[13] The results are expressed as equivalent to mmol Trolox. Oxidative stress index of the samples is defined as the percentage of TOS levels to the ratio of TAS of the samples. Before calculating, we converted the mmol levels of the TAS test units into micromole units, as in the TOS test.

For statistical evaluation, we used SPSS for Win. Ver. 18.0 (SPSS Inc., Chicago, Illinois, USA). While as the data were normally distributed, the data were expressed as mean±standard deviation, as the data were not normally distributed the data were expressed as median (Min-Max). As the data were normally distributed and dependent, groups were compared using a paired-sample t test. As the data were not normally distributed and independent, groups were compared using a Mann-Whitney U test. As the data were not normally distributed and dependent, groups were compared using a Wilcoxon test. When the data were not normally distributed and independent, Kruskal Wallis test was performed to evaluate the differences of TAS, TOS, OSI levels at the arrival on the sting region. The relationship between variables was analyzed using the Pearson Correlation Coefficient. In all comparisons, $p<0.05$ was considered statistically significant.

RESULTS

Half of the 44 patients who applied for scorpion envenomation were male, and the average age was 45.22±17.99. Of patients, 40.9% (n=18) were stung by a single scorpion on the upper extremity, 31.8% (n=14) were stung on the lower extremity. Twelve people were stung by multiple scorpions in multiple regions. Of patients, 59.1% (n=26) applied directly to the emergency department of Gaziantep University, 40.9% (n=18) were referred from other centers. All patients were conscious on arrival. Blood was taken from 68.2% (n=30) of the patients within the first four hours, from 31.8% patients within the second four hours, and so on. Twenty six patients received scorpion serum therapy, while 18 patients were followed without any scorpion serum. Of patients, 43.18% of the patients did not develop any symptoms; 22.72% had nausea-vomiting-dizziness, 11.36% had nausea-vomiting-dizziness-chest pain, 9.09% had nausea, 9.09% had vomiting, 2.2% had dizziness, and 2.2% had abdominal pain. 72.72% of the patients, 22.72% had severe pain and numbness in the extremity, and 4.5% had edema and severe pain in the extremity. All patients were followed in the emergency monitoring room. Grade 1 patients received analgesia and main support (0.9% isotonic), grade 2 patients received scorpion serum in addition to these therapies. None of our patients required intensive care.

We studied TAS/TOS counts from blood samples taken from the patients, and calculated OSI for each patient. There was no correlation between age and TAS, TOS, or OSI levels ($p>0.05$ for all). The difference in mean arrival TAS, TOS, and OSI levels for male and females were not statistically significant ($p>0.05$) (Table 1).

TAS, TOS, and OSI levels in the first application were higher than the control levels that were detected one month later ($p<0.05$) (Table 2).

TAS, TOS, and OSI arrival levels were higher than healthy control group levels and this difference was also statistically significant ($p<0.05$) (Table 3).

Early or late blood draws in scorpion envenomation did not significantly impact TAS, TOS and OSI levels ($p>0.05$) (Table 4).

Both arrival and control TOS levels and OSI arrival levels in patients who received scorpion serum were higher than that of patients that did not receive scorpion serum (Table 5).

The cardiac parameters, whole blood, and biochemical and coagulation levels of enrolled patients were compared, whereas glucose, urea, AST, ALT, CK, MB, platelet count, and aPTT levels recorded at arrival were significantly higher than the levels taken at the control ($p<0.05$), the increases in other levels were not significant ($p>0.05$) (Table 6). Although fasting and postprandial status of patients was not considered during blood draws, either at arrival or when they were called for control, control glucose mean level was lower than arrival mean level (Table 6).

TABLE 1: Comparison of gender and TAS, TOS, OSI levels in scorpion envenomations.

	Male (n=22)	Female (n=22)	p*
TAS-arrival-mean	1.76 (1.36-2.42)	1.66 (1.23-2.94)	0,177
TOS-arrival-mean	4.86 (2.30-26.45)	8.35 (2.36-36.41)	0,227
OSI-arrival-mean	0.31 (0.12-1.56)	0.54 (0.17-1.43)	0,080

* Mann-Whitney U test, All data were expressed as median (min-max).
TAS: Antioxidant status; TOS: Oxidant status; OSI: Oxidative stress index.

TABLE 2: Comparison of TAS, TOS and OSI levels at arrival time and one month later.

	Mean arrival level n=44	Mean level one month later n=44	p*
TAS	1.74±0.33	1.56±0.37	0.005
TOS	8.90±7.32	4.51±2.50	<0.001
OSI	0.55±0.40	0.36±0.24	<0.001

* Paired-sample t test, All data were expressed as mean±SD.
TAS: Antioxidant status; TOS: Oxidant status; OSI: Oxidative stress index.

TABLE 3: Comparison of TAS, TOS and OSI levels of the healthy control group with the patient values at arrival.

	Averages of arrival levels n=44	Averages of healthy control group n=20	p*
TAS	1.72 (1.03-2.94)	1.58 (1.30-1.98)	0.018
TOS	7.18 (2.30-32.41)	2.89 (1.82-6.62)	<0.001
OSI	0.46 (0.12-1.68)	0.15 (0.12-0.42)	<0.001

The data were expressed as median (min-max),* Mann-Whitney U test.
TAS: Antioxidant status; TOS: Oxidant status; OSI: Oxidative stress index.

TABLE 4: Comparison of TAS, TOS, OSI levels with the time of blood drawn.

	First 4 hours (n=26)	Second 4 hours and later then (n=18)	p*
TAS	1.74 (1.03-2.94)	1.68 (1.40-2.11)	0,563
TOS	6.62 (2.30-36.41)	7.64 (2.48-26.45)	0,840
OSI	0.41 (1.43-1.31)	0.53 (0.13-1.68)	0,504

* Mann-Whitney U test, All data were expressed as median (min-max).
TAS: Antioxidant status; TOS: Oxidant status; OSI: Oxidative stress index.

TABLE 5a: Comparison of TAS, TOS, OSI levels in patients that received or not received scorpion antivenom

Treatment	Arrival	Control	p*
Treated with scorpion serum (n=29)			
TAS	1.68 (1.03-2.94)	1.52 (1-2.46)	0.012
TOS	7.58 (2.30-36.41)	4.42 (1.86-11.84)	<0.001
OSI	0.47 (0.12-1.68)	0.36 (0.03-0.87)	0.023
Followed with a supportive treatment without scorpion serum (n=15)			
TAS	1.74 (1.23-2.11)	1.56 (0.98-1.43)	0.156
TOS	6.76 (2.36-16.92)	2.78 (1.85-8.97)	0.001
OSI	0.43 (0.13-0.88)	0.15 (0.01-0.88)	0.001

* Wilcoxon test, All data were expressed as median (min-max).
TAS: Antioxidant status; TOS: Oxidant status; OSI: Oxidative stress index.

When TAS, TOS, and OSI results were compared according to sting regions for scorpion envenomation, TOS and OSI arrival levels were higher in patients who applied with more than one envenomation, and this result was statistically significant ($p<0.05$) (Table 7). When TAS, TOS, and OSI arrival levels in patients with (n=19) electrocardiogram (ECG) modification (such as PR distance, UQTc, QTd, QpTc, Pmin) and in patients without ECG modification (n=25) were compared, no statistically significance difference was observed.

TABLE 5b: Comparison of differences in the levels of TAS, TOS, OSI in patients (in admission and control) that were treated or not treated with scorpion serum.

	Treated with scorpion serum (n=29)	Followed with a supportive treatment without scorpion serum (n=15)	p*
TAS	9.6 (-61:59)	8.3 (-26:37)	0.674
TOS	32 (-79:72)	42 (7:79)	0.720
OSI	2.2 (-180:83)	43 (0:95)	0.090

* Mann-Whitney U test, All data were expressed as median (min:max).
TAS: Antioxidant status; TOS: Oxidant status; OSI: Oxidative stress index.

TABLO 6: Comparison of mean values for blood biochemisty, coagulation, whole blood and cardiac parameters observed at arrival and at control one month later.

	Mean arrival level	Mean control level	p**
Glucose	116.65±36.82	97.68±12.72	0.002
Urea	35.36±17.56	20.77±6.14	<0.001
Creatinine	0.83±0.29	0.81±0.60	0.844
Sodium	136.65±21.16	140.19±2.53	0.274
Potassium	4.22±0.47	4.16±0.25	0.428
AST	28.36±24.8	17.93±5.63	0.003
ALT	20.13±13.04	14.40±6.11	<0.001
CK	150.56±145.19	101.1±215.34	0.041
CK MB	36.81±35.75	17.22±12.62	<0.001
Troponin	0.006±0.029	0.0009±0.006	0.195
Hemoglobin	13.80±1.4	13.85±1.54	0.780
Platelet	245.09±54.24	275.02±54.97	<0.001
INR	1.026±0.071	1.027±0.078	0.815
aPTT	13.47±1.43	11.79±0.93	<0.001
LDH	396.79±143.29	398.6±146.65	0.939

**Paired sample t test, All data were expressed as mean±SD.
AST: Aspartate aminotansferase; ALT: Alanine aminotransferase; CK: Creatine kinase; CK MB: MB fraction of creatine kinase; INR: International normalized ratio; aPTT: Activated partial thromboplastin time; LDH: Lactate dehydrogenase.

DISCUSSION

Scorpion venoms consist of a complex of several toxins that exhibit a wide range of biological properties and actions, as well as chemical compositions, toxicity, and pharmacokinetic and pharmacodynamic characteristics. These venoms are associated with high morbidity and mortality, especially among children.[14,15] Victims of envenoming by scorpions suffer a variety of pathologies, involving mainly both sympathetic and parasympathetic stimulation as well as central manifestations such as irritability, hyperthermia, vomiting, profuse salivation, tremor, and convulsion.[16] The clinical signs and symptoms observed in humans and experimental animals are related to an excessive systemic host inflammatory response to stings.[16] Under normal conditions, host cells are protected from the toxic effects of reactive oxygen species by enzymatic and non-enzymatic antioxidants. The marked increase in reactive oxygen species production during pathologic conditions, such as during acute and chronic inflammation, can overwhelm the body's defense mechanisms and lead to oxidative cell and tissue injury.[17] The term "oxidative stress" is adopted to describe any condition that results in an accumulation of free radicals that are deactivated by molecules known as antioxidants.[18]

In our study, all patients who applied for medical treatment due to scorpion stings were conscious at arrival. Of patients, 43.18% did not develop any symptoms, the remaining developed some or all of the following symptoms, nausea, vomiting, dizziness, chest pain, and abdominal pain. The most frequent sign was pain and paralysis. Major systemic findings such as severe cardiac signs and lung edema did not occur, and none of our patients required intensive care. In a 120-case prospective study performed by Al et al,[19] the most prevalent sign was localized pain, and patients did not develop severe systemic effects. This result was similar to the results of our study. Bouaziz et al[20] prepared a classification according to the presence or absence of systemic signs such as localized pain

TABLE 7: Comparison of TAS, TOS, OSI levels at arrival with the sting regions.

	Upper extremity and single scorpion n=18 (A)	Lower extremity and single scorpion n=14 (B)	More than one extremities and more than one scorpions n=12 (C)	p*	P**
TAS	1.67 (1.03-2.42)	1.73 (1.40-2.46)	1.65 (1.36-2.94)	0.891	
TOS	7,04 (2.36-25.26)	4.18 (2.30-18.91)	10.49 (2.48-36.41)	0.040	A-B 0.464
					A-C 0.087
					B-C 0.041
OSI	0.38 (0.12-1.3)[a]	0.29 (0.13-1.12)	0.78 (0.14-1.68)	0.033	A-B 1.000
					A-C 0.015
					B-C 0.031

*Kruskal-Wallis test, **Mann-Whitney U test, All data were expressed as median (min-max).
TAS: Antioxidant status; TOS: Oxidant status; OSI: Oxidative stress index.

in grade 1 scorpion stinging region, grade 2 systemic signs, cardiac findings or convulsion including grade 3 cardiogenic shock, pulmonary edema, or severe neurologic findings including coma. Although grade 3 signs did not occur in our study, 26 patients were evaluated as grade 2, and 18 patients as grade 1. The maximum follow-up period in emergency was calculated as 48 hours. Twenty-six of the patients received scorpion serum, 18 did not receive scorpion serum. Only symptomatic treatment was sufficient in patients who did not receive scorpion serum. In our study, no severe conditions such as lethal arrhythmia, cardiogenic shock, tamponade, or lung edema were observed.

In our study, no significant correlation was found between blood TAS-TOS-OSI levels of patients with or without ECG alteration ($p>0.05$ for all). This suggests that there is no significant correlation between the toxic effect of the scorpion poison and antioxidant, oxidant, or oxidative stress. In a compilation by Petricevich,[16] a study was mentioned regarding the structure of poisons obtained from various types of scorpions and the regions they affected in the body. According to this, alpha and beta toxins that affect sodium channels cause extension of the potential of action in nerves and muscles, and the presence of negative membrane potential. The same article suggested that potassium channel toxins varies according to type, and voltage-dependent potassium channels play a role in immune response, and cause T cell proliferation and IL–2 production. In addition, the article states that the blockage of potassium channels that are activated by calcium causes relative hyperkalemia and catecholamine release. It is observed that, in rats following scorpion envenoming, different proinflammatory and inflammatory cytokine releases occurs depending on the type, and as the severity of these symptoms increases so does the amount and variance of these cytokines.[21] The experimental studies emphasize that, in scorpion envenoming, immune response is initiated by nitric oxide (NO) and complement system.[22] NO, which is a free radical, is a crucial vasodilator and neurotransmitter. Insulin resistance plays a role in many serious effects, including septic shock and hypertension. According to another experimental study on rats stung by *Tityus* types, oxidant and antioxidant levels are not only related to membrane lipids, cellular proteins, nucleic acid function and integrity, but also cause signal production and gene expression in immune cells; whereas complement activation is directly related to tissue damage.[23] It is observed that, with the increase of LDH, AST, ALT, CK, CKMB, glucose 6 phosphate dehydrogenase rises in cardiac and liver effects.[24,25] Excessive release of catecholamines was detected in some animal models to increase glucagon, cortizol, thyroid hormone, and insulin secretion, and all of these increments cause hyperglycemia.[26] Another result of this study[26] is that insulin, which is an hormone with pleotropic effect, plays a remedial role in cardiovascular, hemodynamic, and neurologic symptoms. In other animal model studies, cytokines also had hyperglycemic effects.[27] In our study, when glucose, urea, AST, ALT, CK, CK MB, platelet

count, aPTT levels of patients' arrival whole blood, biochemistry, and coagulation parameters were compared to the control levels obtained one month later, they were significantly higher. Although fasting and postprandial status were not considered while taking blood, the mean arrival glucose level was significantly higher than the mean control glucose level. No increase or decrease occurred in the sodium and potassium levels of the patients.

According to the results of an animal experiment study by Dousset *et al*,[28] free radicals occur in scorpion poison toxicity, antioxidant substances are injected to rats concomitantly with venom, and toxicity does not decrease in repeated experimental envenoming. Only N-acetylcysteine (NAC) decreased venom toxicity. In our study, we observed that arrival TAS, TOS, and OSI levels were higher than control levels taken one month later. This result reveals that scorpion poison-dependent toxicity is present, and this cannot be efficiently taken under control by the body's own antioxidant system, which supports the study by Dousset et al.[28] New studies are required on strengthening the body's defense mechanism by administering antioxidants (such as NAC) against toxicity that occurs due to scorpion envenoming. TAS, TOS, and OSI levels of our patients were statistically higher than the levels of the healthy control group. If more clinically severe cases (grade 3) had occurred, then we suppose these results would be different. The statistically high levels of arrival TAS, TOS, and OSI compared to the control levels taken one month later reveal that the antioxidant system of the body easily and quickly tolerates toxicity due to grade 1 and 2 scorpion stinging. High levels of TAS taken following four hours, although statistically insignificant, suggest that antioxidant system activity is not sufficient. TOS and OSI arrival levels in patients who receive scorpion serum are significantly higher. However, the increase in TAS levels is not significant. This result reveals that scorpion serum does not effectively activate the antioxidant system and that, moreover, it may have an oxidative effect. As envenomation region and number of scorpions that sting increases, so does the response of the oxidant-antioxidant system.

LIMITATIONS OF THIS STUDY

The number of patients in our study is small. The absence of clinically grade 3 patients caused inability to evaluate TAS, TOS, and OSI levels in this patient group. In addition, the influence of scorpion serum on the oxidant-antioxidant system in grade 3 patients could not be investigated. It is a significant limitation that the scorpion antivenom we use is not specific to particular scorpions. TAS, TOS, and OSI levels a short time later than scorpion antivenom administration (24–48 hours) are unknown. Thus, the short-term effect of treatment with antioxidants in this study is unknown, and this should be investigated in further studies.

In conclusion, as we know, this is the first clinical study performed on a patient group regarding the comparison of TAS, TOS, and OSI levels. There is no correlation between age and gender in terms of TAS, TOS, and OSI levels in scorpion envenoming ($p>0.05$ for all). Patients who received or did not receive scorpion serum were improved and TAS, TOS, and OSI levels regressed. The significant increase in TOS and OSI levels in patients who received scorpion serum was not experienced in TAS levels. This suggests that scorpion antivenom may also be an oxidant, and its role in treatment is not effective. The effect, advantage, or harm of scorpion antivenom used in existing scorpion envenoming is disputable according to the results of our study. This study should be repeated with an antivenom exclusively against the poisons of scorpions living in the Southeastern Anatolian Region. When we consider cardiac parameters, there is no significant relation between cardiac influence and TAS, TOS, and OSI levels, and the cardiac effect of scorpion poison is not related to toxins.

REFERENCES

1. Sagheb MM, Sharifian M, Moini M, Sharifian AH. Scorpion bite prevalence and complications: report from a referral centre in southern Iran. Trop Doct 2012;42(2):90-1.
2. Harman M. [Bee and scorpion stings, and snake bites]. Turkiye Klinikleri J Surg Med Sci 2006;2(3):16-21.
3. Kartal M. [Scorpion envenomation]. Satar S, editör. Acilde Klinik Toksikoloji. Birinci Baskı. Adana: Nobel Yayınevi; 2009.p.603-9.
4. Tuuri RE, Reynolds S. Scorpion envenomation and antivenom therapy. Pediatr Emerg Care 2011;27(7):667-72; quiz 673-5.
5. Andriantsitohaina R, Duluc L, García-Rodríguez JC, Gil-del Valle L, Guevara-Garcia M, Simard G, et al. Systems biology of antioxidants. Clin Sci (Lond) 2012;123(3):173-92.
6. Sakaguchi S, Furusawa S. Oxidative stress and septic shock: metabolic aspects of oxygen-derived free radicals generated in the liver during endotoxemia. FEMS Immunol Med Microbiol 2006;47(2):167-77.
7. Agarwal A, Allamaneni SS. Free radicals and male reproduction. J Indian Med Assoc 2011;109(3):184-7.
8. Col C, Dinler K, Hasdemir O, Buyukasik O, Bugdayci G. Oxidative stress and lipid peroxidation products: effect of pinealectomy or exogenous melatonin injections on biomarkers of tissue damage during acute pancreatitis. Hepatobiliary Pancreat Dis Int 2010;9(1):78-82.
9. Braunersreuther V, Jaquet V. Reactive oxygen species in myocardial reperfusion injury: from physiopathology to therapeutic approaches. Curr Pharm Biotechnol 2012;13(1):97-114.
10. Abdollahi M, Ranjbar A, Shadnia S, Nikfar S, Rezaie A. Pesticides and oxidative stress: a review. Med Sci Monit 2004;10(6):RA141-7.
11. Sulthana SM, Kumar SN, Sridhar MG, Bhat BB, Rao KR. Antioxidant enzyme activity in children with Down syndrome. Curr Pediatr Res 2012;16(1):43-7.
12. Felti L, Pacáková V, Stulík K, Volka K. Reliability of carotenoid analyses: A review. Current Analytical Chemistry 2005;1(1):93-102.
13. Dehghani R, Fathi B. Scorpion sting in Iran: a review. Toxicon 2012;60(5):919-33.
14. Erel O. A novel automated direct measurement method for total antioxidant capacity using a new generation, more stable ABTS radical cation. Clin Biochem 2004;37(4):277-85.
15. Yılmaz HL, Boşnak M. [Management guidelines of scorpion sting and snake-bite in children]. Turkiye Klinikleri J Surg Med Sci 2007;3(50):104-11.
16. Petricevich VL. Scorpion venom and the inflammatory response. Mediators Inflamm 2010;2010:903295. doi: 10.1155/2010/903295.
17. Teixeira de Lemos E, Pinto R, Oliveira J, Garrido P, Sereno J, Mascarenhas-Melo F, et al. Differential effects of acute (extenuating) and chronic (training) exercise on inflammation and oxidative stress status in an animal model of type 2 diabetes mellitus. Mediators Inflamm 2011;2011:253061. doi: 10.1155/2011/253061.
18. Morgan PE, Dean RT, Davies MJ. Inactivation of cellular enzymes by carbonyls and protein-bound glycation/glycoxidation products. Arch Biochem Biophys 2002;403(2):259-69.
19. Al B, Yılmaz DA, Söğut Ö, Orak M, Üstündağ M, Bozkurt S. Epidemiological, clinical characteristics and outcome of scorpion envenomation in Batman. The Journal of Academic Emergency Medicine 2009;3(8):51-5.
20. Bouaziz M, Bahloul M, Kallel H, Samet M, Ksibi H, Dammak H, et al. Epidemiological, clinical characteristics and outcome of severe scorpion envenomation in South Tunisia: multivariate analysis of 951 cases. Toxicon 2008;52(8):918-26.
21. Petricevich VL, Peña CF. The dynamics of cytokine d nitric oxide secretion in mice injected with Tityus serrulatus scorpion venom. Mediators Inflamm 2002;11(3):173-80.
22. Annane D, Sanquer S, Sébille V, Faye A, Djuranovic D, Raphaël JC, et al. Compartmentalised inducible nitric-oxide synthase activity in septic shock. Lancet 2000;355(9210):1143-8.
23. Pessini AC, Kanashiro A, Malvar Ddo C, Machado RR, Soares DM, Figueiredo MJ, et al. Inflammatory mediators involved in the nociceptive and oedematogenic responses induced by Tityus serrulatus scorpion venom injected into rat paws. Toxicon 2008;52(7):729-36.
24. Bahloul M, Ben Hamida C, Chtourou K, Ksibi H, Dammak H, Kallel H, et al. Evidence of myocardial ischaemia in severe scorpion envenomation. Myocardial perfusion scintigraphy study. Intensive Care Med 2004;30(3):461-7.
25. Bawaskar HS. Management of severe scorpion envenomation at rural sittings: what is the role of scorpion antivenom? J Venom Anim Toxins Incl Trop Dis 2005;11(1):3-7.
26. Vasconcelos F, Sampaio SV, Garófalo MA, Guimarães LF, Giglio JR, Arantes EC. Insulin-like effects of Bauhinia forficata aqueous extract upon Tityus serrulatus scorpion envenoming. J Ethnopharmacol 2004;95(2-3):385-92.
27. Murthy KRK. The scorpion envenoming syndrome: a different perspective. The physiological basis of the role of insulin in scorpion envenoming. J Venom Anim Toxins 2000;6(1):4-51.
28. Dousset E, Carrega L, Steinberg JG, Clot-Faybesse O, Jouirou B, Sauze N, et al. Evidence that free radical generation occurs during scorpion envenomation. Comp Biochem Physiol C Toxicol Pharmacol 2005;140(2):221-6.