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The Comparison of Oxidant and Antioxidant Parameters in Inflamed Paw Tissue Formed by Carrageenan Injection in Intact and Adrenalectomized Rats

[İntakt ve Adrenalektomili Sıçanlarda Karrageninle Oluşturulan İnflamasyonlu Pençe Dokusunda Oksidan ve Antioksidan Parametrelerin Karşılaştırılması]

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ABSTRACT

Aim: In this study, oxidative stress, antioxidant response and inflammation parameters were investigated in intact and carrageenan-inflamed paw tissues in intact and adrenalectomized rats. The effects of prednisolone and adrenaline on these parameters were investigated.

Materials and Methods: 37 Albino Wistar male rats were divided into six groups as follows: I: only adrenalectomized, II: adrenalectomized + carrageenan-given, III: adrenalectomized + prednisolone-and carrageenan-given, IV: adrenalectomized + adrenaline-and carrageenan-given, V: only carrageenan-given and VI: intact rats. Carrageenan was injected as the inflammatory agent. At the end of experiments, the paw tissues were obtained and homogenised. Thiobarbituric acid reacting substances and nitric oxide levels, myeloperoxidase, superoxide dismutase, glutathione peroxidase and glutathione S-transferase activities were measured in the supernatants.

Results: The highest mean thiobarbituric acid reacting substances and nitric oxide, the lowest superoxide dismutase and glutathione peroxidase values were determined in group II. In group III and IV were significantly suppressed thiobarbituric acid reacting substances, nitric oxide and myeloperoxidase values and increased superoxide dismutase, glutathione peroxidase and glutathione S-transferase activities. On the other hand, there was not any statistically significant difference between group III and IV's parameters.

Conclusion: These results showed that adrenalectomy resulted increase in oxidant parameters and decrease in antioxidant parameters, particularly in inflammation. Prednisolone and adrenaline significantly suppressed the increase in oxidants and decrease in antioxidants in adrenalectomized + carrageenan-given rats.

Key Words: Adrenalectomy, paw edema, carrageenan, oxidants, antioxidants

ÖZET

Amaç: Bu çalışmada, intakt ve adrenalektomize sıçanların, intakt ve karrageninle oluşturulmuş inflamasyonlu pençe dokularında oksidatif stres, antioksidan yanıt ve inflamasyon parametreleri ölçüldü. Ayrıca prednisolon ve adrenalin suplementasyonunun bu parametreler üzerine etkileri de araştırıldı.

Gereç ve Yöntem: 37 adet Albino Wistar cinsi erkek sıçan altı gruba ayrıldı; grup I: yalnızca adrenalektomi, grup II: adrenalektomize + karragenin verilen, grup III: adrenalektomize + prednisolon-karragenin verilen, grup IV: adrenalektomize + adrenalin-karragenin verilen, grup V: yalnızca karragenin verilen ve grup VI: intakt sıçanlar. Karragenin inflamatuvar ajan olarak pençelere enjekte edildi. Deneysel işlemlerin sonunda pençe dokuları alındı, uygun tamponlara konuldu ve homojenize edildi. Tiyobarbitürik asit reaktif maddeleri ve nitrik oksit düzeyleri, miyeloperoksidaz, süperoksit dismutaz, glutatyon peroksidaz ve glutatyon S-transferaz aktiviteleri süpernatanlarda ölçüldü.

Bulgular: En yüksek ortalama tiyobarbitürik asit reaktif maddeleri ve nitrik oksit değerleri ve en düşük süperoksit dismutaz ve glutatyon peroksidaz değerleri grup II'de elde edildi. Grup III ve IV'te tiyobarbitürik asit reaktif maddeleri, nitrik oksit ve miyeloperoksidaz değerleri belirgin olarak baskılandı, süperoksit dismutaz, glutatyon peroksidaz ve glutatyon S-transferaz değerleri arttı. Diğer yandan grup III ve IV arasında parametreler açısından belirgin bir fark yoktu.

Sonuç: Bu sonuçlar göstermiştir ki adrenalektomi, oksidan parametrelerde artışa, antioksidan parametrelerde azalmalara (özellikle inflamasyonda) yol açmıştır. Prednizolon ve adrenalinin verilmesi adrenalektomize + karragenin verilen sıçanlarda oksidanlarda artışı ve antioksidanlarda azalışı belirgin bir şekilde baskılamıştır.

Anahtar Kelimeler: Adrenalektomi, pençe ödemi, karragenin, oksidanlar, antioksidanlar

Introduction

Reactive oxygen species (ROS) (oxygen ions, free radicals and peroxides) are highly reactive substances due to the presence of unpaired valence shell electrons. Lipid peroxidation, which is taken place by free oxygen radicals, is an indicator of oxidative damage particularly seen in cell membranes, and it leads to excess Ca^{2+} accumulation within a cell by altering membrane permeability (1, 2). Dysfunction of a cell membrane results in cell swelling and even death. Malondialdehyde (MDA) is an end product of lipid peroxidation and is used for showing the level of oxidative damage (3-5). It is well known that oxidative stress plays an important role in commence and development of the pathology in many diseases (6). Free radical injury may be the primary reason for some diseases, may lead to an increase in complications, or may increase the cell damage depending on other factors (7, 8).

Antioxidant defense systems of the body are important in terms of maintaining and protecting homeostasis. Cells protect themselves against the damage of ROS and other radicals by using repairing processes, defense enzymes, and internal and external antioxidants, namely, the free radical scavengers (9). Enzymatic defense takes place through enzymes like superoxide dismutase (SOD), catalase and glutathione peroxidase (GPx) (10). SOD removes free superoxide radicals. Whereas GPx removes H_2O_2 and lipid peroxides (9). Glutathione S-tranferase (GST) catalyzes the conjugation of some potentially toxic electrophilic xenobiotics to nucleophilic GSH, thus prevents damage to DNA, RNA or proteins (11). Similarly, the endocrine system fulfills the same duty. The adrenal gland, which is a part of this system, undertakes an important part of this duty through both cortical and medullar hormones. Secretions of the adrenal gland are controlled by internal and external stimuli, apart from their role of affecting other functions responsible from maintaining homeostasis. Hormones of the sympathoadrenal system are not vital for survival, while being required for the adaptation of acute and chronic stress. Adrenaline, noradrenaline, and dopamine are the most important hormone molecules of severe stress response (12).

Some studies have revealed that glucocorticoid levels increase in the inflamed tissues (with the aim of inhibiting inflammation) and the nitric oxide (NO) production, which increases in inflammation, is inhibited by glucocorticoids (13,14). Furthermore, investigations have demonstrated that glucocorticoids are a regulatory factor for antioxidant enzymes in the peripheral tissues containing corticosteroid receptors (15,16). Carrageenan, a local inflammatory agent used in the present study, was showed that it leads to an increase in the peripheral release of NO (17). Myeloperoxidase (MPO) is abundantly present in neutrophils and produces hypoclorous acid from H_2O_2 and Cl^- during respiratory burst, oxidizes tyrosine to tyrosyl radical and is responsible from the green color of inflammation. In adrenalectomized rats, the severe inflammation due to carrageenan would be the reason of the damage seen in cell membranes due to the more rigorous carrageenan inflammation in adrenalectomized animals when compared with the intact ones (18).

In a study performed on adrenalectomized rats, subcutaneous glucocorticoid injection was demonstrated to extenuate the antioxidant defense system of the brain, while increasing SOD activity and decreasing GPx activity of the liver tissue (19). In another study, SOD and GPx activities along with H_2O_2 production decreased significantly in the peritoneal macrophage cells of adrenomedullectomized rats, when compared with the control group; however, this decrease was stated to be less in the group that dexamethasone was given and it was suggested that in these cells, glucocorticoids and adrenaline could control antioxidant enzyme activities physiologically (20).

In the light of all the above-mentioned information, the present study was aimed to compare the oxidant and antioxidant parameters affecting inflammation, to investigate the effect of adrenalectomy and local inflammation on oxidant (MPO, MDA and NO) and antioxidant parameters (SOD, GST and GPx), and to evaluate the effects of external glucocorticoid (prednisolone) and cathecholamine (adrenaline) on these parameters in the paw tissue of rats. Measurement of Thiobarbituric acid reacting substances (TBARS) was used as an indicator of MDA value.

Materials And Methods

A total of 37 Albino Wistar male rats, with a mean weight of 195 ± 15 g were used. All animals were provided by The Center of Experimental Research and Practice in Atatürk University and the experiments were performed according to the approved ethical rules. Ethical approval of Atatürk University was received in 06.12.2006 with the number of 2006 4.1/13. Most of the chemicals used were purchased from MERCK and SIGMA Chemical Co. unless otherwise stated.

Experimental Groups and Procedure

All experimental animals were categorized into 6 different groups; group I (n=6): only adrenalectomized rats, group II (n=7): adrenalectomized + carrageenan-given rats, group III (n=7): adrenalectomized + prednisolone-and carrageenan-given rats, group IV (n=7): adrenalectomized + adrenaline-and carrageenan-given rats, group V (n=5): rats with only carrageenan-given rats and group VI (n=5): intact rats (the control group).

After grouping, 27 rats (group I, II, III and IV) underwent bilateral adrenalectomy under thiopenthal anesthesia. After operation, the animals were housed in their cages and were fed with standard commercial rat chow, tap water and 1 % NaCl solution. Group V and VI animals were also fed with the same chow and tap water except NaCl solution. At the eighth day of operation, an intraperitoneal injection of prednisolone (5 mg/kg) was

administered to group III animals, and adrenaline (100 µg/kg) to group IV animals. These doses of adrenaline and prednisolone provided the effect, which is provided by the physiologic levels of these hormones (21).

After 15 minutes of prednisolone and adrenalin injections of groups defined above, carrageenan (0.1 mL, 1 % dilution) was injected into the right paws of rats as an inflammatory agent in grup II-V. Carrageenan, which is a polymer of galactose located at the cell wall, is soluble in water and mainly used for food and ice-cream industry. At the same time, it is used for forming inflammation in experimental animals as a powerful inflammatory agent (22). In this study, carrageenan was provided by Sigma-Germany. Carrageenan, which is found as powder, was diluted with injectable water. A criterion that inflammation has been developed was the increase in the paw volume, which is measured by pletismometer. Inflammation was developed 4 hours later than carrageenan injection. Following the completion of the experimental procedures, tissue sampling was performed by extracting whole paw tissue when the inflammation was developed. After buffered appropriately, the tissue was homogenized. Supernatants were obtained from these samples and stored at -80 °C until the day of biochemical analyses.

Biochemical Analyses

Determination of GPx Activity

The GPx enzyme, in the presence of H_2O_2, reduces H_2O_2 to water while catalyzing the reaction of GSH turning into oxidized glutathione (GSSG). The GSSG formed is reduced again to GSH by the glutathione reductase reaction using NADPH as the reducing substrate. There is a decrease in absorbance during the oxidation of NADPH to $NADP^+$ and this is measured by a spectrophotometer at 340 nm for the calculation of the GPx activity (23). The results were given as U/mg protein.

Determination of GST Activity

Total GST activity was measured via the conjugation of 1-chloro 2,4-dinitrobenzene with the reduced glutathione. The increase in the absorbance of this compound at 340 nm was calculated as it is directly proportional with GST activity (24). The results were given as U/mg protein.

Determination of SOD Activity

SOD activity was determined by the method of Sun et al. (25). SOD estimation is determined by the inhibition of the reduction of nitroblue tetrazolium by superoxide radicals, which are produced by the xanthine oxidase system. Enzyme activity was measured by the degree of inhibition of this reaction, and was expressed as U/mg protein.

Determination of MPO Activity

For establishing MPO activity, MPO mediated H_2O_2 oxidation reaction was used with the 4-aminoantipyrine/phenol solution being the substrate (26). The results were given as mU/mg protein.

Measurement of NO Levels

Since NO is a very short-lived radical, nitrite + nitrate levels were measured instead of NO. Griess reagent (sulfanylamide and N-(1-naphtyl)-ethylenediamine) was used for this assay. The method is based on a two-step process. The first step is the conversion of nitrate into nitrite using a nitrate reductase. The second step is the addition of the Griess reagent, which converts nitrite into a deep purple azo compound; photometric measurement of absorbance at 540 nm is due to the fact that this azo chromophore accurately determines nitrite concentration (27). The results given as µmol/g wet tissue.

Measurement of TBARS Levels

For TBARS assay at high temperatures (95 °C) the thiobarbituric acid and TBARS form a pink-colored complex and its absorbance is measured at 532 nm by spectrophotometry (28). The results are given as nmol/g wet tissue.

Measurement of protein levels

The measurement of protein in tissue was defined according to the Bradford method (29). The amount of protein was established by measuring the absorbance of the blue-colored complex at 595 nm with spectrophotometer and this blue-colored complex was formed as a result of the negative charged Coomassie Brilliant Blue G-250 binding to the positive charges on the proteins.

Statistical Analysis

The results obtained were given as "mean value ± standard deviation" ($\bar{x}$ ± SD). The importance of differences between groups was described with the one-way ANOVA test. Following this, Fisher's post-hoc LSD (least significant differences) was performed. Spearman test was used for correlation analyses. All statistical analyses were performed with the "SPSS for Windows, version 11.5" statistical software and $p<0.05$ was accepted as statistically significant.

Results

A total of 37 Albino Wistar male rats, with a mean weight of 195 ± 15 g were used in the present study. All experimental animals were categorized into 6 different groups. The aim was to evaluate the affects of adrenalectomy and local inflammation on oxidation, on enzymes forming the antioxidant response, and the effects of exogenous glucocordicoids and adrenaline on this response. According to this, the groups were formed and homogenates of paw tissues of all animals were prepared and TBARS and NO levels, SOD, GPx, GST and MPO activities were measured and all mean ± SD, min-max levels were calculated. Increase in paw volume, which is considered an indicator of local inflammation was 100 %, 29 %, 36 % and 80 % for Group II, III, IV and V, respectively.

As seen in Figure 1, the highest mean TBARS levels were determined in group II (adrenalectomized + carrageenan-given rats) and the lowest in the intact group (group VI). Group III and IV had similar TBARS levels. There were differences between only adrenalectomized group (group I) with group VI ($p<0.05$), group V ($p<0.01$) and group II ($p<0.001$). Group II having the highest TBARS levels (31.8±2.5 nmol/g wet tissue) had significant differences with the remaining groups in terms of TBARS ($p<0.001$ for all). The difference between group III and IV was statistically insignificant ($p>0.05$). On the other hand, the difference between intact group and only carrageenan-given group was statistically significant ($p<0.001$) (Figure 1).

In the evaluation of NO levels in paw tissue samples, the highest mean levels was found in group II (adrenalectomized + carrageenan-given rats) and the lowest levels in group VI (intact). The values were similar in groups I, III and IV. In terms of NO levels, group I had lower values than group II and group V ($p<0.001$). Additionally, the difference between group I with III, IV and VI was statistically insignificant. Like in TBARS, the highest NO levels were observed in group II ($p<0.001$ for all). The difference between group III and IV was statistically insignificant (Figure 2).

As for the SOD, the highest value (25.0±4.7 U/mg pro-

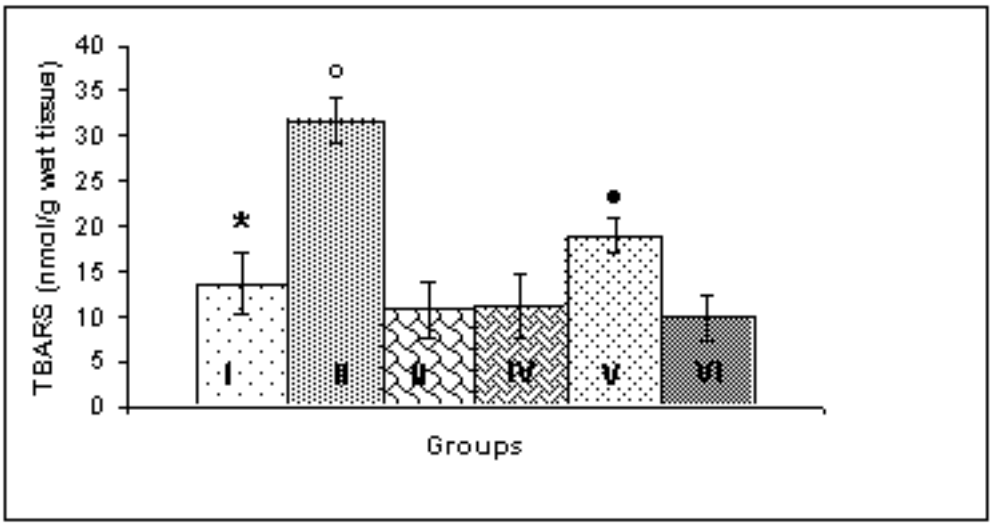

Figure 1. Distribution of TBARS levels in the study groups
*Group I with VI $p<0.05$, group I with V $p<0.01$
°Group II with all groups $p<0.001$
•Group V with III, IV and VI $p<0.001$

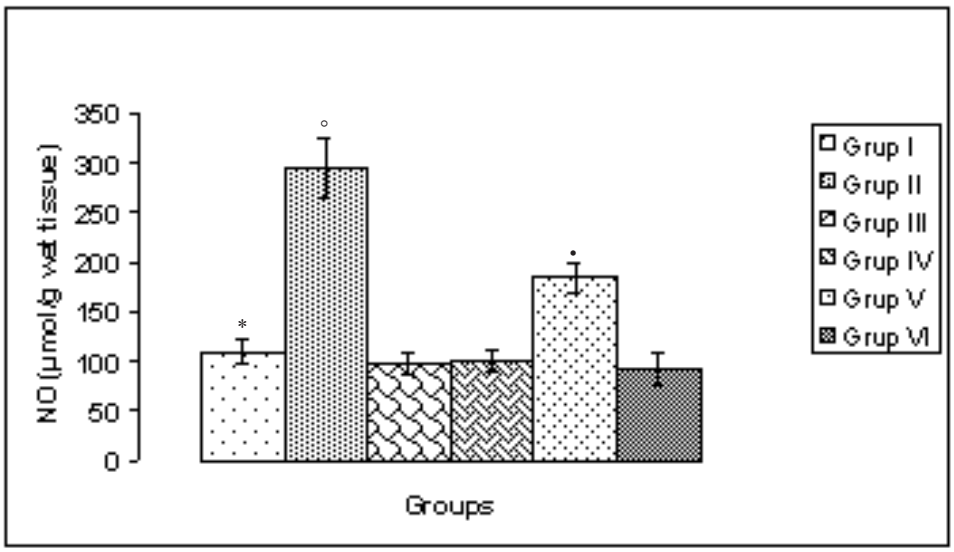

Figure 2. Distribution of NO levels in the study groups
*Group I with II and V $p<0.001$
°Group II with all groups $p<0.001$
•Group V with III, IV and VI $p<0.001$

tein) was belonging to the intact group (VI) and the lowest (8.5±2.5 U/mg protein) to adrenalectomized + carrageenan-given rats group (II). While there was a borderline difference between group I and II (p=0.049), no significant difference was present between group III and IV. The difference was very significant in the comparison of adrenalectomized + carrageenan-given group (II) with others (p<0.001). The difference was at an intermediate level between intact and only carrageenan-given rats (p<0.01) and at a weak level between only carrageenan-given rats with both prednisolone and adrenaline groups (p<0.05) (Figure 3).

Tissue GPx activity was highest in groups III and IV and lowest in group II. In statistical comparisons, only group II had significant differences with other groups (p<0.001 with group III and IV, p<0.01 with group VI, p<0.05 with group V) (Figure 4).

Among antioxidant parameters, tissue GST activity was found to be highest (1.43±0.46 U/mg protein) in group III (adrenalectomized + prednisolone-and carrageenan-given rats) and lowest (0.50±0.14 U/mg protein) in group II (adrenalectomized + carrageenan-given rats). The activities were similar in groups III and IV. In group comparisons, while there were significant differences between groups II with III and IV (p<0.001 for both), no significant difference was detected between V and VI. Both prednisolone and adrenaline groups had significantly increased GST activities when compared to intact (p<0.001 for both) and only carrageenan-given rats (p<0.001 and p<0.01) (Figure 5).

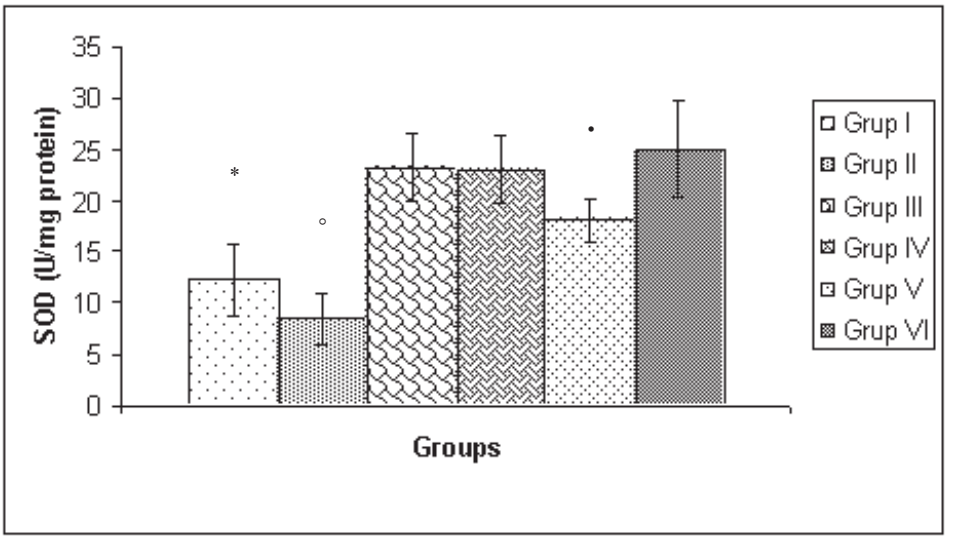

Figure 3. Distribution of SOD activities in the study groups
*Group I with III, IV and VI p<0.001, group I with V p<0.01
°Group II with all groups p<0.001
•Group V with VI p<0.01, group V with III and IV p<0.05

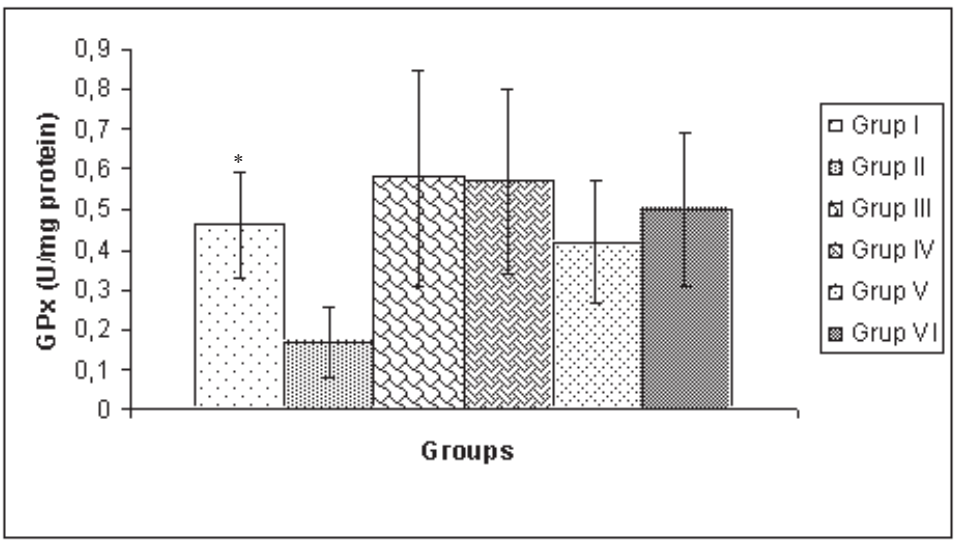

Figure 4. Distribution of GPx activities in the study groups
*Group II with III and IV p<0.001, group II with VI p<0.01, group II with V p<0.05

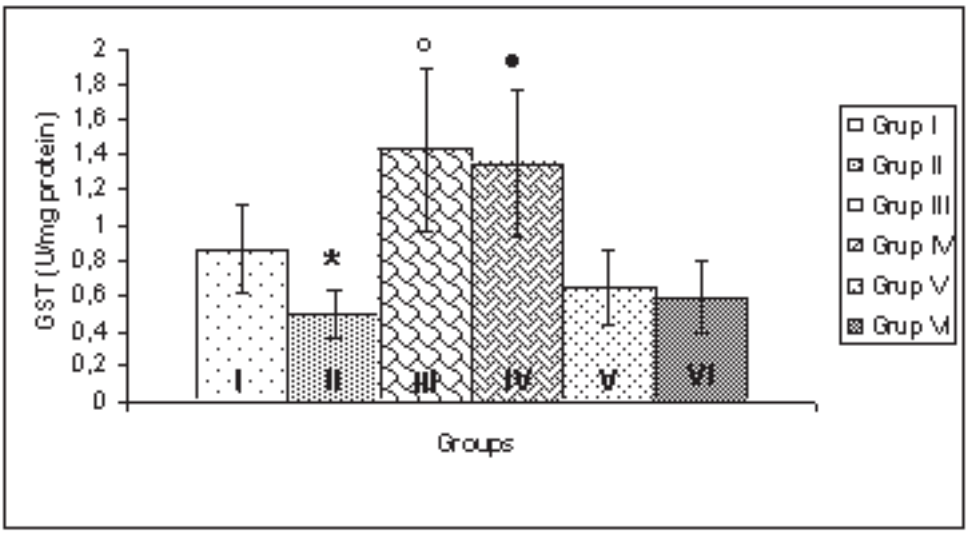

Figure 5. Distribution of GST activities in the study groups
*Group II with III and IV p<0.001
°Group III with V and VI p<0.001
•Group IV with V p<0.01, group IV with VI p<0.001

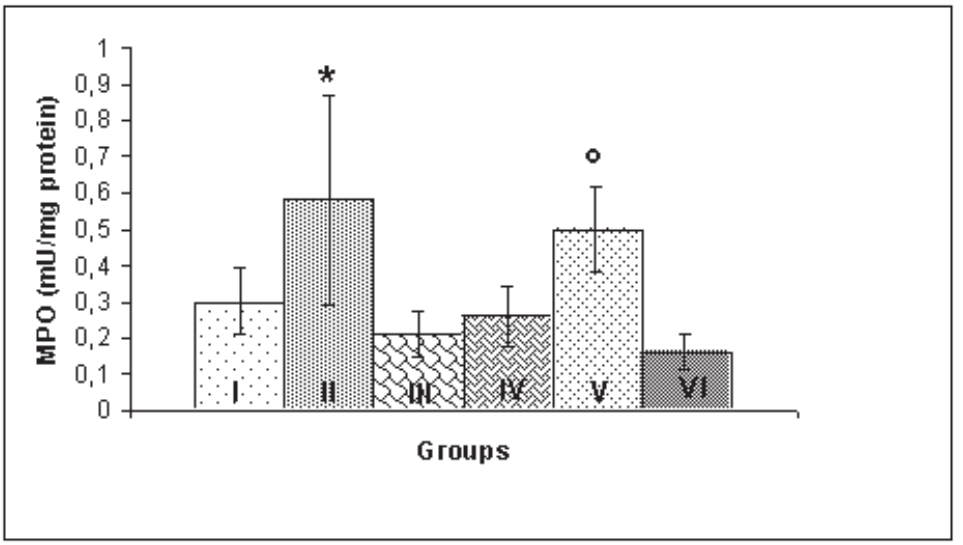

Figure 6. Distribution of MPO activities in the study groups
*Group II with I p<0.01, group II with III, IV and VI p<0.001
°Group V with IV p<0.05, group V with III and VI p<0.01

In the evaluation of MPO, as an inflammation marker, adrenalectomized + carrageenan-given rats group had the highest value (0.58±0.29 mU/mg protein) and intact group had the lowest value (0.16±0.051 mU/mg protein). The differences were at p<0.05 level between I-V and IV-V, and at p<0.01 level between V-VI, V-III and I-II. Adrenalectomized + carrageenan-given rats group had highyl significant differences when compared to both adrenaline and prednisolone groups (p<0.001). On the other hand, there was not any significant difference between intact group with adrenaline and prednisolone groups (p>0.05 for both) (Figure 6).

Correlation analyses revealed that in group I (adrenalectomy group) there was a positive correlation between NO-TBARS (r = 0.89, p<0.05) and NO-MPO (r = 0.94, p<0.01), however, negative correlations at various levels were observed between NO-SOD (r = -0.94, p<0.01), SOD-TBARS (r = -0.67, p<0.05), and SOD-MPO (r = -0.83, p<0.05). In group II, there were negative correlations between NO-SOD, SOD-TBARS, SOD-MPO, TBARS-GST, GST-MPO, and GPx-MPO, while there were positive correlations between NO-TBARS, NO-MPO, SOD-GST, SOD-GPx, TBARS-MPO, and GST-GPx. In group III, there were only positive correlations

between TBARS-NO (r = 0.86), TBARS-MPO (r = 0.78) (for both $p<0.05$) and NO-MPO (r = 0.93, $p<0.01$). In group IV, there was a negative correlation between MPO-GST. However, positive correlations were calculated between NO- TBARS, NO-MPO, and TBARS-MPO. In group V, there was a positive correlation between NO-MPO and negative correlations between NO-SOD and SOD-TBARS ($p<0.05$ for all). In group VI, only TBARS and MPO were correlated (r = 0.9, $p<0.05$).

Discussion

It is well known that there is a balance between oxidant and antioxidant events in living organisms. But, sometimes this balance is destroyed and consequently, some pathologic processes develop. The sources of oxidative stress are mainly free radicals (7). Reactive oxygen species are molecules like hydrogen peroxide, ions like the hypochlorite ion, and radicals like the hydroxyl radical. It is the most reactive of them all; note how it differs from the hydroxyl ion. The superoxide anion is both ion and radical. In cells, there are some mechanisms, called as antioxidant system, struggle with oxidative damage. The antioxidant system has an enzymatic (SOD, GST, catalase etc.) and non-enzymatic (vitamins, carotenoids, phenoles etc.) components. There are also some drugs acting as antioxidant (7).

In the light of these data, we prepared an experimental model in which we aimed to investigate the effect of adrenal gland removal and local inflammation on oxidative stress and antioxidant parameters. For this purpose, TBARS, NO, SOD, GPx, GST and MPO were measured in the paw tissue (adrenelactomized, inflamed and intact) samples of the rats.

Results of the study showed that in the paw tissues of intact and adrenalectomized rats with inflammation, there was a statistically significant difference in oxidant and antioxidant parameters.

The highest TBARS levels were observed in group II, in which carrageenan inflammation associated the adrenalectomy, following this TBARS levels from highest to lowest were in groups V, I, IV, and III, with the lowest levels measured at the intact group, in which no procedure was undertaken. In the adrenalectomized group with local inflammation given prednisolone and adrenaline, the levels were close to each other and that of the intact group. In groups with inflammation, TBARS levels of especially groups II and V were significantly higher than the other groups. Thus, TBARS levels increase in the injured tissue increase significantly, even when the paw tissue of rats adrenalectomized only are compared with intact rats. This indicates that adrenal gland hormones protect the tissue from lipid peroxidation injury. MDA is an end-product in lipid peroxidation and is formed at the end of some serial reactions in lipid peroxidation and it is widely used as an indicator of oxidative stress (30, 31). An experimental study has revealed that in adrenalectomized rats, carrageenan lead to a stronger injury when compared with the intact rats (18). In the present study, the TBARS levels of the paws of adrenalectomized rats given carrageenan were very high, when compared with the intact rats given carrageenan. This information obtained from the present study and the literature indicates that the increase in TBARS levels is parallel to the severity of inflammation.

The present study also showed that giving adrenaline to adrenalectomized rats before carrageenan inflammation significantly decreased TBARS levels when compared with groups II and V. The differences between adrenaline, prednisolone, and intact rat groups were statistically insignificant. According to the results of this experiment, it can be stated that both adrenal cortex and adrenal medulla hormones play a role in the suppression of TBARS levels in the inflamed tissue. In the study of Süleyman *et al*. (32) in the adrenalectomized rats, in which inflammation was formed with carrageenan, the anti-inflammatory effect of the NSAID given was significantly more prominent than rats given adrenaline and prednisolone, however, there were no significant differences between groups given adrenaline only, prednisolone only, and both adrenaline and prednisolone.

Another parameter measured in the present study was NO levels showing oxidation and the highest levels were observed in the adrenalectomy + carrageenan group, highest to lowest levels were seen in groups V, I, IV, and III, respectively, with the lowest levels being seen in the intact group (VI). This showed similarity with the TBARS levels.

In the present study, NO levels of the inflamed tissue were parallel to the severity of inflammation. In the paw tissue of adrenalectomized rats in which carrageenan was given (grup II), NO level was significantly higher than all other groups, including the group in which only carrageenan was administered. Again, NO levels of the group to which only carrageenan was given was significantly higher than the group that was adrenalectomized only. NO levels of the adrenalectomized rats, in which adrenaline and prednisolone was administered, did not reveal statistically significant differences. Furthermore, NO levels of the adrenalectomized rats in which adrenaline and prednisolone was administered, were not too different from the intact (control) group. Thus, it suggests that especially inflammation plays a major role in increasing NO levels, however they are not very efficient in the absence of the adrenal gland.

The highest SOD activity was observed in intact group and lowest in group II. But, low values were observed in only carrageenan given group and only adrenalectomized group. Here, the decreasing effect of inflammation on SOD activity can be considered. In the inflammation originating from oxidative stress, ROS might inhibit SOD activity.

The effect of adrenalectomy is the decrease in SOD activity, being more prominent in inflammation and the difference between intact and adrenalectomy only

groups were statistically significant. However, the difference between the adrenalectomized groups, in which adrenaline and prednisolone was administered, was also statistically significant. This shows that the control of SOD activity can be related with adrenal gland hormones. In the study of Yıldırım *et al*. (33), it was found that following adrenalectomy, the SOD activity in the stomach tissue was significantly lower, when compared with the sham operated group. In the same study, especially adrenaline was suggested to play a role on SOD and GPx activities.

In the present study, the GPx activity of intact and adrenalectomized rat paw tissues did not reveal statistically significant differences, however, when carrageenan was administered to adrenalectomized rats, the enzyme activity decreased significantly in relation to these groups. On the other hand, in groups in which adrenaline and prednisolone was administered, this activity increased significantly. This suggests that adrenaline and prednisolone also play important roles in the protection of GPx activity.

The GST activity of the adrenalectomized rats-given carrageenan was lower than the intact rats. However, the GST activities of adrenalectomized rats given adrenaline and prednisolone were 2.6 and 2.1 times higher than healthy (intact) rats, respectively. There was no statistically significant difference between only carrageenan-given group and intact group. This indicates that endogenous adrenaline and glucocorticoids may affect the prevention of GST exhaustion.

MPO, as the marker of leukocyte accumulation in tissues, forms hypochlorus acid and chloramines with long-half life from H_2O_2. Subjects with MPO deficiency have a problem in a defense mechanism leading to some fungal and bacterial infections (34-36). In the present study, the MPO values in the paw tissue of intact and adrenalectomized rats given carrageenan were also significantly higher than that of intact rats. When the MPO activity of adrenalectomized-inflamed tissues given prednisolone and adrenaline were evaluated, there was a significant decrease when compared only with the adrenalectomized group-given carrageenan. Thus, this can be considered as a finding supporting the antiinflammatory effects of prednisolone and adrenaline.

As a result, adrenal gland hormones play an important role in preserving the oxidant-antioxidant balance and the response of the organism to local inflammation being more pronounced. The absence of the adrenal gland induces the oxidative stress by itself and endangers the organism to get more damage during local inflammation. Related future studies supporting these findings will be beneficial.

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