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RESEARCH ARTICLE

The Investigation of the Effect of Boron on Intestinal Incision Wound Healing in Rats

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ABSTRACT

In this study 250-300 g 38 adult male Wistar Albino rats were used, randomly divided into six groups as four groups that received boron (n=7/group) and two control groups (n=5/group). Etibor-48 (Borax pentahydrate) was diluted in saline and administered by gavage. The Boron 1 group received 10 mg.kg-1 boron for three days and the Boron 2 group received 10 mg.kg-1 for seven days, Boron 3 group received 30 mg.kg-1 for three days, and the Boron 4 group, received 30 mg.kg-1 for seven days before the start of the study. On day 0, colon incisional wound was performed for all rats under general anesthesia and sutured. Control groups took no drugs and were euthanized on the third and seventh postoperative days. Blood sampling were done via cardiac punction at necropsy to measure Interleukin-1, IL-6, TNF-α, MPO, MDA, NO, GSH, and AOA. The sutured bowel incision line was evaluated histopathologically. Inflammatory cell measurement results in Boron 1, 2, 3, and 4 groups, and Control 3- and 7-day groups were 2.93±0.41, 3.93±0.41, 3.61±0.55, 3.93±0.41, 1.26±0.41, and 1.43±0.51, respectively. There were statistical difference between the groups ($p<0.05$). AOA measurement results in the Boron 1, 2, 3, and 4 groups, and Control 3- and 7-day groups were 7.38±0.64, 8.27±0.57, 9.07±1.16, 9.06±0.86, 10.00±1.47, and 9.86±0.54 mmol.L-1, respectively. There were statistical difference between the Boron 1 and the Control 3- and 7-day groups ($p<0.05$). It is concluded that Borax pentahydrate solution 30 mg.kg-1 had a positive effect on intestinal incisional wound healing, contrary to the literature.
.**Keywords:** Antioxidant activity, Borax pentahydrate, intestinal incisional wound healing, rat

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Ratlarda Barsak Ensizyon Yarası İyileşmesi Üzerine Bor'un Etkisinin Araştırılması

ÖZ

Bu çalışmada 250-300 g, 38 erişkin erkek Wistar Albino rat kullanıldı, rastgele olarak Bor verilen dört grup (n=7/grup) ve iki kontrol grubu (n=5/grup) olmak üzere altı gruba ayrıldı. Etibor-48 (Boraks pentahidrat) serum fizyolojik içinde seyreltildi ve gavaj yoluyla uygulandı. Bor 1 grubuna üç gün 10 mg.kg-1 bor, Bor 2 grubuna yedi gün 10 mg.kg-1 bor, Bor 3 grubuna üç gün 30 mg.kg-1 bor ve Bor 4 gruba, çalışmanın başlamasından yedi gün önce 30 mg.kg-1 bor verildi. 0. günde tüm ratlara genel anestezi altında kolon anastomozu yapıldı. Ameliyat sonrası kontrol gruplarına hiçbir ilaç uygulanmadı ve postoperatif üçüncü ve yedinci gün ötenazi yapıldı. MPO, IL-1, IL-6, TNF-α, MDA, NO, GSH ve AOA'yı ölçmek için nekropsi sırasında tüm sıçanlardan kardiyak punksiyonla kan örneği alındı. Anastomoz bölgesi histopatolojik olarak değerlendirildi. Bor 1, 2, 3 ve 4 grupları ile Kontrol 3 ve 7 günlük gruplarda inflamatuar hücre ölçüm sonuçları sırasıyla 2,93±0,41, 3,93±0,41, 3,61±0,55, 3,93±0,41, 1,26±0,41 ve 1,43±. 0.51 olarak bulundu. Gruplar arasında istatistiksel farklar vardı ($p<0.05$). Bor 1, 2, 3 ve 4 grupları ile Kontrol 3 ve 7 günlük gruplarda AOA ölçüm sonuçları sırasıyla 7,38±0,64, 8,27±0,57, 9,07±1,16, 9,06±0,86, 10,00±1,47 ve 9,86±0,54 mmol/l idi. Bor 1 ve Kontrol 3- ve 7 günlük gruplar arasında istatistiksel fark vardı ($p<0.05$). 30 mg.kg-1 Boraks pentahidrat solüsyonunun bağırsak yara iyileşmesi üzerine literatürün aksine olumlu etkisi olduğu sonucuna varılmıştır.
Anahtar Kelimeler: Antioksidan aktivite, barsak ensizyonel yara iyileşmesi, Borax pentahydrate, sıçan.

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INTRODUCTION

Wound healing is mainly comparable in all tissues; however, it has some distinct features in the gastrointestinal system (Kiliçoğlu et al. 2005). In intestinal wounds, unlike skin wounds, smooth muscle cells also synthesize collagen together with fibroblasts, and the tensile force occurs much faster in the intestine (Yağci 2011). In the gastrointestinal tract, many different features exist that are not found in the skin, such as the presence of a large pool of microorganisms, the effect of serosa on suture line closure, and vascular nutrition specific to the gastrointestinal system. Although our knowledge about local and systemic factors affecting gastrointestinal anastomosis healing is increasing, anastomotic leak and separation are common serious problems with high mortality (Yağci 2011).

Boron is denoted by the symbol 'B', which is first in group 3A in the periodic table (Nielsen et al. 1987; Saritaş et al. 2019). It has an atomic number of 5, an atomic weight of 10.82, a specific gravity of 2.84, and a melting point of 2190±20°C. Boron is not found in a pure form in nature. Instead, it is found in the form of boron salts or silicates by combining with oxygen. There are nearly 200 boron compounds in nature, the primary ones being boric acid and borax (Moseman 1994; Demir 2005).

The present study aimed to examine the histopathological and biochemical effects of Borax pentahydrate on intestinal incisions wound healing in rats.

MATERIAL and METHODS

This research was initiated with the approval of Afyon Kocatepe University Animal Experiments Local Ethics Committee (AKUHADYEK), dated 24.05.2018 with Registry No. 64-17.
A total of 38 adult male rats weighing 250-300 g were used in the study. Rats were housed in standard cages with a 12/12 h light/dark cycle. Animals were allowed to drink water ad libitum and provided rat chow until two hours before the study started. Orally administered Etibor-48 (Borax pentahydrate) obtained from Eti Boron Mining Enterprises was used in the study.

Anesthesia Protocol

General anesthesia was provided using a combination of 13 mg.kg-1 xylazine hydrochloride (Rompun, 50 ml, 23.32 mg.ml-1, Bayer-Germany) and 87 mg.kg-1 ketamine hydrochloride (Alfamine 10%, Ata-Fen, İzmir, TÜRKİYE).

Surgical Procedure

For all rats, on day 0, under general anesthesia, after reaching the descending colon, a 2-3 cm longitudinal incision was made in the antimesenteric region, and a double layer of 6-0 Prolene non-absorbable polypropylene suture was applied (Korkmaz et al. 2015) and checked for leaks. The abdominal wall and skin were closed using established methods in all groups. Gentamicin 4 mg.kg-1 was administered parenterally for five days, and wound care was carried out until the end of the study.

Study Groups

The 38 rats used in the study were divided into six groups at random. Groups 1 through 4 were administered boron diluted in physiologic saline by oral gavage, while groups 5 and 6 served as untreated controls.
The Boron 1 (n=7) group received boron (10 mg.kg-1) for three days before the start of the study and continued until the third postoperative day, at which time the rats were euthanized.
The Boron 2 (n=7) group received boron (10 mg.kg-1) for seven days before the start of the study and continued until the seventh postoperative day, at which time the rats were euthanized.
The Boron 3 (n=7) group received boron (30 mg.kg-1) for three days before the start of the study and continued until the third postoperative day, at which time the rats were euthanized.
The Boron 4 (n=7) group received boron (30 mg.kg-1) for seven days before the start of the study and continued until the seventh postoperative day, at which time the rats were euthanized.
The Control 3-day group (n=5) and Control 7-day group (n=5) did not receive boron and were euthanized three and seven days postoperatively.
At the time of euthanasia on day 3 or 7, samples were taken from the suture line for histopathological examination, and cardiac blood samples were obtained for biochemical measurements.

Biochemical Measurements

Determination of IL-1, IL-6, TNF-α, and MPO Activity

The serum activity of IL-1 (Biont, Rat Interleukin 1 [IL-1] ELISA Kit Catalog No: YLA0153RA), IL-6 (Biont, Rat Interleukin 6 [IL-6] ELISA Kit Catalog No: YLA0031RA), TNF-α (Biont, Rat TNF-α ELISA Kit, Catalog No: YLA0118RA), and MPO (Biont, Rat Myeloperoxidase [MPO] ELISA Kit Catalog No: YLA0046RA) were determined using commercial ELISA kits.

Determination of Malondialdehyde (MDA) Level:

The MDA level was determined based on the double-boiling method, which was modified by Draper and Hadley (1990). During the first boiling, the bound MDA in the samples is liberated from the proteins, and the proteins are precipitated. In the second boiling, the absorbance of the colored complex formed by reacting with total MDA and thiobarbituric acid (TBA) is measured at 532 nm. The concentration of MDA is then calculated using the molar absorption coefficient.

Two test tubes, a control, and a sample were prepared. Trichloroacetic acid (TCA) 10% solution (2.5 ml) was placed in both tubes, after which 0.5 ml of sample was added to the sample tube and 0.5 ml of distilled water to the control tube. The tubes were sealed and kept in a boiling water bath for 15 minutes, then cooled under cold water and centrifuged at 3000 rpm for 10 minutes, after which 2 ml of the upper supernatant was transferred to another tube and 1 ml of 0.675% TBA solution added. The lids were tightly closed, and the tubes were placed in a boiling water bath again for 15 minutes, then cooled in cold water. The absorbance of the sample against the blank was measured at 532 nm in a spectrophotometer. Using the extinction coefficient of the MDA-TBA complex at 532 nm, the MDA value was determined in nmol.ml-1 for serum and nmol.mg-1 for tissue samples.

Determination of Antioxidant Activity (AOA)

The AOA was determined based upon the fact that Fe-EDTA complex standard solution reacts with hydrogen peroxide by the Fenton reaction, leading to formation of hydroxyl radicals. Reactive oxygen radicals degrade benzoate because of TBARS release. Antioxidants added to human fluid cause suppression of TBARS production. This reaction is measured calorimetrically, with suppression of color development detected as AOA (Koracevic et al. 2001).

Determination of Nitric Oxide (NO)

A modified method determined by nitrite + nitrate (NOx) levels, as reported by Miranda et al. (2001), was used to measure NO in tissue and serum samples.

Determination of Glutathione (GSH)

A glutathione assay kit (Cayman Chemical Company, item no.703002, USA) was used to determine the GSH level.

Histopathological Examination

Samples from animals that underwent necropsy were fixed in 10% neutral buffered formaldehyde solution. After 48 hours, they were trimmed and placed into cassettes, followed through a series of alcohol and xylene, then blocked in paraffin and cut to a thickness of 4-5 microns with a microtome and placed on slides. These sections were marked with hematoxylin-eosin and analyzed under a light microscope.

Statistical Analysis

The results were statistically evaluated by a one-way ANOVA test using the SPSS 16.0 statistical package program. The Duncan test was applied to results with statistical differences. Data were expressed as mean ± standard deviation. The level of significance was set as $p<0.05$.

RESULTS

The aim of this study was to investigate the effects of 10 mg.kg-1 and 30 mg.kg-1 orally given Boron on the healing of the intestinal incision line in the 3rd, 7th day and control groups, with histopathological and biochemical parameters.

In this study, histopathological examination results of tissue sections taken from the incision line in Boron 1, Boron 2, Boron 3, Boron 4, Control 3 and Control 7 groups, respectively; as inflammatory cell, fibroblastic activity, neovascularization and collagen level were given. Inflammatory cell measurement results in the groups, found as 2.93±0.41, 3.93±0.41, 3.61±0.55, 3.93±0.41, 1.26±0.41, 1.43±0.51, respectively. Fibroblastic activity values in the groups were recorded as 3.26±0.75, 3.43±0.82, 3.77±0.52, 3.77±0.52, 1.1±0.10, 1.43±0.52, respectively. Neovascularization measurement values found as 2.93±0.41, 3.77±0.52, 3.43±0.52, 3.93±0.41, 1.26±0.41, 1.43±0.52, respectively. Collagen measurement results were determined as 3.26±0.41, 3.43±0.52, 3.43±0.52, 4.10±0.00, 1.10±0.00, 1.10±0.00, respectively. Inflammatory cells, fibroblastic activity, neovascularization and collagen levels obtained as a result of histopathological examinations were found to be statistically significant when compared between all groups ($p<0.05$) (Table 1, Figure 1).

In this study, hemogram (WBC, LYM, MID, GRA, Hb, MCH, RBC, MCV, HCT, PLT) and biochemistry parameters in blood samples taken by intracardiac injection of Boron 1, Boron 2, Boron 3, Boron 4, Control 3 and Control 7 groups (MDA, IL-1, IL-6, TNF-α, MPO, AOA, GSH, NO) measurements were made. When the hemogram results were evaluated, WBC, LYM, MID, RBC, MCV, HCT and PLT levels were not statistically significant when compared between groups ($p>0.05$), while GRA, Hb and MCH parameters were statistically significant ($p<0.05$) (Table 2).

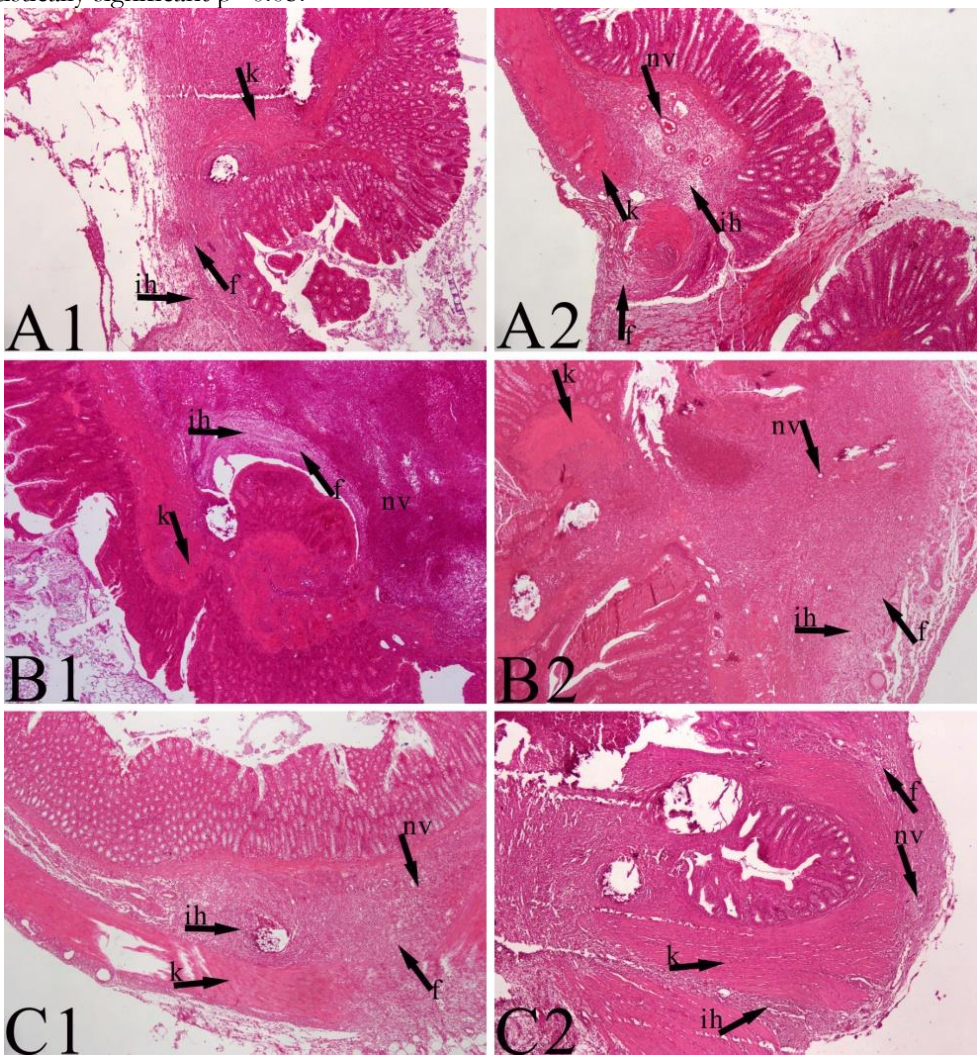
When all groups were compared as a result of biochemical measurements, no statistically significant difference was observed in GSH, TNF-α and IL-1 values ($p>0.05$) (Table 3).

When statistical comparison was made for all groups, it was found that between Boron 1 group and Boron 3, Boron 4, Control 3 and Control 7 groups, Boron 2 group and Boron 4, Control 3 and Control 7 groups, Boron 3 group and Control 7 groups difference was observed ($p<0.05$). According to IL-6 measurement results, the increase in Boron 1 group was found to be statistically significant in comparison with Boron 2, Boron 3, Boron 4, Control 3 and Control 7 groups ($p<0.05$). The increase in Boron 3 group is statistically significant in comparison to Boron 4, Control 3 and Control 7 groups ($p<0.05$) (Table 3).

Table 1: Histopathological results of inflammatory cells, fibroblastic activity, neovascularization, and collagen in groups.

	Histopathological results			
Groups	**Inflammatory cell**	**Fibroblastic activity**	**Neovascularization**	**Collagen**
Boron 1, 10 mg.kg-1 3 days	2.93±0.41[b]	3.26±0.75[a]	2.93±0.41[b]	3.26±0.41[b]
Boron 2, 10 mg.kg-1 7 days	3.93±0.41[a]	3.43±0.82[a]	3.77±0.52[b]	3.43±0.52[b]
Boron 3, 30 mg.kg-1 3 days	3.61±0.55[a]	3.77±0.52[a]	3.43±0.52[a]	3.43±0.52[b]
Boron 4, 30 mg.kg-1 7 days	3.93±0.41[a]	3.77±0.52[a]	3.93±0.41[a]	4.10±0.00[a]
Control 3 days	1.26±0.41[c]	1.11±0.10[b]	1.26±0.41[c]	1.10±0.00[c]
Control 7 days	1.43±0.51[c]	1.43±0.52[b]	1.43±0.52[c]	1.10±0.00[c]
P	**0.000**	**0.000**	**0.000**	**0.000**

[a, b, c:] Inflammatory cells, fibroblastic activity, neovascularization, and collagen values with different letters in the same column are statistically significant $p<0.05$.



A1: Control Day 3; A2: Control Day 7; B1: Boron 10 mg Day 3; B2: Boron 10 mg Day 7; C1: Boron 30 mg Day 3; C2: Boron 30 mg Day 7, nv: Neovascularization, f: Fibroblastic activity, ic: Inflammatory cell, c: Collagen.

Figure 1: Microscopic views of histopathological examination results in groups.

Table 2. Hemogram Results in Groups (Mean ±SD).

	Hemogram Results									
Groups	**WBC (10^9/L)**	**LYM (%)**	**MID (%)**	**GRA (%)**	**Hb (mg/dl)**	**MCH (pg)**	**RBC (10^{12}/L)**	**MCV (fl)**	**HCT (%)**	**PLT (10^9/L)**
Boron 1 10 mg.kg-1 (3 days)	6.24±1.67	47.27±9.41	9.00±1.95	43.72±9.10^{b}	15.15±1.12^{a}	18.85±0.88^{a}	8.06±0.75	45.77±1.89	36.81±2.58	617.71±93.85
Boron 2 10 mg.kg-1 (7 days)	8.46±5.92	52.27±22.07	11.08±3.45	36.64±21.34^{c}	14.11±1.44^{a}	19.30±1.20^{b}	7.35±1.07	47.28±1.85	34.66±4.37	818.85±168.84
Boron 3 30 mg.kg-1 (3 days)	5.76±4.08	51.90±6.22	11.55±2.51	36.54±6.47^{c}	15.41±0.46^{a}	18.01±0.63	8.57±0.45	45.41±1.96	38.87±1.30	524.85±212.64
Boron 4 30 mg.kg-1 (7 days)	6.12±1.74	42.20±14.54	10.95±6.66	46.84±12.88^{d}	13.84±1.76^{b}	18.11±0.96^{abc}	7.66±1.14	45.08±2.13	34.44±4.76	785.42±155.44
Control (3 days)	8.04±2.66	38.90±5.34	7.96±3.93	53.14±9.10^{e}	14.98±1.12^{b}	17.94±1.21	8.36±0.51	44.50±1.98	37.20±2.51	496.40±123.36
Control (7 days)	5.66±2.35	37.18±9.90	8.18±3.53	54.64±12.78^{a}	14.20±0.66^{b}	18.30±0.89^{abd}	7.79±0.59	45.08±2.13	35.02±1.54	855.40±81.62

WBC: Leukocytes (White Blood Cells), LYM: Lymphocyte, MID: Monocytes, GRA: Granulocyte, Hb: Hemoglobin, RBC: Red Blood Cells, PLT: Platelets, MCV: Mean Cell Volume, HCT: Hematocrit, MCH: Mean Corpuscular Hemoglobin

Values with different letters (a,b,c,d,e) between groups in the same column are statistically significant ($P < 0.05$).

Table 3. Biochemistry Analysis Results in Groups (Mean ±SD).

Biochemistry Analysis Results								
Groups	**MDA (mcmol/gH)**	**IL-1 (pg/ml)**	**IL-6 (ng/L)**	**TNF-α (ng/L)**	**MPO (ng/ml)**	**AOA (mmol/L)**	**GSH (nmol/gHb)**	**NO (mcmol/ml)**
Boron 1 10 mg.kg-1 (3 days)	5.03±0.11[a]	20.64±2.98	41.55±8.68[a]	47.12±8.17	22.72±5.61[a]	7.38±0.64[a]	2.41±0.32	12.56±0.80[a]
Boron 2 10 mg.kg-1 (7 days)	4.96±0.16[a]	19.52±1.70	25.34±5.30[b]	42.66±4.85	16.57±2.40[b]	8.27±0.57[ab]	2.25±0.27	10.52±1.20[ab]
Boron 3 30 mg.kg-1 (3 days)	4.37±0.44[ab]	21.41±2.10	31.43±3.01[abc]	43.46±4.93	19.98±1.43[ef]	9.07±1.16[ab]	2.20±0.37	11.33±0.74[ab]
Boron 4 30 mg.kg-1 (7 days)	3.81±0.61[ac]	21.24±1.76	19.62±3.61[abd]	45.36±10.29	15.51±1.66[c]	9.06±0.86[ab]	2.21±0.33	10.12±1.63[b]
Control (3 days)	4.03±0.80[ad]	22.67±1.43	25.54±4.99[afb]	39.82±6.31	16.11±2.86[d]	10.00±1.47[b]	2.11±0.06	10.12±1.55[b]
Control (7 days)	3.28±0.25[ae]	20.90±2.51	16.14±2.27[abe]	39.22±5.04	14.64±1.44[e]	9.86±0.54[b]	3.05±1.18	9.68±0.85[b]

Values with different letters (a, b, c, d, e) in the same column are statistically significant ($P<0.05$).

MDA: Malondialdehyde, IL-1: Interleukin-1, IL-6: Interleukin-6, TNF-α: Tumor Necrosis Factor-Alpha, MPO: Myeloperoxidase, IMA: Ischemic Modified

Albumin, AOA: Antioxidant Activity; GSH: Glutathione, NO: Nitric Oxide.

According to the statistical comparison of the MPO measurement results of all groups, the increase in Boron 1 group was statistically significant when compared with Boron 2, Boron 3, Boron 4, Control 3 and Control 7 groups ($p<0.05$). There is a statistical difference between Boron 3 group and Control 3 and Control 7 groups. The increase in boron 3 group was significant ($p<0.05$) (Table 3).

When the findings obtained through AOA measurements were compared between all groups, there was a difference between the Boron1 group and the Control 3 and Control 7-day groups ($p<0.05$). There was no difference between Boron 2, Boron 3, Boron 4 groups and any other groups ($p>0.05$) (Table 3).

When the findings obtained as a result of GSH measurements of all groups were compared statistically, the difference in Boron 1 group was statistically significant ($p<0.05$) when the increase in Boron 2, Boron 3, Boron 4, Control 3 and Control 7 groups was compared (Table 3).

DISCUSSION

Colon anastomoses are among the most common surgical procedures in humans and animals. In studies on wound healing and follow-up, anastomotic leakage is a frequent complication. High morbidity has led to an increase in the number of studies and research being conducted. The present study examined the histopathological and biochemical effects of orally administered boron on incisional wound healing of colon in rats.

Healing of wounds is a complicated process in which the tissue repairs itself (Stadelmann et al. 1998; Korkmaz et al. 2015). While the healing process of wounds is similar for different tissues, in the gastrointestinal tract, it has some characteristic properties such as tension time that develops much earlier than in the skin (Cronin et al. 1968; Korkmaz et al. 2015), and collagen is synthesized by smooth muscle cells in intestinal wounds (Graham et al. 1987; Korkmaz et al. 2015).

Intestinal wound healing involves inflammation, proliferation-fibroplasia, and maturation stages. Inflammation begins with vasodilation, secretion of vasoactive materials, increased vascular permeability, and neutrophil infiltration within three hours following vasoconstriction of the wound edges. Macrophages and fibroblasts transfer to the wound site, where macrophages control inflammation through cytokine release (Brasken 1991; Graham et al. 1992; Korkmaz et al. 2015).

There are both local and systemic aspects in the healing of intestinal wounds (Frostberg et al. 2014; Korkmaz et al. 2015). For the extracellular matrix significant factors include collagen fibers, fibroblasts, and immune cells which control wound strength during the early stages of postoperative healing process (Carrico et al. 1984; Frostberg et al. 2014; Korkmaz et al. 2015).

Tissue sections taken from the anastomosis line were evaluated histopathologically for inflammatory cells, fibroblastic activity, neovascularization, and collagen levels (Table 1). The number of inflammatory cells was high in the Boron 2, 3, and 4 groups, with statistical significance ($p<0.001$). Fibroblastic activity measurements from the anastomosis line found the Control 3- and 7-day groups to be statistically lower than in the Boron groups ($p<0.001$). When neovascularization findings obtained in all groups were compared, the highest level was found in the Boron 4 group. On the other hand, neovascularization in the Control 3- and 7-day groups was statistically lower than the Boron groups. The collagen measurement result of the Boron 4 group was statistically significant compared to the other Boron groups. On the other hand, collagen levels in the Control 3- and 7-day groups were statistically lower than all Boron groups.

It was noted that there was a significant increase in fibroblastic activity in the Boron 4 group compared to the Control groups and other Boron groups. It was determined that collagen formation increased 4-fold in the Boron 4 group compared to the Control 7-day group. Inflammatory cell formation was determined in the 7-day 10 mg and 30 mg Boron groups at a high rate. Histopathological evaluations revealed healing to be more uniform in the Boron 4 group, followed by the Boron 2 group; therefore, intestinal wound healing was clinically and histopathologically better in day-7 groups, in line with the literature.

Postoperative pain is one of the widespread issues in surgery. According to various reports, pain treatment is not sufficient in almost half of patients (Gottschalk and Smith 2001; Korkmaz et al. 2015). A multimodal approach to analgesia is necessary for the reduction of discomfort from different mechanisms. Opioids, local anesthetic agents, non-steroidal anti-inflammatory drugs (NSAIDs), paracetamol, and gabapentinoids are examples of drugs used to treat pain (Carstensen and Moller 2010; Wood 2010; Korkmaz et al. 2015).

Today, one of the most common complications in bowel operations is adhesions. Studies have reported that 12% to 17% of patients who undergo abdominal surgery for various reasons develop ileus due to serosal adhesions in the early or late postoperative period (Saribeyoğlu et al. 2008; Koç et al. 2013).

İnce et al. (2010) stated that boron and boric acid reduce liver GSH levels and boosts kidney GSH levels in rats. In our study, there were no statistical differences between any of the groups regarding GSH.

NO, MDA and MPO are substances produced by inflammatory cells or formed as by-products. NO is produced by the enzyme inducible nitric oxide synthetase (INOS) found in neutrophils. As a result of triggering the INOS enzyme in sepsis and inflammation, NO production increases (Faist et al.1996; Anup and Balasubramanian 2000; Koç et al. 2013). In the study by Koç et al. (2013) the NO level was significantly lower in an anastomotic group administered 4% icodextrin compared to a group without icodextrin use. In the present study, comparing the findings in NO levels in all groups, there was a statistically significant difference between the Boron 1 group and the Boron 4, Control 3-, and 7-day groups. There was no difference between the Boron 3 group and the other groups.

MDA is a by-product formed by the cells involved in the inflammatory response when oxygen radicals break down lipid-containing structures such as plasma and cell membranes. It is a parameter used to evaluate both tissue damage and the severity of inflammation (Singal et al. 1983; Kaul et al. 1993; Koç et al. 2013). In the study by Koç et al. (2013), MDA levels were significantly lower in the anastomosis group using 4% icodextrin. In our study, there was a significant difference ($p<0.05$) in the MDA level between the Boron 3 group and the Control 7-day group.

MPO is an enzyme used to create toxic agents that neutrophils use to break down the agents they phagocytize. It is used as an indicator of neutrophil infiltration in tissues. In the same study by Koç et al. (2013), MPO was significantly lower in the group in which 4% icodextrin was used. It has been reported that this low value indicated less adhesion formation due to a less severe inflammatory response. In the present study, when the MPO levels obtained in all groups were compared, there was a statistically significant difference between the Control 7-day and Boron 3 groups.

The biochemical role of boron is still not clear. Although the nutritional importance of boron in some pathological conditions such as arthritis and osteoporosis is not defined, it has been reported that boron increases optimal function throughout the life cycle (Naghii and Samman 1997; Wallace et al. 2002). Various studies have shown that boron is an important element for humans and animals and plays a relative role in macro mineral metabolism, endocrine functions (calcitonin, estrogen, insulin, thyroid), hormones, vitamin D metabolism, bone metabolism, and immune functions (Kabu and Akosman 2013; Kabu et al. 2015; Korkmaz et al. 2019).

A clinical study showed that 17-beta-estradiol and testosterone levels considerably increased in postmenopausal women who received a 3 mg.day-1 boron supplement for seven weeks. According to the same report, boron supplementation results in two-fold increase in testosterone coupling and a significant increase in calcium retention (Nielsen et al. 1987; Naghii et al. 2011). In another study, in males who received 10 mg of boron supplementation daily for four weeks, a significant increase in 17-beta-estradiol levels and increased plasma testosterone was reported (Nielsen 1994; Naghii and Samman 1997; Naghii et al. 2011). Wallace et al. argued that acute supplementation with 11.6 mg of boron as 102.6 mg of sodium tetraborate decahydrate with a meal resulted in a significant increase in plasma boron concentration in comparison to placebo in healthy middle-aged men (Wallace et al. 2002; Naghii et al. 2011). In general, researchers have stated that there was a 10-fold increase in plasma boron from fasting concentrations (Wallace et al. 2002; Naghii et al. 2011).

Naghii et al. (2011) stated that sex hormone-binding globulin showed significantly lower concentrations following boron consumption with no considerable difference, other than TNF-α which again showed low concentrations and six-hour boron supplementation had no significant effect on hormone concentrations.

In a study by Korkmaz et al. (2019) the fact that boron and hyaluronic acid increased antioxidant enzymes, SOD, and catalase levels in both blood and cartilage tissue showed that these two agents contribute to the antioxidant defense system. Also, the researchers reported that it was the first study on MDA, GSH, SOD, and catalase levels in both blood and articular cartilage tissue to evaluate the effect of boron administered intravenously to rats with an osteochondral defect. In this study, we determined that the AOA was significant.

Adhesion development in groups that underwent necropsy on the third and seventh postoperative days (Lange et al. 1995) was evaluated according to a 0-3+ scale. Adhesion development at the level of 0-1+ was observed in all groups; however, no adhesion formation was observed at the sutured incision line to the peritoneum and intra-abdominal organs.

It has been reported that a complex reaction called acute-phase inflammatory response begins immediately after surgical trauma (Pepys 1981), and the production of acute-phase proteins will increase immediately after surgical interventions (Roumen et al. 1992; Wilmore 1997). The substance that regulates the acute-phase protein response is IL-6 (Heinrich et al. 1990; Pullicino et al. 1990), and IL-6 secretion stimulates the secretion of other inflammatory cytokines such as TNF-α and IL-1 (Heinrich et al. 1990; Ertel et al. 1990; Yamamoto et al. 1993). In this study, however, there were no statistical differences in the results between the groups with regard to serum TNF-α values.

The increase in serum IL-6 level in the Boron 3 group was statistically significant compared to the Boron 4 and Control 3- and 7-day groups. Similarly, the increase in the Boron 1 group was statistically significant compared to all other groups. Comparing the findings obtained from all groups for IL-1, there were no differences between the groups ($P>0.05$) (Cruickshank et al. 1990). The acute-phase response reaches its peak value after trauma, and these mediators increase after surgical intervention (Conner et al. 1988; Nishimoto et al. 1989; Gruys 1994). Harada et al. (1997) found a statistically significant relationship between IL-6 and TNF-α levels. As such, an increase in the mentioned parameters was determined in the Boron 1 and Boron 3 groups. Based on these results, it was determined that the levels of inflammatory mediators decreased after the third day in parallel with the literature.

CONCLUSION

In conclusion, in this study, in which Borax pentahydrate solution was given orally by gavage, a 30 mg.kg-1 dose was determined to have positive effects on intestinal wound healing; however, more detailed studies are needed to expand upon these results.

Conflict of interest: The authors declareted that there are no conflicts of interest.

Authors' Contributions: Alarslan carried out the planning, experimental phase and writing of the study. Sarıtaş supervised the planning and writing stages of the study.

Ethical approval: This study was approved by the Afyon Kocatepe University Animal Experiments Local Ethics Committee (AKUHADYEK).

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