International Journal of Legal Medicine
https://doi.org/10.1007/s00414-022-02834-w

ORIGINAL ARTICLE

An experimental rat model of electric shock injury with isolated electric shock and water conduction: the histopathological changes on the skin and internal organs and the effect on biochemical parameters

Ahmet Sedat Dündar[1] · Mucahit Oruç[2] · Osman Celbiş[2] · Emine Türkmen Şamdancı[3] · Ayşe Nur Akatlı[3] · Hasan Okumuş[4] · Çağatay Taşkapan[5] · Onural Özhan[6] · Hakan Parlakpınar[6]

Received: 2 November 2021 / Accepted: 25 April 2022

Abstract

It is difficult to determine the cause of death in electric shock injuries when no trace can be determined on the skin, and this is accepted as a reason for negative autopsy. We aimed to determine useful parameters in the definition of the cause of deaths associated with electric shock and particularly those formed with water conduction. This study used a total of 42 rats, applied with fatal electric shock formed of isolated electric shock at 220 V and with water conduction. The serum NT-ProBNP and H-FABP levels were examined together with histopathological changes in the brain, cerebellum, brainstem, heart, liver and skin and the Bax, caspase-3 and HSP-60 antibody status in these tissues. A statistically significant difference was determined between the groups in respect of the serum H-FABP values and the immunohistochemical staining of the samples taken from the organs. In conclusion, this study is the first in literature with an experimental model of electric shock with water conduction. Using immunohistochemical and biochemical markers in deaths associated with isolated electric shock and electric shock with water conduction, the results of this study can contribute to the clarification of one of the reasons for negative autopsy in forensic medicine.

Keywords Electric shock · Water conduction · H-FABP · Immunohistochemistry · NT-ProBNP

✉ Ahmet Sedat Dündar
dr_asedat@hotmail.com

Mucahit Oruç
mucodr@gmail.com

Osman Celbiş
osmancelbis@gmail.com

Emine Türkmen Şamdancı
esamdanci@gmail.com

Ayşe Nur Akatlı
aysenurakatli@gmail.com

Hasan Okumuş
hokumus86@hotmail.com

Çağatay Taşkapan
taskapanmc@yahoo.com

Onural Özhan
onural.ozhan@inonu.edu.tr

Hakan Parlakpınar
hparlakpinar@hotmail.com

[1] Council of Forensic Medicine, Şanlıurfa, Turkey

[2] Department of Forensic Medicine, Inonu University Faculty of Medicine, Malatya, Turkey

[3] Department of Medical Pathology Malatya, Inonu University Faculty of Medicine, Malatya, Turkey

[4] Council of Forensic Medicine, Gebze-Kocaeli, Turkey

[5] Department of Biochemistry, Inonu University Faculty of Medicine, Malatya, Turkey

[6] Department of Pharmacology, Inonu University Faculty of Medicine, Malatya, Turkey

Published online: 07 May 2022

Introduction

Examination of the scene of the incident and the conditions of death, the medical history of the deceased, histopathological examination, toxicology analyses and chemical, microbiological, serological and genetic examinations are the steps that assist in determining the cause of death in an autopsy. When the cause of death cannot be determined despite all these examinations, it is referred to as negative autopsy. The negative autopsy rate with macroscopic examination only is 10%, but when detailed analyses are added, this rate falls to 1–5% [1–3].

The observation of electricity entry and exit wounds is an important clue in determining the cause of death [4]. In electric shock where traces of the current cannot be determined on the skin such as in electric shock with water conduction, it is difficult to determine the cause of death, and this is evaluated as one of the reasons for negative autopsy [1, 3, 4].

Electric shock can be a cause of death by causing damage to the affected organs and systems related to the type of current, voltage and intensity. The central nervous system (CNS), cardiovascular system, muscle tissue, lungs, liver, skin and other internal organs can be affected by electric shock, and there can be changes in some biochemical parameters [4–9]. The primary cause of death is cardiac damage [5, 10, 11]. An increase may be seen in the ventricle wall stress, in pro-brain natriuretic peptide (ProBNP) and amino terminal fragments (NT-ProBNP), and in cases of acute cardiac injury, heart type fatty acid binding protein (H-FABP) emerges. The changes formed in these two molecules in electric shocks have become a subject of interest to researchers [6, 12, 13].

The aim of this experimental study was to examine the serum NT-proBNP and H-FABP levels obtained from rats applied with fatal isolated electric shock and electric shock with water conduction, together with the histopathological changes in tissue samples taken from the brain, cerebellum, brainstem, heart, liver and skin and the Bax, caspase-3 and HSP-60 antibodies in these tissues. The data obtained in the study were investigated in respect of utility in determining the cause of death in cases of electric shock, which are one of the reasons for negative autopsy.

Materials and methods

Approval for the study was granted by the Experimental Animals Ethics Committee of Inonu University Medical Faculty (decision no:2017/A-46, dated:12.10.2017). This experimental study was conducted in the Experimental Animals Production and Research Centre of Inonu University in accordance with the EU Directive no. 2010/63/EU for experimental animals.

Animals

A total of 42 healthy, female Wistar albino rats, each weighing 200–250 g, were used in the study. The rats were randomly separated into 3 groups of 14, as group A, control group with no intervention; group B, applied with isolated electric shock; and group C, applied with electric shock with water conduction.

Preparation of the experiment and safety

A deep plastic bath was placed on a non-conductive wooden material on a concrete base. A flat piece of plastic material, pierced at certain intervals for fixation of the animals, was placed in the bath. An electrical contactor was placed in the circuit. It was connected to 3 circuit breakers with 6 amp, 15 amp and 32 amp allowed intensity limit, respectively, over the contactor. The circuit breakers which were placed for safety purposes did not activate during the experiment, and no power cut had occurred. An electrical circuit with 220 V input–output connected to an electrical socket was prepared and tested in the Electrical-Electronic Engineering Faculty of Inonu University. The electric current used in industry and domestic environment in our country is alternating current (AC) (Fig. 1). The current source in this study is AC, and the exact frequency is standardized as 60 Hz. By taking samples for analysis, tap water was prepared for use in the group to be applied with electric shock with water conduction (Table 1).

Plastic material was used to prevent electrical conduction. Although the animals were under deep anaesthesia during the experiment, the extremities of the animals were fixed to the plastic material with plastic attachments to prevent movement.

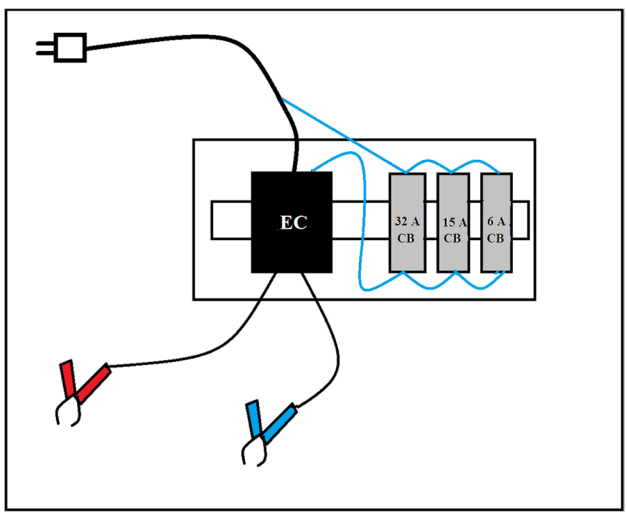

Fig. 1 Schematized electrical circuit assembly

Table 1 Chemical water analysis report made in the public health laboratory (No. 2018–1227-10)

Chemical parameters	Values
pH	7.35
Conductivity (20 0C µs/cm)	221
Free cyanide (µg/L)	0
Bromate (µg/L)	0
Nitrite (mg/L)	0
Fluoride (mg/L)	0
Nitrate (mg/L)	3.40
Boron (mg/L)	0
Nickel (µg/L)	0
Mercury (µg/L)	0
Lead (µg/L)	0.346
Selenium (µg/L)	0
Copper (mg/L)	0
Chromium (µg/L)	0
Cadmium (µg/L)	0
Antimony (µg/L)	0
Benzene (µg/L)	0
Manganese (mg/L)	0
Iron (mg/L)	0.107
Aluminium (mg/L)	0.18
Ammonium (mg/L)	0.01
Sodium (mg/L)	1.24
Chloride (mg/L)	0.63
Sulphate (mg/L)	3.29

While performing the experiment, the researchers wore plastic boots and special gloves to prevent conduction.

Procedures

All the animals were applied with anaesthesia analgesia with an intraperitoneal injection of 90 mg/kg ketamine 10% (Ketalar ® Pfizer) and 15 mg/kg xylazine hydrochloride 2% (Rompun ® Bayer). When anaesthesia was obtained, the rats in group A were sacrificed without the application of any electrical current. For the rats in group B, two groups of electrodes were placed on the right upper and left upper extremities, and the electrical current was applied. These extremities were selected as the most frequently observed route of electrical current in the body in cases of electric shock. For group C, water of 4 ± 1 cm depth was added to a plastic bowl so that it would not reach the airway of the rats. A large container was filled with a large amount of tap water at the beginning of the experiment. The water in the experimental setup was changed for each experiment subject in group C with the same volume. The rats in group C were left into the water, and two free-ended electrode probes (phase and neutral) were placed at the same distance from the head and tail for each one. Two hundred twenty V electric current was applied to the subjects in the experimental setup until they died (approximately 153 ± 27 s). All the animals in the study groups and control group were sacrificed, and then the lower and upper extremities were shaved, and skin samples were taken. In all the subjects, the chest was opened, and blood samples were taken from the vena cava, and the heart was dissected. The skull was opened, and the brain, cerebellum, and brainstem dissections were made. The liver dissections were performed by opening the abdomen. The organs were fixed in 10% formaldehyde solution. Blood samples were centrifuged at 3000 rpm for 10 min, and the serum samples obtained were stored at − 70° until assay.

Biochemical analyses

The numbered serum samples were examined with the NT-ProBNP kit using the chemiluminescence method on a Siemens Immulite-2000 model device (Siemens Healthcare Diagnostics Products Ltd. Llanberis, Gwynedd LL55 4EL UK). In the same way, the serum samples were prepared according to the standard assay principles defined in the H-FABP test kit (Rat H-FABP Cloud Clone Corp. 23,603 W. Fernhurst Dr. Unit 2201, Katy, TX, USA). Analysis was made with the enzyme-linked immunosorbent assay kit (ELISA) method on a BioTek Synergy H1 model ELISA device. Serum H-FABP levels were measured as ng/ml.

Histopathological and immunohistochemical analyses

For histopathological examination, the tissues fixed in formalin were embedded in paraffin blocks, and then Sects. 4 µm in thickness were cut and stained with hematoxylin and eosin (H&E). The immunohistochemical staining was performed in a fully automated immunohistochemistry staining device (Ventana BenchMark Ultra, Ventana Medical Systems, Tucson, AZ, USA).

The primer antibodies used were Bax (Boster Biological Technology, Anti-Bax Rabbit Monoclonal Antibody, Uniprot ID/Q07812) at dilution of 1/200, caspase-3 (Boster Biological Technology, Anti-Caspase-3 Rabbit Monoclonal Antibody, Uniprot ID/P42764) at dilution of 1/500 and HSP-60 (Boster Biological Technology, Anti-HSP60/HSPD1 Picoband Rabbit Monoclonal Antibody, Uniprot ID/P42764) at dilution of 1/2000.

Colon adenocarcinoma tissue was used as the positive control for Bax and tonsil tissue for caspase-3 and HSP-60. The H&E-stained sections and immune antibodies were evaluated under light microscope by two pathologists. The immunohistochemical staining was scored semi-quantitatively (Table 2).

Table 2 Staining criteria in immunohistochemical evaluation

	Staining prevalence	Staining degree
No staining	0	0
Light staining	< %10	1
Moderate staining	%10–50	2
Severe staining	> %50	3

Statistical analyses

The data used in the statistical analysis were stated as median (minimum–maximum) values. As the variables were sequencer discrete variables, the non-parametric Kruskal–Wallis *H* test was applied to determine whether the difference between these variables was statistically significant. Conformity to normal distribution of the continuous numerical variables was assessed with the Shapiro–Wilk test. Multiple comparisons after the Kruskal–Wallis *H* test were made with the Conover test. A value of $p < 0.05$ was accepted as statistically significant.

Results

Biochemical results

The results obtained from the statistical analysis of serum H-FABP (ng/ml) and NT-ProBNP levels at 0 h are shown in Table 3. The serum H-FABP levels of group C (electric shock with water conduction) were observed to be statistically significantly lower than those of the other two groups ($p < 0.05$). Serum NT-ProBNP values were found to be < 20,00 pg/ml in blood samples taken at 0 h for all subjects in groups 1, 2 and 3. Therefore, statistical analysis was not performed.

Histopathological and immunohistochemical results

No significant pathological change was determined in the H&E staining of the brain, cerebellum, brainstem, heart and liver tissue samples of all the subjects. In group B subjects (isolated electric shock), there were observed to be demonstrative findings of electric shock (extension in the epidermal nuclei, dermo-epidermal separation, homogenizaton in the dermis) in the skin samples taken from the electric entry–exit points. The caspase-3, Bax and HSP-60 immunohistochemical staining samples of the groups are shown in Figs. 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12 and 13. The immunohistochemical staining results are shown in Table 4.

Caspase-3

No statistically significant difference was determined between group B and group C in the brain and brainstem samples. The antibody expression in both groups was determined to be significantly higher than in group A ($p < 0.05$). A statistically significant increase was observed between group A and group C in respect of the cerebellum sample ($p < 0.05$). In the heart, liver and upper and lower extremity skin samples, statistically significant differences were determined between all the groups ($p < 0.05$).

Bax

In the cerebellum and brain stem samples, no statistically significant difference was determined between group B and group C, and a statistically significant increase was seen in both these groups compared to group A ($p < 0.05$). In the brain, heart, upper and lower extremities and skin samples, statistically significant differences were determined between all the groups ($p < 0.05$). In the pericentral zone of the liver tissue samples, no statistically significant difference was determined between group B and group C, and a statistically significant increase was seen in both these groups compared to group A ($p < 0.05$). In the staining of the liver zone 2 regions, a statistically significant difference was determined between all the groups ($p < 0.05$).

Table 3 Serum H-FABP levels of the groups

	Group A (control)	Group B	Group C	*P**
H-FABP (ng/ml)				
Median (min–max)	23.73[b] (6.75–63.73)	40.36[b] (10.79–63.73)	10.85 (3.09–63.73)	**0.0037**

Significant at $p < 0.05$

[a]Different from group B

[b]Different from group C

*Kruskal–Wallis test

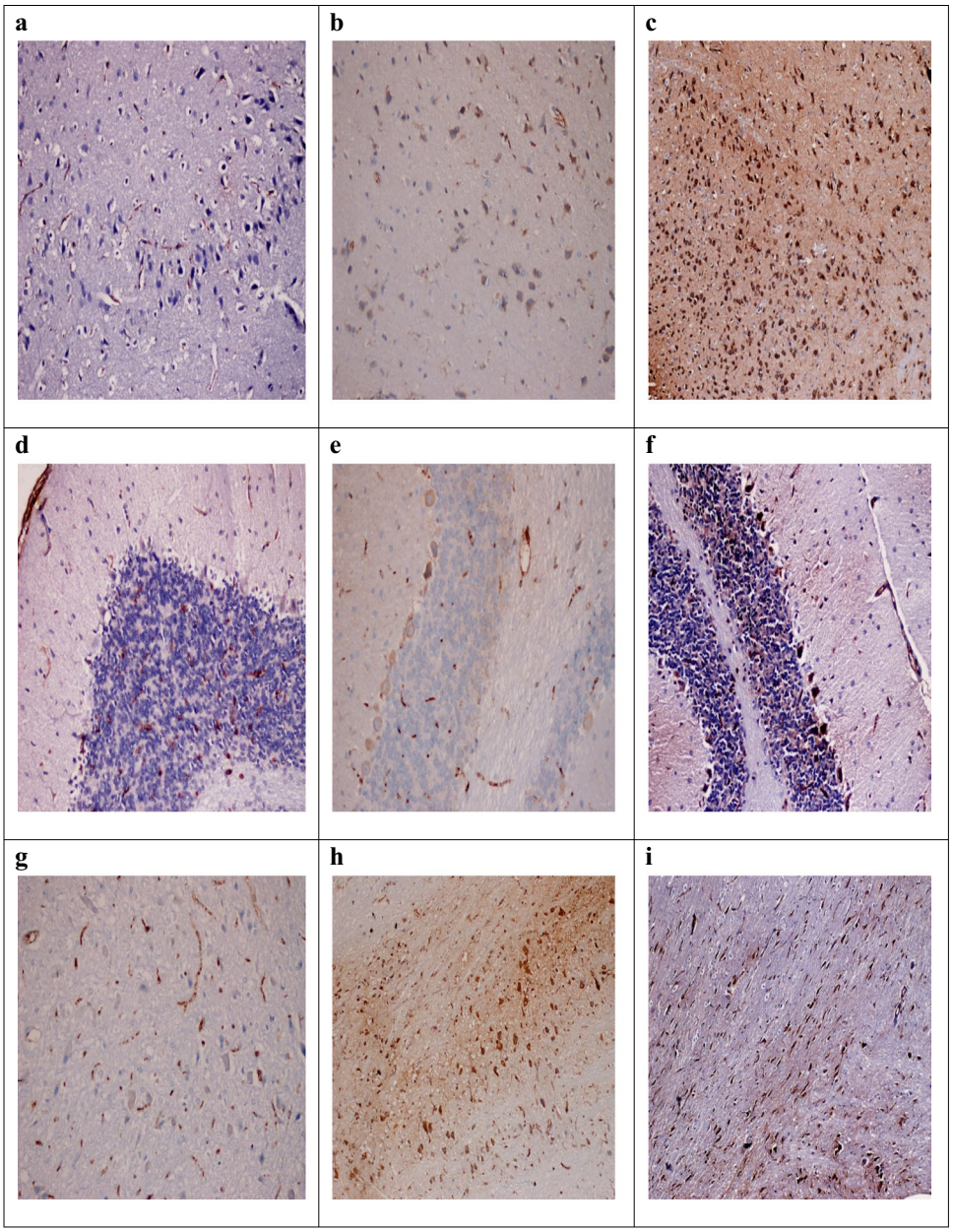

Fig. 2 Immunohistochemical expression of caspase-3 in the cerebrum, cerebellum and brainstem: **a** control, cerebrum score 0, × 200; **b** group 2 cerebrum, score 1, × 200; **c** group 3 cerebrum, score 3, × 100; **d** control cerebellum, score 0, × 200; **e** group 2 cerebellum Purkinje cells, score 1, × 200; **f** group 3 cerebellum Purkinje cells, score 3, × 200; **g** control brainstem, score 0, × 200; **h** group 2 brainstem, score 3, × 100; **i** group 3 brainstem, score 3, × 100

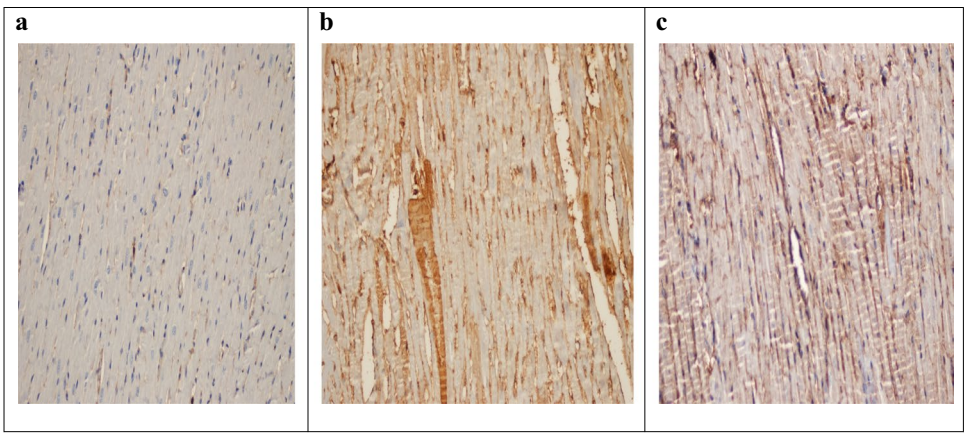

Fig. 3 Immunohistochemical expression of caspase-3 in the heart: **a** control, score 0, × 200; **b** group 2, score 1, × 200; **c** group 3, score 2, × 200

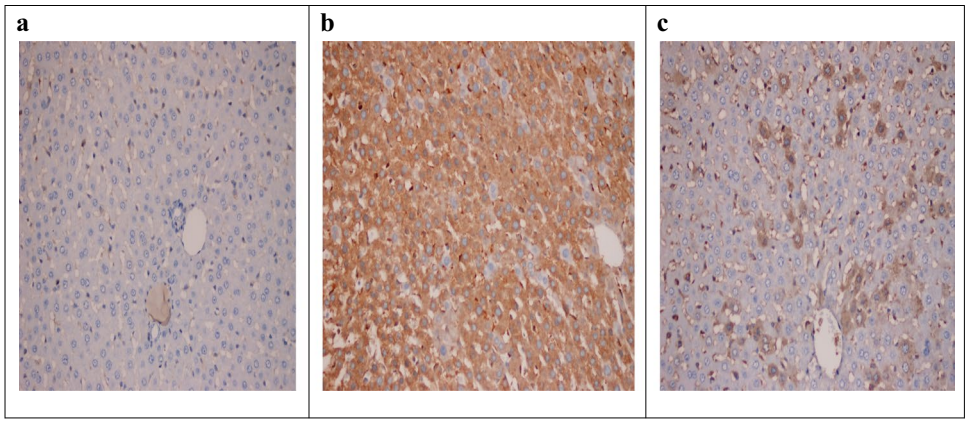

Fig. 4 Immunohistochemical expression of caspase-3 in the liver: **a** control, score 0, × 200; **b** group 2, score 3, × 200, **c** group 3, score 1, × 200

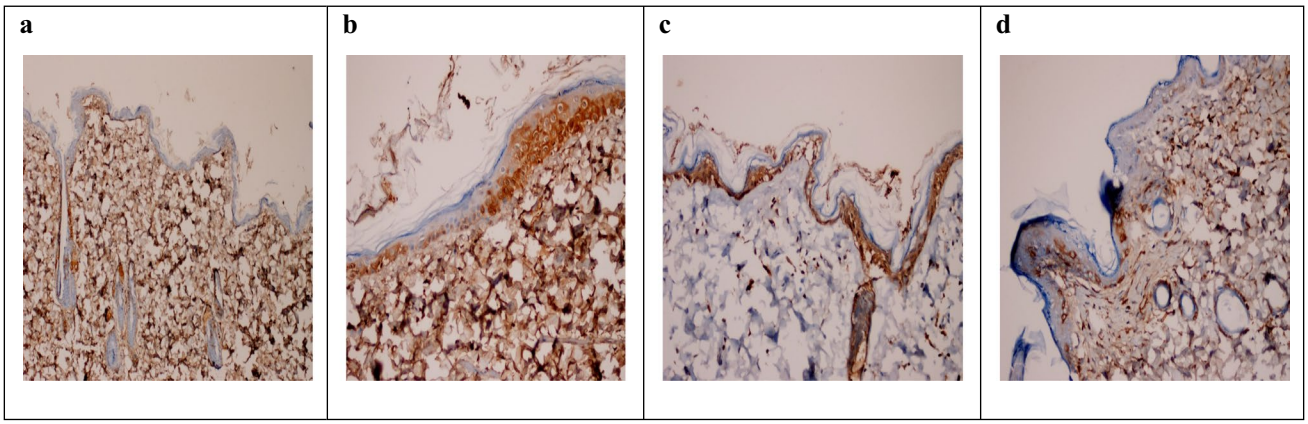

Fig. 5 Immunohistochemical expression of caspase-3 in the skin: **a** control, score 0, × 100, **b** group 2 lower extremity, score 2, × 200; **c** group 2 upper extremity, score 3, × 200; **d** group 3 lower extremity, score 1, × 200

HSP-60 In the staining of the brain, cerebellum, brain stem and liver samples, no statistically significant difference was determined between group B and group C, and a statistically significant increase was seen in both these groups compared to group A ($p < 0.05$). In the heart and upper extremity skin samples, a statistically significant difference was determined between all the groups ($p < 0.05$). In the lower extremity skin samples, a statistically significant increase was seen in group B compared to group A ($p < 0.05$).

Discussion

Death related to electric shock is a process frequently involving cardiac arrythmia or paralysis of the respiratory muscles [4]. Several studies have reported serum NT-ProBNP level as a parameter sensitive to cardiac damage. NT-ProBNP has also been shown to be a good marker in chronic cardiac ischaemia and heart failure [13]. In the current study, the NT-ProBNP levels were within normal limits in all the groups, which was attributed to the fact that none of the animals had any chronic cardiac pathology.

Although the serum H-FABP level is a marker sensitive to cardiac damage, there are few studies of serum H-FABP values associated with electric shock and encompassing electric shock with water conduction [6, 12]. In an experimental study by Özdemir C et al., the electricity of different voltage was applied to animals, and with increasing voltage, there was seen to be a statistically significant difference between the groups in respect of serum H-FABP [6]. In the current study, no significant difference was determined between the control group and group B, the group applied with isolated electric shock at 220 V, in respect of 0-h H-FABP values. However, the serum H-FABP values of group C, applied with electric shock with water conduction, were determined to be statistically significantly lower than those of groups A and B. This suggests that serum H-FABP values taken after electric shock could be a valuable finding in relation to the question of whether there was any water conduction.

As the CNS has low resistance to electric current, this can cause peripheral and central neurological complications, which may emerge early or late [7]. Although there

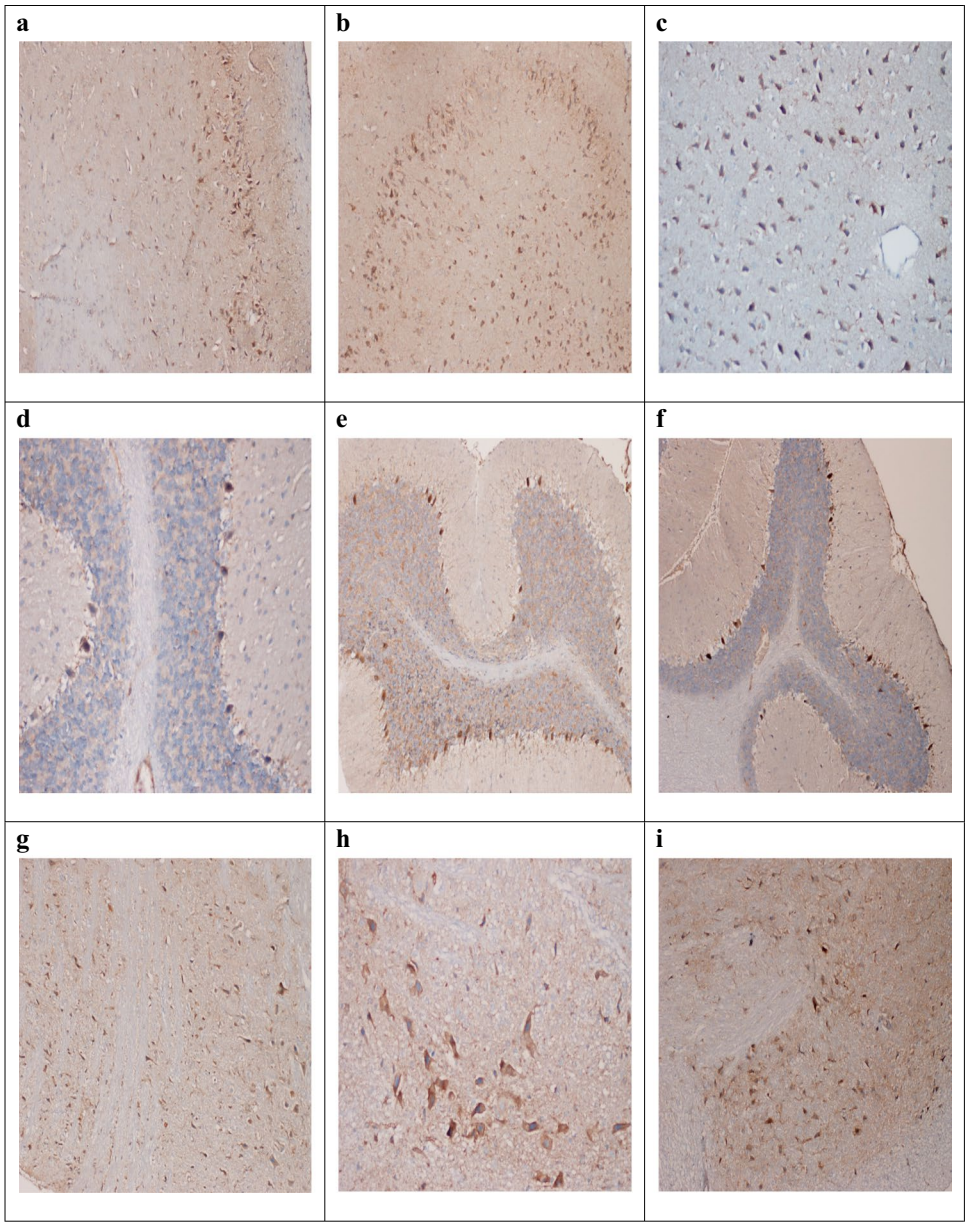

Fig. 6 Immunohistochemical expression of Bax in the cerebrum, cerebellum and brainstem: **a** control cerebrum, score 1, × 100; **b** group 2 cerebrum, score 2, × 100; **c** group 3 cerebrum, score 2, × 200; **d** control cerebellum, score 1, × 200; **e** group 2 cerebellum Purkinje cells, score 3, × 100; **f** group 3 cerebellum Purkinje cells, score 2, × 100; **g** control brainstem, score 1, × 100; **h** group 2 brainstem, score 2, × 200; **i** group 3 brainstem, score 2, × 100

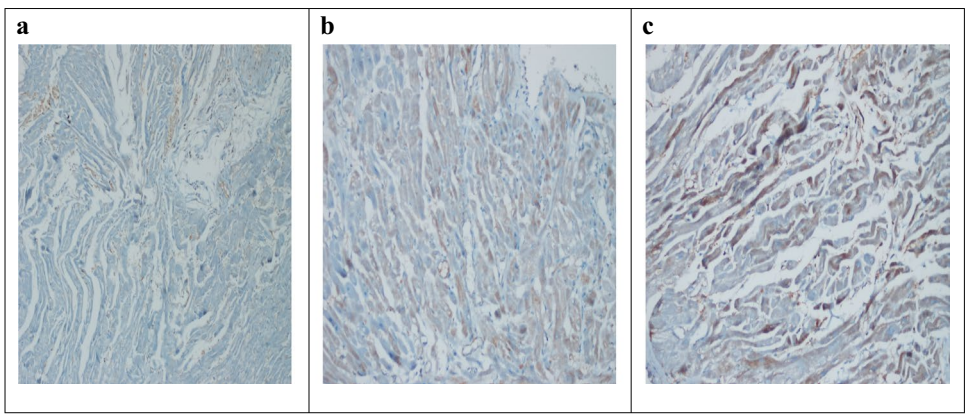

Fig. 7 Immunohistochemical expression of Bax in the heart: **a** control, score 0, × 100; **b** group 2, score 1, × 200; **c** group 3, score 1, × 200

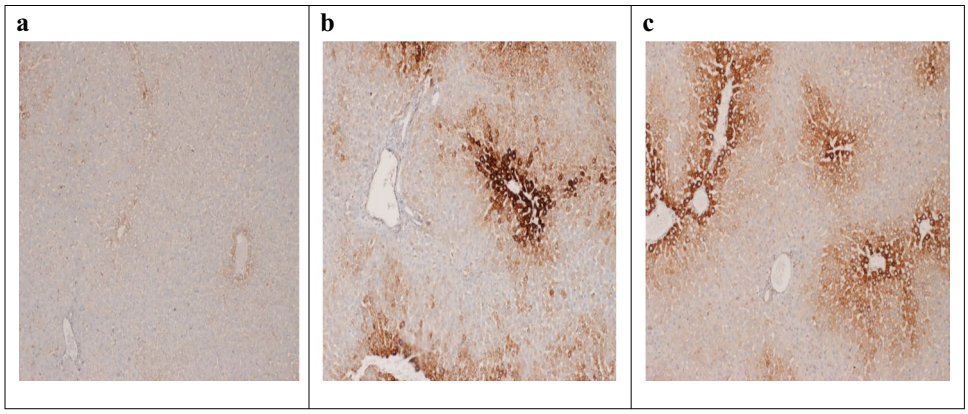

Fig. 8 Immunohistochemical expression of Bax in the liver: **a** control, pericentral zone score 1, × 100; **b** group 2, pericentral zone score 3, × 100; **c** group 3, pericentral zone score 2, × 100

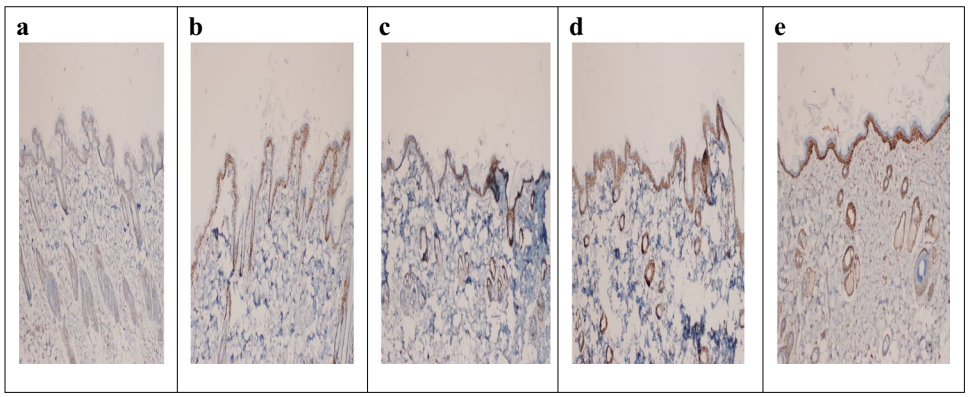

Fig. 9 Immunohistochemical expression of Bax in the skin: **a** control, score 1, × 100; **b** group 2 lower extremity score 2, × 100; **c** group 2 upper extremity, score 2, × 100; **d** group 3 lower extremity, score 2, × 100; **e** group 3 upper extremity, score 3, × 100

are different studies in literature showing the effect of electric shock on the central nervous system, there are very few studies related to the cerebellum and brainstem [7, 14–16]. In studies by Brasko et al. and Larsen et al. to determine the cause of death associated with electric shock, there were seen to be several morphological changes in nerve cells, especially in the cerebellar Purkinje cells [14, 15]. Due to low resistance, the cerebellar Purkinje cells are one of the regions of the CNS most affected by electric current [16, 17].

Although the contact points of the electric current are far from the CNS, damage can occur at any point of the CNS [18, 19]. This indicates that there is an indirect electric current path that can damage the cerebellum, and this may be due to the electrical conductivity of the CNS [18]. A limited number of immunohistochemical studies have been conducted using apoptosis markers and related to histomorphological reflections [8]. Kandeel et al. reported that while the Purkinje cells gave no immune reaction with caspase-3 in the control group, there was a significant increase in immunohistochemical staining with caspase-3 in the group applied with fatal electric shock, and there was a significant increase in the number of caspase-3 positive Purkinje cells in the groups administered non-fatal electric shock [8]. Similarly in the current study, the caspase-3 staining in groups B and C, especially in the brain stem, was similar and was evaluated as score 3. In both groups, a significant difference was observed between the staining of the brainstem neurons and the staining of the brain–cerebellum neurons. Similar results were observed in the Bax and HSP-60 stainings. These results suggest that death related to electric shock could occur through damage to the respiratory centre in particular. However, the intensity of caspase-3 immunohistochemical staining in tissues (brain, cerebellum, brainstem) does not differentiate cases of wet or isolated electric shock.

In a previous experimental study, Bax expression was examined immunohistochemically in the myocardium after electric shock, and the expression levels in the groups with electrical injury were determined to be significantly higher than those of the control group [9]. Similarly in the current study, a statistically significant increase was determined in the Bax staining of the heart tissues in group B compared to group A ($p < 0.001$). Similar results were obtained in group C, and statistically significant differences were determined between all the groups ($p < 0.001$). An increase was seen in pro-apoptosis in the heart tissue in electric shock, and Bax expression in isolated electric shock was greater than in electric shock with water conduction.

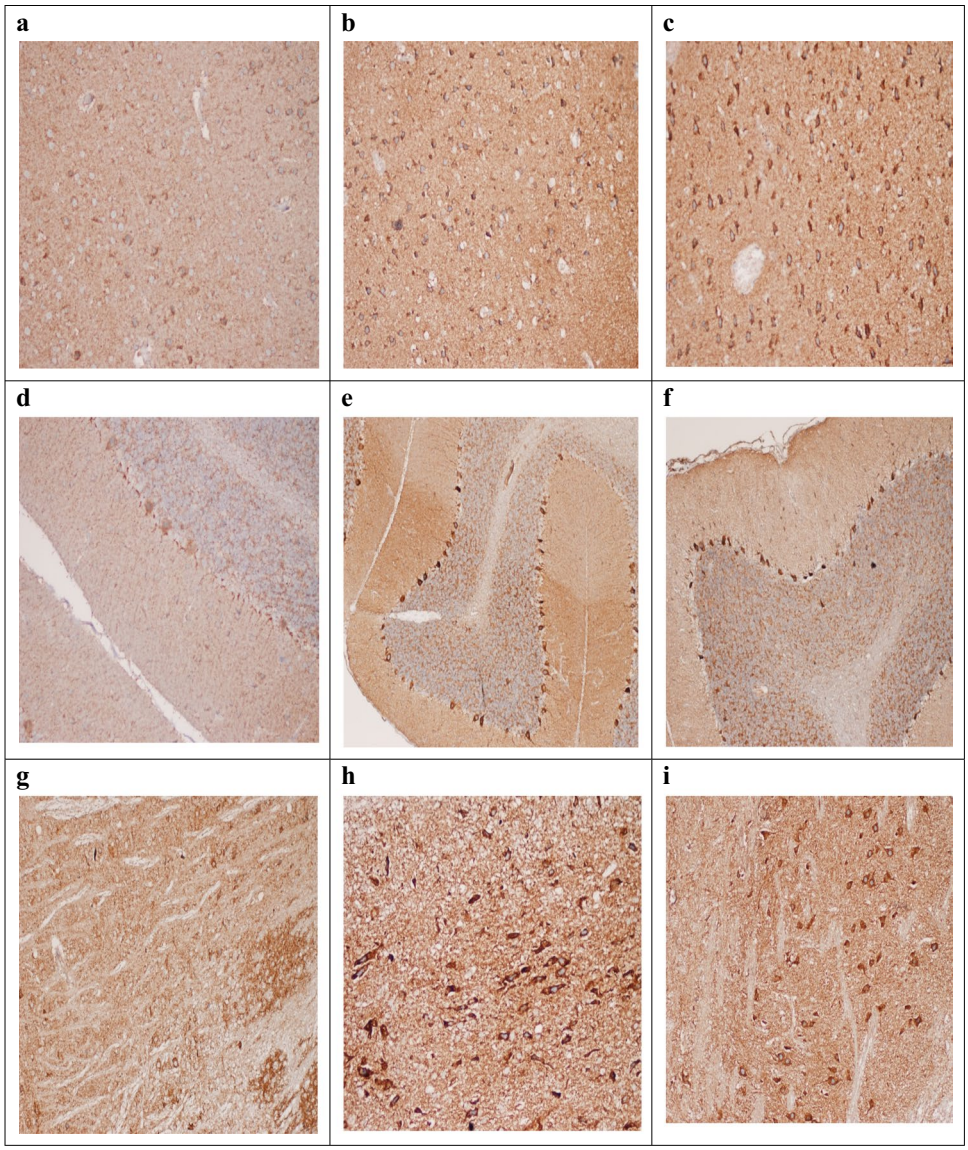

Fig. 10 Immunohistochemical expression of Hsp-60 in the cerebrum, cerebellum and brainstem: **a** control cerebrum, score 1, × 200; **b** group 2 cerebrum, score 2, × 200; **c** group 3 cerebrum, score 3, × 200; **d** control cerebellum, score 1, × 200; **e** group 2 cerebellum Purkinje cells, score 3, × 100; **f** group 3 cerebellum Purkinje cells, score 3, × 100; **g** control brainstem, score 1, × 100; **h** group 2 brainstem, score 3, × 200; **i** group 3 brainstem, score 3, × 100

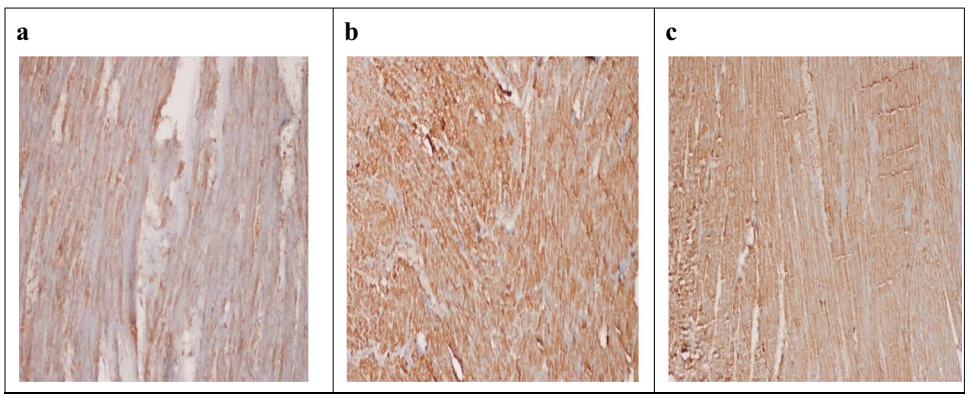

Fig. 11 Immunohistochemical expression of Hsp-60 in the heart: **a** control, score 1, × 200; **b** group 2, score 3, × 100; **c** group 3, score 2, × 100

Huitong Liu et al. showed that in conditions of stress, the expression of stress proteins was activated to assist in the repair of thermal damage and electrostimulation [9]. In the current study, staining with HSP-60 antibody in the heart was increased in all the groups, and the increase in the isolated electric shock group was determined to

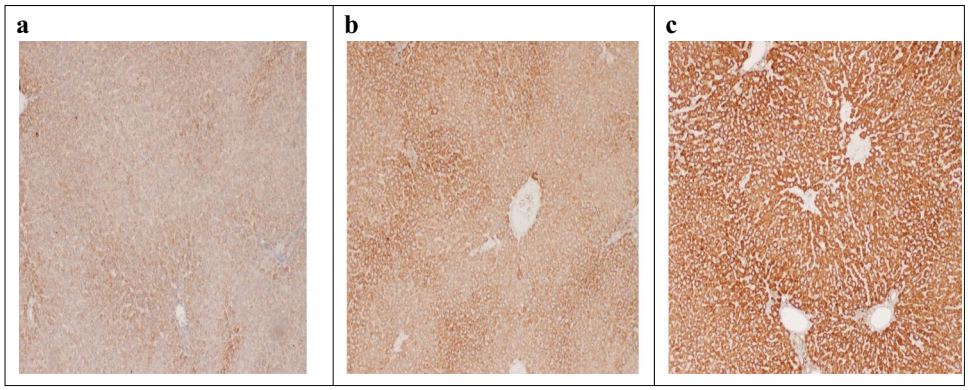

Fig. 12 Immunohistochemical expression of Hsp-60 in the liver: **a** control, score 1, × 100; **b** group 2, score 2, × 100; **c** group 3, score 3, × 100

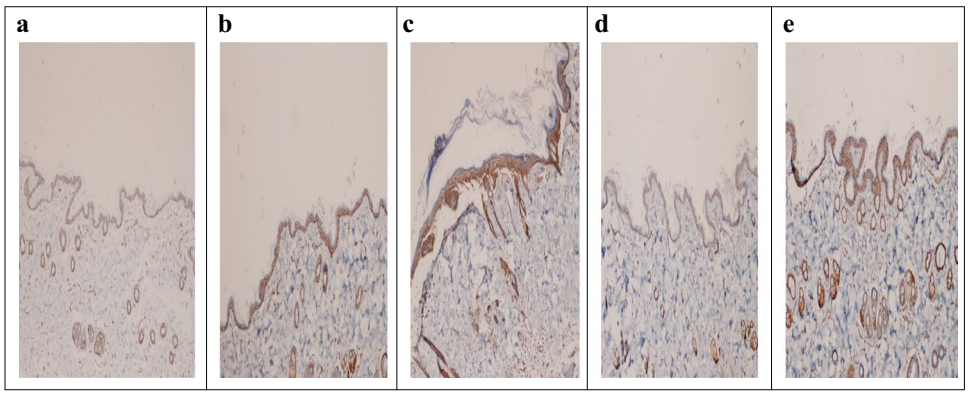

Fig. 13 Immunohistochemical expression of Hsp-60 in the skin: **a** control, score 1, × 100; **b** group 2 lower extremity, score 2, × 100; **c** group 2 upper extremity, score 3, × 100; **d** group 3 lower extremity, score 1, × 100; **e** group 3 upper extremity, score 2, × 100

be statistically significant. In addition, the difference between group A and group B was seen to be statistically significantly different from group C. Again in the study by Huitong et al., the group where the most severe HSP-60 expression was seen in the liver was the group applied with fatal electric shock, and this level of HSP-60 expression was reported to be an indicator that severe destruction had occurred [9]. In the current study, HSP-60 expression in both the isolated electric shock group and the electric shock with water conduction group was observed to be significantly increased especially in the pericentral zone, compared to the control group. These immunohistochemical findings support the view that an electric current forms damage in the liver. Similar to these findings for HSP-60, there was a statistically significant difference between all the groups in respect of the apoptosis markers in the zone 3 region of the liver with caspase-3 and in the pericentral and zone 2 regions of the liver with Bax.

As typical histopathological findings which can be observed on the skin associated with electric shock (vacuoles in the stratum corneum, extension in the cell nucleus, dermo-epidermal separation, homogenization in the dermis) may be seen in several situations such as freezing and burns, these findings are not pathognomonic [1, 2, 20]. In the current study, statistically significant differences were seen between all three groups in the staining with immunohistochemical markers (caspase-3, Ba, HSP-60) of skin samples taken from the upper extremities. Compared to the control group, there was increased staining in the groups exposed to the electric currents, and the increase in group B (isolated electric current) was significant.

Similarly, there were seen to be statistically significant differences between all the groups in the caspase-3 and Bax staining in the lower extremity skin samples. In the staining with HSP-60, only the difference between groups A and B was statistically significant. In addition, the staining in the upper extremities was seen to be more intense than in the lower extremities. It was thought that this could be due to the severity of the burn injury formed in the skin with direct contact of the isolated electric shock. As no previous study could be found in literature which had similarly examined immunohistochemical markers in the skin tissue of cases with death due to electric shock, these findings could form the foundation of future studies.

The results of this study showed that there were statistically significant differences in the immunohistochemical

Table 4 Immunohistochemical evaluation of the groups

	Group A (control)	Group B	Group C	Kruskal–Wallis test
Brain				
Median (min–max)	0[a,b] (0–1)	0 (0–1)	1 (0–3)	***p* = 0.0175**
Caspase-3	0[a,b] (0–1)	1.5[b] (1–3)	1 (0–2)	***p* < 0.001**
Bax	1[a,b] (0–1)	1 (1–3)	2 (1–3)	***p* < 0.001**
HSP-60				
Cerebellum				
Median (min–max)	0[b] (0–0)	0 (0–2)	0 (0–3)	***p* = 0.0456**
Caspase-3	0.5[a,b] (0–1)	1.5 (0–3)	2 (1–3)	***p* < 0.001**
Bax	1[a,b] (1–2)	3 (2–3)	3 (2–3)	***p* < 0.001**
HSP-60				
Brainstem				
Median (min–max)	0[a,b] (0–1)	3 (0–3)	3 (3–3)	***p* < 0.001**
Caspase-3	1[a,b] (0–1)	2 (1–3)	2 (1–3)	***p* < 0.001**
Bax	1[a,b] (1–3)	3 (2–3)	3 (2–3)	***p* < 0.001**
HSP-60				
Heart				
Median (min–max)	0[a,b] (0–0)	1[b] (0–2)	2 (0–2)	***p* < 0.001**
Caspase-3	0[a,b] (0–1)	1[b] (0–1)	0 (0–1)	***p* < 0.001**
Bax	1[a,b] (1–1)	3[b] (1–3)	1.5 (1–3)	***p* < 0.001**
HSP-60				
Liver				
Median (min–max)	0[a,b] (0–0)	3[b] (2–3)	1 (1–1)	***p* < 0.001**
Caspase-3	1[a,b] (1–1)	3 (2–3)	3 (2–3)	***p* < 0.001**
Bax (pericentral zone)	0[a,b] (0–0)	1[b] (0–3)	2 (0–2)	***p* < 0.001**
Bax (Zon-2)	1[a,b] (1–1)	2 (1–3)	2 (1–3)	***p* < 0.001**
HSP-60				
Skin (upper extremity)				
Median (min–max)	0[a,b] (0–0)	3[b] (2–3)	1 (0–1)	***p* < 0.001**
Caspase-3	0[a,b] (0–0)	3[b] (2–3)	1 (0–1)	***p* < 0.001**
Bax	1[a,b] (1–1)	3[b] (2–3)	2 (1–3)	***p* < 0.001**
HSP-60				
Skin (lower extremity)				
Median (min–max)	0[a,b] (0–0)	2[b] (1–2)	1 (0–1)	***p* < 0.001**
Caspase-3	0[a,b] (0–0)	2[b] (1–2)	1 (0–1)	***p* < 0.001**
Bax	1[a] (1–1)	2 (1–2)	1 (1–2)	***p* < 0.001**
HSP-60				

*Significant at $p < 0.05$. [a]Different from group B. [b]Different from group C

staining (caspase-3, Bax, HSP-60) in the brain, cerebellum, brainstem, heart, liver and upper and lower extremity skin samples and in the blood serum biochemical parameters (H-FABP) as a result of the damage caused by an electric current. To the best of our knowledge, there has been no previous experimental study in literature related to electric shock with water conduction.

In conclusion, this is the first study to have examined isolated electric shock and electric shock with water conduction in an experimental animal model. With the use of immunohistochemical and biochemical markers in this study, the findings can be considered to contribute to the clarification of one of the reasons for negative autopsy in forensic medicine. However, comparative studies are needed on real victims of electric shock in water.

Author contribution Research concept and design: Ahmet Sedat Dündar, Osman Celbiş and Emine Türkmen Şamdancı.

Collection and/or assembly of data: Ahmet Sedat Dündar, Hasan Okumuş and Onural Özhan.

Data analysis and interpretation: Çağatay Taşkapan, Ayşenur Akatlı and Mucahit Oruç.

Writing the article: Ahmet Sedat Dündar and Mucahit Oruç.

Critical revision of the article: Çağatay Taşkapan and Ayşenur Akatlı.

Final approval of article: Osman Celbiş, Emine Türkmen Şamdancı and Hakan Parlakpınar.

Funding This research was supported by Inönü University Scientific Research Projects with 924 project ID code.

Declarations

Conflict of interest The authors declare no competing interests.

References

1. Saukko PJ, Knight B (2004) Knight's forensic pathology, 3rd edn. Hodder Arnold, London, New York
2. Di Maio VJ, Di Maio D (2001) Forensic pathology, 2nd edn. CRC Press, Boca Raton
3. Cohle SD, Sampson BA (2001) The negative autopsy: sudden cardiac death or other? Cardiovasc Pathol 10:219–222. https://doi.org/10.1016/s1054-8807(01)00093-x
4. Wang Y, Liu M, Cheng W, Liao FLZ, Wang Y (2008) Endothelial cell membrane perforation of aorta and pulmonary artery in the electrocution victims. Forensic Sci Int 178:204–206. https://doi.org/10.1016/j.forsciint.2008.04.003
5. Fineschi V, Di Donato S, Mondillo S, Turillazzi E (2006) Electric shock: cardiac effects relative to non fatal injuries and postmortem findings in fatal cases. Int J Cardiol 111:6–11. https://doi.org/10.1016/j.ijcard.2005.07.060
6. Özdemir Ç, Asil H, Yazıcı C, Akgün H, Akcay A (2016) Heart-type fatty acid binding protein and cardiac troponin I may have a diagnostic value in electrocution. J Forensic Leg Med 39:76–79. https://doi.org/10.1016/j.jflm.2016.01.015
7. Kurtulus A, Acar K, Adiguzel E, Boz B (2009) Hippocampal neuron loss due to electric injury in rats: a stereological study. Legal Med 11:59–63. https://doi.org/10.1016/j.legalmed.2008.08.001
8. Kandeel S, Elhosary NM, El-Noor MM, Balaha M (2017) Electric injury-induced Purkinje cell apoptosis in rat cerebellum: histological and immunohistochemical study. J Jchemneu 81:87–96. https://doi.org/10.1016/j.jchemneu.2017.02.010
9. Huitong L, Qiaofeng W, Ze Z, Yanan X, Suzhen D, ZhenyuanW (2016) The clinical and medicolegal analysis of electrical shocked rats: based on the serological and histological methods. BioMed Res Int 1-12. https://doi.org/10.1155/2016/4896319
10. Price TG, Cooper MA (2014) Electrical and lightning injuries. In: Marx JA, Hockberger RS, Walls RM, Biros MH (eds) Rosen's emergency medicine: concepts and clinical practice. Elsevier Saunders, Philadelphia, pp 1906–1914
11. Dzhokic G, Jovchevska J, Dika A (2008) Electrical injuries: etiology, pathophysiology and mechanism of injury. Macedonian J Med Sci 1:54–58
12. Aartsen WM, Pelsers M, Hermens WT, Glatz J, Daemen M, Smits J (2000) Heart fatty acid binding protein and cardiac troponin T plasma concentrations as markers for myocardial infarction after coronary artery ligation in mice. Eur J Physiol 439:416–422. https://doi.org/10.1007/s004249900180
13. Zhu BL, Ishikawa T, Michiue T et al (2007) Postmortem pericardial natriuretic peptides as markers of cardiac function in medicolegal autopsies. Int J Legal Med 121:28–35. https://doi.org/10.1007/s00414-006-0102-1
14. Brasko J, Rai P, Sabol MK, Patrikios P, Ross DT (1995) The AMPA antagonist NBQX provides partial protection of rat cerebellar Purkinje cells after cardiac arrest and resuscitation. Brain Res 699:133–138. https://doi.org/10.1016/0006-8993(95)01015-n
15. Larsen A, Stoltenberg M, West MJ, Danscher G (2005) Influence of bismuth on the number of neurons in cerebellum and hippocampus of normal and hypoxia-exposed mouse brain: a stereological study. J Appl Toxicol 25:383–392. https://doi.org/10.1002/jat.1061
16. Winkelman MD (2008) Neurological complications of thermal and electrical burns. In: Aminoff MJ, Josephson SA (eds) Amin Off's Neurology and General Medicine, 4th edn. Churchill Livingstone, pp 1040–1045
17. Schulze C, Peters M, Baumgartner W, Wohlsein P (2016) Causes, pathogenesis and morphological findings. Vet Pathol 53:1018–1029. https://doi.org/10.1177/0300985816643371
18. Park E, McKnight S, Ai J, Baker AJ (2006) Purkinje cell vulnerability to mild and severe forebrain head trauma. J Neuropathol Exp Neurol 65:226–234. https://doi.org/10.1097/01.jnen.0000202888.29705.93
19. Pliskin NH, Meyer GJ, Dolske MC (1994) Neuropsychiatric aspects of electrical injury: a review of neuropsychological literature. Ann N Y Acad Sci 720:213–218. https://doi.org/10.1111/j.1749-6632.1994.tb30450.x
20. Visonàa SD, Chena Y, Bernardib P, Andrelloc L, Osculatia A (2018) Diagnosis of electrocution: the application of scanning electron microscope and energy-dispersive X-ray spectroscopy in five cases. Forensic Sci Int 284:107–116. https://doi.org/10.1016/j.forsciint.2018.01.001