Electric injury-induced Purkinje cell apoptosis in rat cerebellum: Histological and immunohistochemical study

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ABSTRACT

Introduction: Electrical injury is a prominent problem in low income countries with increased morbidity and mortality rate. Nervous system is one of the most susceptible systems to electrical current because of its low resistance. There were different studies demonstrated electrocution effect on the nervous system, however little was made on the cerebellum.

Aim: This study was conducted to produce an experimental suggestion concerning injury of the nervous system through evaluating Purkinje cell apoptosis and number in rat cerebellum by fatal and non-fatal electric current using histological and immunohistochemical study. Also to support the diagnosis of electrocution as a probable cause of death and delayed neurological damage as well as disability.

Materials & methods: Fifty male Wistar rats were divided into five groups (10 rats each); control group: normal rats that were sacrificed without exposure to electric current, groups 1–3 (non-fatal electrocution groups): rats were exposed to alternating electric current (220 v, 50 Hz) for one minute then were sacrificed immediately, after 2 h, and after 4 h respectively, and group 4 (fatal electrocution group): rats were sacrificed after being electrified up to death (153 ± 27 s). Sections from cerebellum were processed for histological and caspase-3 immunohistochemical study.

Results: Purkinje cells showed marked histopathological changes in the form of shrunken dark stained cells with significant reduction of their number in H & E stained sections when compared to control, widespread argyrophilia, and degenerated organelles along with shrunken irregular nuclei. For caspase-3 staining, there was significantly increased number of caspase-3 positive cells in groups 1–3 (non-fatal electrocution groups) and markedly increased in group 4 (fatal electrocution group) in comparison to control group. These changes were gradually increased with the increased duration after exposure to the electric current.

Conclusion: Apoptosis and loss of Purkinje cells were involved in the pathogenesis of immediate and long term effect of electrical injury on Purkinje cells, which will be an aid to the forensic pathologist to determine the cause of death and residual damage as well as disability after electric shock.

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1. Introduction

Electrical injury due to contact with electrical circuits has become a prominent problem because of the extensive use of electricity in our life (Mashreky et al., 2012). This type of injury could result in direct effect of the electrical current on the body or burns from ignition of clothes by striking the skin with heat dissipation or may lead to trauma either from associated intense muscular contractions or from associated falls (Wang et al., 2005; Fish and Geddes, 2009; Jayanth et al., 2014). The degree of injury depends on the type, and intensity of the electrical current, the current pathway, duration of contact, location of damage and the physical conditions under which the event takes place (e.g. Wet environment or dry environment) (Duff and McCaffrey, 2001).

Electrocution has been defined as a death caused by electric shock through whom a sudden, violent response of a person’s body occurs due to electric current flow (Viswakanth and Shruthi, 2015). It is mostly unintentional; occurs in children in domestic environment or in adults as an occupational hazard (Nguyen et al., 2004;
Arkuszewski et al., 2014). Immediate death that occurs after contact with circuits at low voltages is supposed to be due to ventricular fibrillation. However, the cause of death after high voltage circuits, thought to be due to central inhibition of the nervous system with subsequent respiratory failure (Jia-ke et al., 2009).

Electric injury can affect many body tissues including nervous system which is one of the most susceptible systems to the electrical current because of its low resistance. Electrical shock can result in neurological complications, involving both peripheral and central nervous systems which may present immediately or later on (Oehmichen et al., 2003; Kurtulus et al., 2009). Additionally, Brasko et al. (1995) and Larsen et al. (2005) revealed many morphological changes of the nerve cells due to electric injury which may be an area of interest in forensic pathology in order to determine the cause of death.

The cerebellum is an important organ for motor function, cognition, and emotion (Strata et al., 2011; Lisberger and Thach, 2013). Its cortex is made of molecular, Purkinje, and granular layers. Cerebellar Purkinje cells are the sole output neurons of the cerebellar cortex that project to the deep cerebellar and vestibular nuclei (Lisberger and Thach, 2013). It plays a fundamental role in controlling the motor movement through the release of gamma-aminobutyric acid; a neurotransmitter which exerts inhibitory actions on certain neurons and thereby reduces the transmission of nerve impulses thus regulate and coordinate motor movements (Louis et al., 2014).

There were different studies demonstrated the effect of electrocution on the nervous system like that made by Kurtulus et al. (2009), however little was made on the cerebellum. Moreover, cerebellar Purkinje cells considered as one of the largest neuron of the nervous system with many branching extensions, so any pathological changes could be easily seen. Additionally, cerebellar Purkinje neurons are found to be one of the most affected regions in the central nervous system by electrical currency because of its low resistance. In animals, cerebellar Purkinje cells seem to be particularly vulnerable to electrical current (Winkelman, 2008).

The delayed neurological complications of an electric shock are of significant medicolegal importance where an electric occupation or accident may result in lifelong neurological disability requiring large financial compensation (Winkelman, 2008). One of the most difficult problems in forensic pathology is to trace a chain of events from the injury to the disability to prove that the injury is the basic cause. This causal relationship is crucial for civil compensation (Jayanth et al., 2014).

Apoptosis is the process of programmed cell death, and is regulated by multiple genes (Wang et al., 2006). Neuronal apoptosis play a major role in brain development and many neurodegenerative diseases and neuronal cell death from brain ischemia has been found to be mediated by caspase-3 which exists as an inactive pro-enzyme that go through proteolytic processing to form active caspase-3 enzyme (D’Amelio et al., 2012).

In this light, our study was conducted to produce an experimental suggestion concerning injury of the nervous system by electrical current through histological and caspase-3 immunohistochemical evaluation of rat cerebellar Purkinje cells in case of fatal and non-fatal electric injury to support the diagnosis of electrocution as a probable cause of death and delayed neurologi- cal damage as well as disability.

2. Materials and methods

2.1. Animals

This study was carried out on 50 adult male Wister rats weighing 200 – 250 g, obtained from the animal house, Faculty of Medicine, Tanta University, Egypt. All animals were housed in plastic cages at a temperature of 22 ± 1 °C, with a relative humidity (60 ± 5%). Animals were exposed to a 12 h light/dark cycle, fed a standard laboratory diet and water ad libitum.

2.2. Ethical considerations

Experimental procedures were performed according to the guide on the care and use of laboratory animals approved by the Ethical Committee of Faculty of Medicine, Tanta University, Egypt.

2.3. Induction of electrical injury

According to Wang et al. (2006), rats of the experimental groups were subjected to an electric current via an electrical energy transfer device consisted of a double copper cable with a pair of ends. One end peeled 1 cm in length and the other connected to an electrical energy source (conveying AC of 220 V, 50 Hz alternating current). The animals were fixed on a plate and one clamp connected to rat left hind limb while the other to right forelimb.

2.4. Experimental design

After one week of acclimatization, animals were divided into five experimental groups; control group: included 10 normal rats not exposed to electric current and were sacrificed at the same period of their corresponding experimental groups; groups 1–3 (non-fatal electrocution groups): included 30 rats (10 rats for each group) that were exposed to electric current wasn’t of lethal intensity for one minute then sacrificed immediately, after 2 h, and after 4 h respectively (Li et al., 2015), and group 4 (fatal electrocution group): included 10 rats that were sacrificed after being electrified up to death (153 ± 27 s).

Animals of the control group as well as groups 1–3 (non-fatal electrocution groups) were anesthetized by i.p. injection of pentobarbital (50 mg/kg). Then specimens of the cerebellum of all groups obtained immediately from the lateral lobes and processed for histological and caspase-3 immunohistochemical study according to Suvarna et al. (2013) and Ramos-Vara (2005). In brief, specimens were divided into two parts, one was fixed in 10% formalin buffered saline; dehydrated, cleared, embedded in paraffin, and finally sections of 5 μm obtained for histological and immunohistochemical study using light microscope. The other specimen was fixed in 3% glutaraldehyde for electron microscopic examination.

2.5. Histological examination

2.5.1. Haematoxylin and Eosin (H&E) stain

In short, sections were deparaffinized and were hydrated following by staining with H&E. Finally, dehydration, clearing and mounting in Canada balsam.

2.5.2. Gleece and Marsland’s silver staining

In brief, wax was removed from the slides, then dipping into 0.5–1% celloidin followed by washing in 70% alcohol. Next, slides were placed in 20% silver nitrates, washed in 10% formalin, flooded with ammoniacal silver solution then with 10% formalin. After that, slides were fixed in 5% sodium thiosulfate, washed, dehydrated and mounted in DPX.

2.5.3. Electron microscopic examination

According to Kuo (2007), sections were cut into very small pieces, fixed in 3% glutaraldehyde for 24 h, incubated in 1% osmium tetroxide at 4 °C, dehydrated, and embedded in epoxy resin. Next, sections were cut (50 nm) with an LKB ultramicrotome, stained
with 2% uranyl acetate and lead citrate, then were examined by JEOL transmission electron microscopy at the Electron Microscopy Unit, Faculty of Medicine, Tanta University, Egypt.

### 2.6. Immunohistochemical examination

Sections were deparaffinized, rehydrated and placed in 0.3% hydrogen peroxide/methanol for 20 min to block endogenous peroxidase activity. Then, sections were immersed in 10 ml of citrate buffer (pH 6) and microwaved. Afterwards, blocking of non-specific protein binding sites through treating sections with a serum-free protein blocking solution for 20 min at room temperature. Next, the sections were incubated at 4°C with a 1:150 dilution of anti-caspase-3 antibody (Abcam, UK) followed by incubation with the secondary antibody (Dako, North America, Inc., CA, USA). After that, 1–2 drops of diaminobenzidine was applied to the sections, counterstained with hematoxylin, dehydrated, cleared and were examined by light microscope.

### 2.7. Statistical analysis

By using the image analyzer computer system (Leica Q500 MCO analyzer, Central Lab. Faculty of Medicine, Tanta University, Egypt), 5 μm sections stained with H&E as well as caspase-3 were used to examine the relative number of Purkinje cells per 1000 μm length of Purkinje cell line in 10 non-overlapping fields from 10 different sections of each experimental group using power 200.

![Image](image_url)

**Fig 1.** (Effect of electrocution on rat cerebellar Purkinje cells using H&E stain): A) Control: flask-shaped Purkinje cells (→) with basophilic cytoplasm, vesicular, centrally located nuclei and prominent nucleoli arranged in one row between molecular (M) and granular layer (G), instead of "A) Control: flask-shaped Purkinje cells (→) arranged in one row between molecular (M) and granular layer (G) with basophilic cytoplasm, vesicular, centrally located nuclei and prominent nucleoli." A) Control: flask-shaped Purkinje cells (→) arranged in one row between molecular (M) and granular layer (G) with basophilic cytoplasm, vesicular, centrally located nuclei and prominent nucleoli. B) Group 1: Deformed shrunken Purkinje cells with deeply stained cytoplasm (→) in-between normally appeared ones (●). instead of "Deformed, shrunken Purkinje cells, deeply stained cytoplasm (→) in-between normally appeared ones (●). C) Group 2: irregular distorted and shrunken Purkinje cells with pericellular unstained haloes (→) instead of "Group 2: irregular, distorted, shrunken Purkinje cells with pericellular unstained haloes (→). D) Group 3: abnormal arrangement with irregular shrunken cells and pericellular unstained haloes (→), abnormal arrangement, irregular, shrunken cells with pericellular unstained haloes (→). E) Group 4: fallen off cells leaving empty spaces (●), disturbed linear organization, marked cell degeneration with deeply stained cytoplasm and irregular shrunken outlines of the perikarya. instead of "fallen off cells, leaving empty spaces (●), disturbed linear organization, marked degeneration, irregular shrunken outline of perikarya, deeply stained cytoplasm, fallen off cells, leaving empty spaces (●), disturbed linear organization, marked degeneration, irregular shrunken outline of perikarya, deeply stained cytoplasm (→) (H&E x1000).
Values were presented as mean ± SD. Statistical significance between the different experimental groups was estimated using the two-tailed Student’s t-test after evaluation using F-test. P value <0.05 considered to be significant.

3. Results

3.1. Histological results

3.1.1. H&E results

The control group showed Purkinje cells, arranged in one row as a middle layer between the molecular and granular layer of the cerebellar cortex. They were large flask-shaped cells with basophilic cytoplasm, large rounded, vesicular and centrally located nuclei with prominent nucleoli (Fig. 1A). Group 1 examination showed deformed, shrunken Purkinje cells with deeply stained cytoplasm in between normally appeared ones (Fig. 1B). In group 2; Purkinje cells appeared irregular, distorted and shrunken losing their characteristic flask shape with pericellular unstained halos (Fig. 1C). For group 3; the Purkinje cells showed disorganization through which some displaced upwards in the molecular layer while others displaced downwards in the granular layer. They markedly appeared, irregular, distorted, and shrunken with pericellular unstained halos (Fig. 1D). As regards group 4; there was fallen off cells leaving empty spaces together with disturbed linear organization and loss of its flask-shaped appearance. Markedly degenerated and irregular shrunken cell outlines noticed in association with deeply stained cytoplasm (Fig. 1E).

The number of Purkinje cells in H&E stained sections revealed significant decrease in the experimental groups 1–3, and markedly decreased in group 4 (fatal electrocution group) when compared to the control (Fig. 2).

3.1.2. Silver stain results

In the control group, regular homogenous cytoskeletal elements were seen in the cytoplasm of Purkinje cell perikarya (Fig. 3A). Conversely, gradually affected cells were seen in the other experimental groups; through which some Purkinje cells with mild accumulation of its cytoskeletal elements were noticed in group 1 (Fig. 3B) to be widespread cytoplasmic argyrophilia in group 2 (Fig. 3C). While in group 3 intense argyrophilia of Purkinje cell cytoplasm was seen (Fig. 3D). However, in group 4 markedly distorted cells with dis-arrangement was observed in association with massive accumulation of cytoskeletal elements in its perikarya (Fig. 3E).

3.1.3. Electron microscopic results

The control group showed large Purkinje cell with euchromatic nucleus, dispersed chromatin and prominent nucleolus. Its cytoplasm was granular rich in ribosomes, rough endoplasmic reticulum (RER), Golgi apparatus as well as electron dense mitochondria (Fig. 4A1 & A2). In group 1, decreased Purkinje cell size with surface irregularity was noticed when compared to control. This was associated with the presence of perinuclear space, slightly dilated RER, and Golgi apparatus in addition to secondary lysosomes (Fig. 4B1 & B2). Regarding group 2; irregular shrunken Purkinje cell was seen in association with nuclear irregularity, dilated RER and Golgi apparatus, swollen degenerated mitochondria besides secondary lysosomes (Fig. 4C1 & C2). Concerning group 3, markedly affected cell was seen in the form of shrinkage, irregular nuclear outlines in addition to disorganization of the organelles, dilated RER and Golgi apparatus, and swollen mitochondria with destructed cristae (Fig. 4D1 & D2). Group 4 showed severely contracted dark stained Purkinje cell with irregular outlines and spinoous processes on its surface when compared to the control. Moreover, the nuclei showed irregularities of its nuclear envelope along with nuclear indentation. These changes were accompanied with markedly dilated RER and Golgi apparatus, and markedly swollen degenerated mitochondria with destructed cristae (Fig. 4E1&E2).

3.2. Caspase-3 immunohistochemical results

Purkinje cells of the control group showed negative immune reaction for caspase-3 and only scattered cells exhibited faint light brown granules in their cytoplasm (Fig. 5A). Instead, increased number of Purkinje cells with positive caspase-3 reaction was seen in the other groups through which group 1 showed fewer Purkinje cells with caspase-3 positivity (Fig. 5B) to be many cells in group 2&3 (Fig. 5C & D). Whereas, in group 4, most of the Purkinje cells appeared with positive cytoplasmic reaction for caspase-3 (Fig. 5E).

In our best knowledge, little was known about the electrical injury induced apoptosis. So, apoptosis by using caspase-3 was studied as a mechanism of cerebellar damage due to electrical injury. By using caspase-3 immunohistochemical staining, there was significantly increased number of caspase-3 positive Purkinje cells in groups 1–3 (non-fatal electrocution groups) to be markedly increased in group 4 (fatal electrocution group) when compared to control (Fig. 6).

4. Discussion

Electrical injury is an overwhelming traumatic event associated with different body system damage causing increased morbidity and mortality rate (Koumbourlis, 2002; Chudasama et al., 2010). Electrocution is an uncommon accidental cause of death. In the United States, about 1000 deaths each year with a mortality rate of 3–5% ensues (Lucas, 2009). The pathological findings that will be detected in the present study may greatly help the forensic pathologist to determine the cause of death from fatal electrocution in case of absent external signs, as well as survival time in case of non-fatal electrical injury. Moreover, the degree of damage as well as disability from occupational electric injury could be detected and thus assists in the process of civil compensation (Jakubieniene et al., 2006).
Moreover, lived electrical rats histopathological neurological chemical changes were explained; in accordance with Schulze et al. (2016) who expressed Purkinje cell changes as a result of exposure to electrical injury.

Furthermore, the duration of exposure to electric current in group 4 (fatal electrocution group) was $153 \pm 27$ s and this difference between rats was attributed to the individual variations between the animals.

In the present research, rats were chosen because upon inducing electrical injury; their hearts recovers spontaneously from ventricular fibrillation. Consequently, when any deaths occur; an attention to the nervous system should be considered (Chilbert, 1990). Moreover, the alternating circuits produced different types of lesions in the central nervous system with the cerebellar Purkinje cells susceptible to such current. On the other hand, the neurological complications due to electrical injury are not commonly explained; histological and caspase-3 immunohistochemical study was used to give an evidence concerning the injury of cerebellar nerve cells (in particular; Purkinje cells) as a result of electrocution. In this light, the present research was to study the histopathological changes in the rat cerebellar Purkinje cells in non-fatal and fatal electrical injuries. Additionally, non-fatal electrical injuries were studied immediately, 2 and 4 h after the rats were electrocuted to show its immediate and delayed effects. Moreover, the duration of exposure of the electric current in non-fatal groups (1–3) was one minute, which wasn’t of lethal intensity, through which all exposed rats to one minute electric current were lived until the time of scarification even after their loss of consciousness either from cardiac or CNS complication.

In the present study, H&E stained sections showed progressive deformed shrunken Purkinje cells with deeply stained cytoplasm in group 1 together with pericellular unstained halos in groups 2 and 3 and fallen off cells with disturbed linear organization and loss of flask-shaped appearance in group 4. These results were in accordance with Pliskin et al. (1994). Additionally, the indirect cerebellar injury was assured by impairment of motor function besides coordination and cognitive deficits (Wesner and

Fig. 3. (Effect of electrocution on rat cerebellar Purkinje cells using silver stain): A) Control: regular homogenous cytoskeletal elements in the cytoplasm of Purkinje cell perikarya (→). B) Group 1: some Purkinje cells with mild accumulation of cytoskeletal elements (→). C) Group 2: Purkinje cell with wide spread cytoplasmic argyrophilia (→). D) Group 3: marked cytoplasmic argyrophilia (→). E) Group 4: arrangement of Purkinje cells in two rows with massive accumulation of cytoskeletal elements in its perikarya (→) (Silver stain x1000).
This could be attributed to the loss of Purkinje cells after the traumatic brain damage in rats (Park et al., 2006).

According to Andrews (2012) cognitive and psychological disorders has become manifest in some cases even when the electrical current apparently does not cross the brain and when structural brain damage is not found. This may be due to increased cortisol levels and/or severe psychological and physical stress from electrical trauma as well as overstimulation of the excitatory
neurotransmitters and receptors including glutamate. Consequently, the elevated cortisol and excited glutamate receptors may lead to damage to the memory through a disruption in long-term potentiation (Martin et al., 2003).

The current study showed significant reduction of Purkinje cell number in H&E stained sections of the experimental groups 1–3 to be marked reduction in group 4 (fatal electrocution group) when compared to control. These results were in agreement with Kurtulus et al. (2009) who revealed loss of neurons after the electrical injury. Also, the study made by Hausmann et al. (2007) on the cerebellum of individuals with a history of acute or prolonged hypoxia/ischemia before death revealed that about 60% of Purkinje cells were lacking of nuclear staining and become shrunken with its number decreased with advancing survival time after circulatory arrest. Additionally, the neuronal cell loss may occur indirectly from respiratory arrest secondary to paralysis of the respiratory center or muscles that results in hypoxia or from ischemia resulting from thrombosis (Pliskin et al., 1994; Koumbourlis, 2002).

Shock risen from alternating current lead to hemorrhage in the nervous system with a large number of nerve cells that were permanently damaged with unexpected recovery. The mechanism of these hemorrhages after injury with alternating circuits is not entirely clear. It has been suggested that it was due to the strong tetanic contraction of the back musculature at the time the circuit is closed or might due to marked venous congestion that pertains during the shock or due to the sharp rise in blood pressure immediately following the injury (Huan-Jui et al. 2010).

Likewise, the neuronal cytoskeleton was disrupted not only by the mechanical injury, but also by the hypoxia and ischemia (Fitzpatrick et al., 1998). Consequently, to measure the extent of Purkinje cytoskeletal protein degeneration, silver stain was used. Our study showed an increase in the percentage of silver stained Purkinje cells by the increased duration after submission to electric current in groups 1–3 relative to the control group. Moreover, in group 4 (fatal electrocution group), many distorted Purkinje cells with massive accumulation of cytoskeletal elements in its perikarya was observed.

In the present work, apoptosis of Purkinje cells was confirmed by electron microscopic examination. There was apparently decreased Purkinje cell size, irregular cell outlines, and ultrastructural changes to the organelles besides nuclear abnormalities. This was directly proportional to the time interval between the exposure to electrical shock and decapitation together with the duration of exposure to electric current. These findings were inconsistent with Anders et al. (2001) who disclosed that there were histological and ultrastructural changes in Purkinje cells induced by low voltage electrocution in the form of hemorrhage, edema, necrosis, inflammation, abnormal mitochondria as well as degenerated nucleus.

This could be explained by the fact that during re-oxygenation the reperfused neuron exposed to potential toxins that are injurious or even lethal to mitochondria, including oxygen free radicals associated with oxidative stress along with important proinflammatory mediators (del Zoppo et al., 2000; Ritter et al., 2000; Polster and Fiskum, 2004).
Dong and Chen (2002) stated that electrocution acts by two mechanisms. The first mechanism was by thermal tissue damage through which high temperature lead to dissolution of cell membrane components (e.g. phospholipids) resulting in loss of the cell membrane function with consequent cell death. The second mechanism was the electroporation damage secondary to a strong electric field resulting in formation of pores in the phospholipids component of the cell membrane leading to cell damage. Chen et al. (1998) added that, the elongated permeable state of the cell membrane will permit leak of potassium and other cellular metabolites out of the cells, calcium into the cell, and leak of sodium into the cell losing the ionic driving force and osmotically driving water flow thus triggering apoptosis and cell death.

Furthermore, the membrane proteins’ electrical damage was reported by Tropea and Lee (1992) and Andrews (2012). They mentioned that, when cells exposed to an intensive electric current, generation of a supra-physiological potential occurs across the cell membrane which drives a huge transmembrane currents with the net result is electroconformational damage of the membrane proteins that may threaten cell wall integrity as well as cellular function. In addition, the transmembrane protein molecules contain polar amino acid residues that can change their orientation in any electric field leading to protein degradation that may be irreversible with cell death (Arnolodo and Purdue, 2009).

In the present study, apoptosis was studied as a mechanism of cerebellar damage due to electrical injury attributable to both direct action and hypoxia (Yuan, 2009). Apoptosis has been mediated by the intrinsic mitochondrial pathway and the extrinsic cell death receptor-mediated pathway (Friedlander, 2003). Nickolls and Budd (2000) and Snider et al. (1999) reported that mitochondria played a role in mediating either necrotic or apoptotic neuronal cell death during ischemia/reperfusion and this was confirmed by the marked ultrastructural mitochondrial changes seen in the present work.

In the present study, immunohistochemical examination by caspase-3 was done to evaluate Purkinje cell apoptosis, which demonstrated significantly increased number of caspase-3 immuno-stained positive Purkinje cells of different groups relative to control. Moreover, the percentage of apoptosis was increased with increasing survival time in groups 1–3 to be marked in fatal electrocution group (group 4). Similarly, caspase-3 staining was found to be correlated with the other imaging data as seen in the change of cell size, number, and cytoskeletal density through light
microscopic examination as well as organelle changes seen by electron microscopic examination.

Caspace-3 has been identified as a central player of apoptosis and may be a target for regulating cell death (Friedlander, 2003; Machnicka et al., 2012). This was through proteolysis of DNA repair proteins, cytoskeletal proteins, and inhibition of Caspase-activated deoxyribonuclease, ending with morphological cell changes and eventually apoptotic cell death (Clark et al., 2000).

The results of the present study revealed more marked damage of the cerebellar Purkinje cells in the fatal electrocution group. This could be explained on the basis that the severity of the electrical injury depends on the duration of the contact with the source of the current (Koumbourlis, 2002).

In conclusion, apoptosis was involved in the pathogenesis of immediate and long term effect of electrical injury on Purkinje cells, which will be an aid to the forensic pathologist to determine the cause of death and residual damage and disability after electric shock. So we recommend further studies for determining the relationship between delayed neurological dysfunction and cerebellar Purkinje cell loss due to electrical injury. Furthermore, there is a great necessity for postmortem human researches to detect the effects of fatal and non-fatal electrical injuries to human cerebellar Purkinje cells and if they can be relied upon to identify the cause of death. Additionally histopathological examination is very important to be included for a final diagnosis of electrocution and it is advisable to use its different techniques as routine examinations for the diagnosis of all electrocution cases. On the other hand, we recommended the use of other methods for evaluation of Purkinje cell apoptosis such as TUNEL, DNA fragmentation, as well as COMET assay.

References


