Death by electrocution: Histological technique for copper detection on the electric mark

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Abstract

The current observation of deaths by electrocution, both for domestic and work-related accidents as well as those in other contexts, has deepened the scope of investigation into electric marks, especially from the histological point of view. This is one of the few investigation tools that may lead to the diagnosis of death by electrocution in this distinct area, bearing in mind the diagnostic difficulties that this type of fatality presents.

Our attention has been placed on the phenomenon of metallization. In particular, we focused on using the Timm's method [1] to locate the copper deposits. The phenomenon of metallization, usually could be caused by the copper deposit, this happens due to the copper debris released onto the skin by the live conductor. To date, this technique has only been used in the pathological field. Nevertheless, we tried to assess its application in seven selected cases, after partially modifying the technique, comparing it with the most common staining detection techniques and analysing the specificity, sensitivity as well as the potential for its application in the routine.

INTRODUCTION

In Italy, deaths by electrocution are still numerically significant, despite the existence of specific safety standards.

According to data obtained from a study in 1967 [2], in our country, there were 7.6 deaths by electrocution per million inhabitants, while the same figure in Switzerland was 5.7, 5.3 in Germany, 5.2 in Australia, 5.0 in the US, 4.9 in Japan, 4.5 in both Spain and Belgium, 4.3 in Canada and 4.2 in France. In nine other countries in Europe and Oceania there was a decrease, with a range of values between 3.7 and 1.3 deaths.

As already known, death by action of the electrical energy is a "functional" death-type, because fatality may happen as a result of disorders of the heart rhythm (such as an ventricular fibrillation) or electrically induced contraction of the respiratory muscles: internal findings are non-specific and therefore may substantially overlap with those of sudden death [3].

The only specific sign of the passage of electricity is the electric burn mark: this skin lesion is caused by electro-thermal heating of the epidermis and dermis generated by the passage of the current: the so-called Joule effect. In such domestic electric shocks, where the voltage is not high, it may not be always recognizable [4]. Electric marks generally have an irregularly rounded shape, however is not uncommon to see other shapes. Sometimes, they can be hidden by either folds of skin, hairy formations, hand calluses, or by the same skin burns such as those caused by an arc or related to clothing fire [5].

Electric burn marks are very resistant to putrefactive phenomena, as argued by Fernando *et al.*[6] and so it is often recognizable even when the corpse is found after a long interval from the time of death, e.g. when there is the need to perform an exhumation. It is usually located close to the point of contact between the skin and the live conductor, in this area the current enters and runs through the body, coming out when the body is earthed using the shortest pathway [7]. Even if signs of electrocution on the skin are not seen, the possibility of death due to electrocution cannot not be ruled out. Sometimes the mark is not formed either due to the current having low intensity, or to the skin resistance at the point of contact being very low. For example, burn marks may be absent when the skin is wet (e.g. sweat) or if the victim is completely immersed in liquid (e.g. in a bathtub): in the latter scenario electric burn marks could be found in a few cases, where the majority are linear and at the waterline [8].

Diagnosing the cause of death: electric marks.

The electric burn mark appears when the skin [3,4,9] has been in firm contact with an electrical conductor when the flow of current passes through the high resistance of the skin, heating up the tissues, and in addition the fluids underneath will produce steam.

However, when the contact is less firm and there is an air gap is formed between the conductor and the skin, the current jumps over this space, creating a lesion by spark (spark lesion)[10]**.** The spark-like burn is caused by a very high temperature and therefore causes carbonization of the keratin layer.

In the first type of electric marks, the ones caused by firm contact, by using the common staining method (haematoxylin-eosin)[11], we are able to observe a thickening of the *stratum corneum* whose lamellae are seen as dark compact wavy bundles; on this occasion (and also in the layers of the epidermis) irregular honeycomb-shaped cavities are formed, as well as corneas clavicular ("clave corneas"). The *stratum lucidum* is often disrupted and it is also frequently affected by the formation of clave corneas. The *stratum granulosum*, however, shows protoplasmic oedema, cell lysis and the disappearance of nuclei, which migrate into the epidermal layer below (Fig. 1). The cells of the *stratum spinosum* appear elongated and organized as tufts, sheaf or palisades-like, arranged varyingly on the skin surface.

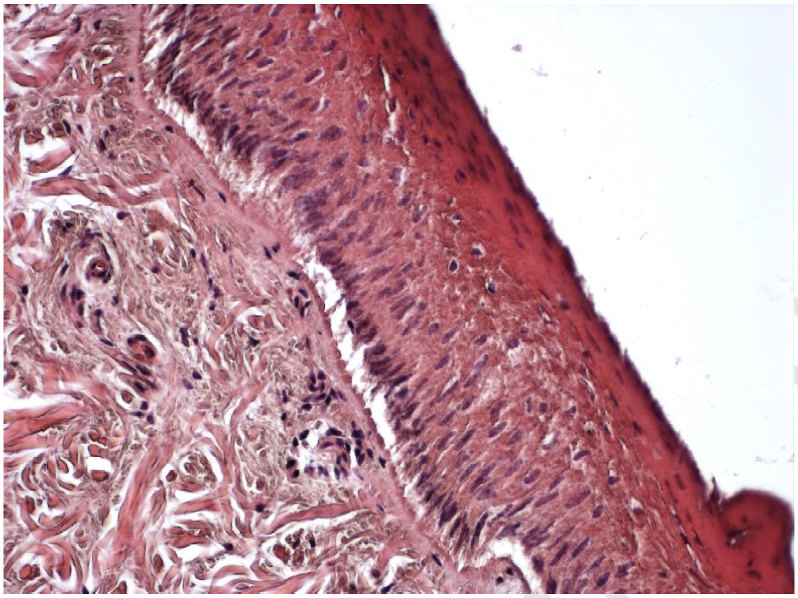

Figure 1 section of handheld skin: the presence of palisade-like nuclei in the stratum spinosum of the epidermis, lysis in the stratum granulosum as well as incomplete detachment from the underlying dermis caused by electric burn mark (HE,10x)

In the spark-like burn a crater-like wound is shown, in which the epidermis is either strongly altered or missing entirely. It is possible to observe residues of deformed basal cells, partially linked to the basement membrane along the edges and at the bottom of this wound.

Another relevant phenomena is the metallization which can be observed at the site of contact with the electrical conductor. Unfortunately, it does not happen in all cases, but as a consequence of a physical process consisting of arcing phenomena that induces the melting of the metals of the conductors (e.g. iron, copper, chromium, zinc, aluminium, molybdenum, manganese) and which are detectable by various methods.

The metallization sometimes clears doubts in challenging cases, as could the deposits of metal ions (e.g. copper, iron, manganese etc.) in correspondence to the electrocution burn marks.

A high voltage, applied for a prolonged period, typically causes profound metallization.

Among the methods used for the detection of metallization, the energy- dispersive X-ray spectroscopy[12] appears very sensitive (it can even detect a few micrograms of the metal element), non-destructive, rapidly performed and allows multi-elemental analysis[13].

The UV-fluorescence technique[14] is instead used to detect traces of aluminium and the diphenyl-carbazide method for those of chromium. The histochemical methods[19] are simple to use and widespread: in particular the colours Perl's Prussian blue[15] for iron and Okamoto - Utamura Rubeanic acid technique for copper. Other staining detection methods are the X-ray microanalysis[16], electron microscopy[17] and radioisotope techniques with cobalt 60[18], with the

latter being able to identify the precise point where the conductor came into contact with the skin surface.
The metallization, generally, allows a reliable diagnosis of electrocution as well as identification of the conductor that the electricity discharge originated from.

CASES.

Our study is based on the observation of seven autopsy cases of individuals who died from electrocution.

Case 1

A 63-year-old man went to a friend´s basement in order to repair an electrical system, in this case a pump, which was used to remove water from the floor. The subject was found dead with an electric porcelain plug firmly held in the left hand. This was plugged in and the floor was very wet.
On the left hand the subject presented a partially excavated, yellowish-coloured area of parchment consistency.

Case 2

A 27-year-old male went into his workshop in the early hours of the afternoon to perform some maintenance on his truck; he was found dead the same afternoon.
The soil surrounding the corpse was soaked with water; next to the dead body a water kettle was found that was linked to an electric plug.
The external examination revealed 2 electrical marks: the first on the palmar surface of the 1st and 2nd finger of the right hand, the second on the lateral side of the right thigh.

Case 3

While a 50-year-old man was working on a power line, he touched some high-tension wires with a metal pole. The external post-mortem examination showed that the subject had electrical marks on the left hand, localized in correspondence to the lateral face of the second phalange of the third finger. On the left foot, at the level of the first metatarsal phalangeal joint there was the exit mark; even on the sole of the foot there were several blackish-coloured disepithelialised areas.

Case 4

A 41-year-old man, a professional electrician, was working on a connection of high voltage cables (380 V) when he was caught up by an electric discharge. The death was almost instantaneous.

An external examination revealed electrical marks on the left hand and half of the right-hand.

An exit mark was seen at the plantar side of the first toe of the left foot.

Case 5

A 27-year-old male was in the basement of his family house fixing a light bulb; he was then found dead, lying on the ground.

During the external examination, an electric mark was found on the palmar surface of the left hand, it ended at the second phalanx of the 3rd finger.

There were several areas of disepithelialization at the plantar surface of the right foot.

Case 6

The subject, a 44-year-old male, was working on a construction site, moving an electrical generator. When he was grounding the machine, the electric current hit him.

An electric mark was found during an external examination on the 1st and 4th finger of left hand, and on the palmar area of the right hand as well.

Case 7

During a domestic accident, a 40-year-old man was electrocuted while he was helping to tidy up some electrical wires at his friend's house.

An external examination revealed that the subject had an electrical mark on the 4th finger of the left hand.

The skin samples taken during the post-mortem examinations at sites of injury were fixed in 10% buffered formalin. After washing and dehydration these samples were then embedded in paraffin, and sections were obtained by sliding microtome. The tissue sections were stained with haematoxylin / eosin staining method, as routine.

From the blocks stored at the Forensic Medicine Department, sections no more than 4 μm thick were cut and used to perform histochemical staining methods. To detect the presence of iron, the Perls' Prussian blue staining method was used[15], with this method, the iron debris becomes blue-coloured, and the cell nuclei red.

In order to highlight the presence of copper we decided to use Rhodamine [15] in sodium acetate solution and formic aldehyde for about twenty hours, which was followed by a five minute treatment with Mayer's haemalum; with such preparation copper debris takes a rust-red colour, while the cell nuclei are coloured blue.

However, given the limited sensitivity of the method mentioned above, we decided to apply the Timm's method for copper stain detection (so far used for the diagnosis of Wilson's Disease [20]) in a suitably modified form: the sections were left for five minutes in a solution of 0, 5% ammonium sulphide, then treated with 15% trichloroacetic acid (to remove the sulphides of iron and zinc). At this point, the sections were treated with a solution obtained by mixing silver nitrate to 5% and hydroquinone, in order to transform the copper sulphide to silver sulphide: by virtue of the latter debris copper, if present, their colour will change to black.

RESULTS.

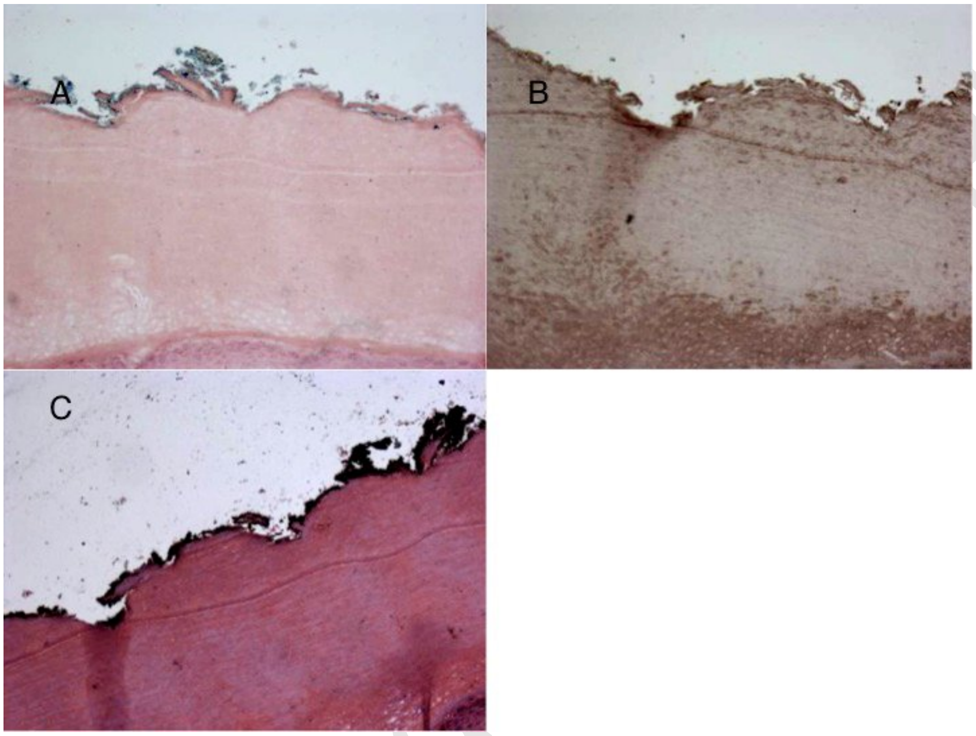

Figure 2 A) shows the slide coloured by Perls' Prussian Blue which detects the Iron debris; B) Rhodamine technique which poorly detects the copper deposit; C) TIMM's method which detects in a very clear way the Copper deposit on the skin.

This work aims to affirm the importance of the use of histopathological tools in forensic investigations, which allow the identification of the "signs of the current passage," hence enabling a confident diagnosis of electrocution to be made. The techniques used were Perls' Prussian Blue to detect iron, and both Rhodamine and the Timm's method to detect copper (Fig.2). In the 7 cases that we analysed, the Perls' Prussian Blue technique, which detects accumulations of iron, enabled us to identify the presence of the respective metal deposits in 4 cases out of 7. The Rhodamine method, however, used for copper detection, gave us (almost) positivity only in one case. Finally the Timm's method allowed identification of the deposition of copper very clearly in 5 out of 7 cases (Tab.1).

We have to bear in mind that electrical conductors are not made from a single metal: nowadays in the industrial production of electrical components, various materials are used, e.g. copper, iron and aluminium, especially to avoid the high costs in using a "pure" material, so it is often useful to perform more than one staining technique.

	CASE 1	CASE 2	CASE 3	CASE 4	CASE 5	CASE 6	CASE 7
Perls' Prussian Blue	-	+	-	+	+	-	+
Rhodamine	-	+/-	-	-	-	-	-
TIMM's	-	+	-	+	+	+	+

Table 1 shows the results of the different techniques used to detect the depositions of ions on the electrocution marks on the skin with concerns on our cases.

DISCUSSIONS.

Besides the search for iron ions and confirmation of Perls' Prussian Blue staining technique as a cheaper, faster, but still reliable means to achieve a definite diagnosis in cases of death by electrocution, this study evaluated the effectiveness of a new method, namely the Timm's method [1], for the search of copper ions. This method has never been applied to human cases of electrocution and it was found to be an unequivocally more sensitive and specific staining method than Rhodamine [16], the most commonly used method that does, however, present a lot of difficulties. We must take into account that this (rhodamine) staining technique can be performed by any laboratory which can carry out histological analysis. Nevertheless it has relatively long production times (about 48 hours) and could be significantly affected by the quality of the preparation; in the final analysis, therefore, this method has been involved in a higher percentage of doubtful cases, which might be due to be artefacts arising from the difficulties of implementing the technique itself.

Cases 2, 4, 5, 6 and 7, in fact, in which the object involved in the electrocution phenomena was an electric wire made by copper, were completely negative to the rhodamine staining technique, results were then denied by the subsequent repeated staining technique with Timm's. In cases 1 and 3, however, where the conductor was not a wire composed of copper, this technique was found to be negative.

CONCLUSIONS.

Our study has revealed some interesting results about the use of the Timm's method regarding copper detection in electrical burn marks on the skin. The first data are very important, emerging from both the crime scene view and the external examination in the autopsy suite: on the basis of these reports it is possible to determine when the presence of copper can be assumed, to be found with our method. Our study also shows a high reliability of the methodology and a higher sensitivity than the use of the histological staining technique with rhodamine. Moving on to consider the fundamental aspects that suggest routine use of Timm's method, we must also emphasize the economic/management issue. With its high sensitivity, this method has obvious limitations as to the quantification of copper in the fabric (of little use to the forensic pathologist in cases of death from electrocution). Such limitations may be rectified using other methods, such as probe X-ray and mass spectrophotometry, however they are expensive and hardly accessible in the territory.

We suggest the routine use of the Timm's method, which is sensitive, economical, easily repeatable in every laboratory, and of fundamental importance for the reconstruction of the causal relationship between "not pathognomonic" injury and death by electrocution.

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