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DNA-typing of cellular material on current conductors

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Abstract The examination of deaths due to electricity may require a comparison between current marks on the body and the electrodes suspected to have caused them. Normally the identification of the responsible electrode is carried out by analysing metal traces on the current marks. We however examined the conductor for traces of biological material after experimentally produced current marks. The surfaces of the conductors were investigated using a low-power macroscope and burnt tissue could always be recognised. Subsequently, all electrodes were carefully swabbed, extracted with chelex and typed for short tandem repeat polymorphisms using PCR. This procedure was successful in all cases. Therefore, DNA analysis can be a powerful tool to supplement conventional scene reconstruction in cases of deaths due to electricity.

Key words Current · DNA typing · Current marks · Electrode · Conductor

Introduction

The diagnosis of death due to electricity often necessitates the exclusion of other causes of death (Bonte et al. 1986; Fechner et al. 1990; Rothschild et al. 1997). Technical examinations as well as the investigation of current marks are required if present (Xiaohu et al. 1995). However, current marks are only found in 30–60% of cases (Klein 1958). A comparison of suspected conductors with the current marks is based on a comparison of metal traces (Böhm 1970; Kernbach-Wighton and Kijewski 1997; Logemann et al. 1996; Marcinkowski and Pankowski 1980) and occasionally on the comparison of the shape of the conductor and the morphology of the current marks (Karlsmark et al. 1984; Schneider 1978).

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When more than one conductor with the same metal composition is found at the scene, no clear assignment can be made based solely on metal comparison. In this article we present a new approach based on the individualisation of biological traces that can be found on the current conductors after skin contact.

Case report

An 8-year-old boy was found standing in an empty bathtub and shortly afterwards he collapsed. He was transported to the clinic, a ventricular fibrillation was diagnosed and resuscitation was performed. However the boy died 2 days later due to hypoxic damage to the brain. Current marks were found on the fingers.

The technical examination revealed that no current source was present in the bathtub but 2 m away there was a partially opened electric water heater under a hand basin. Soldering tin and brass were detected on the current marks which were also present at several possible conducting sites on the equipment (Ortmann et al. 1997).

Despite the identical metal composition there were doubts about the boiler as a source of current because of the spatial distance between the equipment and the position of the child. Therefore the question arose whether biological material would have been transferred from the skin to the conductor. In the literature no method has been published for DNA typing of biological material from current conductors. We therefore investigated whether sufficient material for DNA analysis could be found.

Materials and methods

Experimental current marks were produced on abdominal skin from two corpses removed during autopsies, using electrodes (n = 11) of various metals and shapes. The current source was household current (220 volts, 50 Hz) with a duration of about 5 s and a pressure of contact of approx. 80–100 g. The distance between the electrodes was 1 cm and identical electrodes were used on both poles. Control experiments were carried out under identical test conditions without current.

The electrodes were examined for biological traces under a low-power microscope (magnification × 6.3–32, M420, Wild, Switzerland).

DNA analysis

Sterile swabs were moistened with 100 µl sterile distilled water and the surface of the conductors was carefully swabbed. Only one of the two conductors from each experiment were investigated. The cotton was removed from the swab and incubated in a solution of 450 µL 5% (w/v) Chelex and 50 µL Proteinase K (2 mg/ml) for 36 h at 56° C (Wiegand et al. 1993). The tubes were vortexed, placed in a boiling water bath for 8 min, vortexed and centrifuged for 10 min at 14000 × g. The supernatant was placed on a Centricon 100 micro concentrator, concentrated according to the manufacturer's instructions and dialysed once with 500 µL of sterile water, The residual liquid was collected and diluted to 100 µL with water, quantified according to Waye et al. (1989) and 20 µL of this solution was used as a template for PCR. Amplification was carried out with the AmpFISTR Blue PCR amplification kit (PE Applied Biosystems) according to the manufacturer's instructions. After amplification, samples were separated by capillary electrophoresis (ABI 310 PE Applied Biosystems) according to the manufacturer's instructions using Performance optimised polymer 4.

Results

In the experimental studies a visible current mark was always produced and material on the conductor could sometimes be seen with the naked eye. Using the low-power microscope brownish-black stained spots together with some granular depositions were found on all conductors which were firmly stuck to the surface. On irregular surfaces (e.g. screws) larger particles were found (Figs. 1, 2).

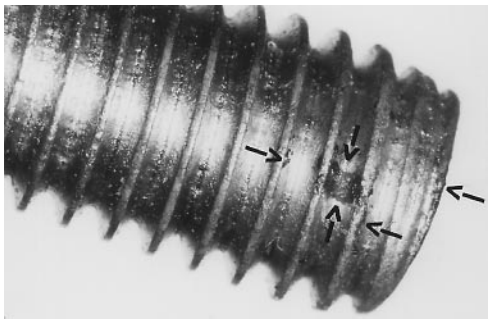

Fig. 1 Burnt organic tissue on an iron screw (magnification 16 ×) with scattered spots on the outside of the thread (↑) and a larger adhesion between the thread

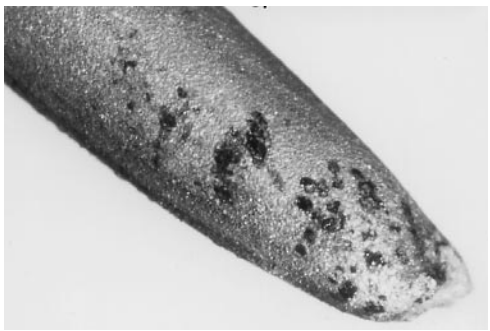

Fig. 2 Multiple spots on the tip of a tin wire. Viewing under a low-power microscope reveals the rough surface of the wire (magnification 22 ×)

Table 1 Amount of DNA experimentally transferred to current conductors

Experimental current conductor	DNA amount
Soldering tin	800 ng
Brass screw 2 cm	400 ng
Varnished iron screw 3 cm	200 ng
Twisted zinc wire	200 ng
Iron nail 1 cm	18.8 ng
Iron nail 2 cm	18.8 ng
Iron screw 3 cm	18.8 ng
Brass screw 5 cm	9.4 ng
Copper paper clip	4.7 ng
Copper pipe	3.1 ng
Aluminium rivet	3.1 ng

Table 2 Genotypes of the stain from the brass clip and the reference blood sample

	D3S1358	VWA	FGA
Ref. Blood	15/18	14/16	23/24
Brass clip	15/18	14/16	23/24

DNA analysis

DNA extracts obtained experimentally from the swabbed surface of current conductors contained between 3 ng and 800 ng of DNA (Table 1). All samples were typed successfully using the AmpFISTR Blue PCR amplification kit and the allele pattern observed was always identical to that of the reference blood samples. Control cases (without current) were always negative.

In the case presented here dirt and dust were found on the possible conduction sites from the water heater but organic tissue was not recognisable. A shutter in the proximity of the presumed conductors showed soot adhesions. All areas of soldering tin and brass were swabbed and typed for DNA and as a substrate control the plastic surroundings of the respective conductors were also swabbed. Only a smear on a live brass clip exhibited a DNA profile identical to that of the victim (Table 2) and no other DNA profile was found. The frequency of the observed profile in the German population is 0.0042% (Rolf et al. 1997; AmpFISTR Blue PCR amplification kit user manual).

Discussion

In many cases of death due to electricity current marks can be found on the skin but the current source cannot be clearly identified. The reasons for this might be that the source was removed or repaired (Sellier 1975) and it is also possible that the current source was not in close proximity (Jellinek 1925, 1932; Ortmann et al. 1997).

A variety of technical approaches exists for the comparison of current marks and electrode metal (Böhm 1970, Kernbach-Wighton and Kijewski 1997; Logemann

et al. 1996). But the detection of the same metal can only exclude conduction sites with different metal composition. If several conductors with similar or identical metal composition are present at a scene, the one responsible for the current mark cannot be clearly identified. Little attention has been paid to contamination of the electrode with burnt skin or other biological material but experience has shown that biological material is transmitted especially by high voltage accidents. The adhesion of biological trace material by low voltage such as 220 V conductors are barely recognisable and it seems that the material is carbonised by the heat. However, investigation under low-power microscope revealed that these stains were not completely carbonised. PCR-based typing of short tandem repeat polymorphisms is presently the most sensitive method available for complete individualisation (Karger et al. 1996, 1997). Additionally, as the DNA fragments are relatively short (100–350 bp) this method is relatively insensitive to degradation of the DNA in the stain material.

Our results clearly demonstrate that sufficient DNA for complete individualisation is still present on these conductors. The electrode distance used in our experiments was similar to that present in the case reported.

The amount of DNA extracted correlates approximately with the amount of material transferred which also seems to be dependent on the surface structure of the conductors as less material was found on smooth surfaces. Because the adhesions were dry and stuck firmly to the metal surface, the DNA typing can be successful even after a longer time interval.

In the case study we could not morphologically detect organic tissue on conduction sites from the heater 1 year after the accident, nevertheless the DNA amplification was successful. The observed genotype matched that of the boy allowing identification of the conductor responsible for the current marks and consequently a reconstruction of the accident.

In conclusion the detection and typing of transferred biological material on current conductors are of forensic significance. PCR-based DNA typing of material on current conductors can be regarded as a new approach which can supplement conventional scene reconstruction in cases of current accidents.

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