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DNA analysis in a case of serial murderers

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Abstract. At the beginning of 2003, in Casablanca (Morocco), three persons (two men and one woman) were found dead in different places and in different slots: they were horribly mutilated. The man suspected for the murders was found hanged. We performed DNA analysis for the identification of each victim by comparison with the alleged relatives to test the paternity of the woman's son, to verify the siblings relation, and to discover to whom some hairs belonged that were found on the scene of the 3rd homicide. © 2003 Elsevier B.V. All rights reserved.

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1. Introduction

At the beginning of 2003, in Casablanca (Morocco), three persons were found dead in different places and in different slots: a man (T11) from whom only the legs were found, a woman (G1) and a young man (S1) whose body was horribly mutilated. All bodies were found stored inside identical big cardboard boxes: inside one of them, four hairs were found. After some weeks of investigations, the man suspected (C1) for the murders was found hanged by the police: it was supposed that he killed himself. Some samples were sent to our laboratory and we performed DNA analysis useful to: (1) identify the victims by comparison with their relatives; (2) verify if T11 and the suspect C1 were cousins; (3) verify who was the father of the G1 son (H1) since the victim was supposed to be the girlfriend of the suspect; (4) verify the presence of semen on the vaginal swab taken from the victim G1; (5) compare hairs found in the crime scene with the ones of the young victim S1 and of the criminal C1.

2. Materials and methods

2.1 . DNA extraction

DNA analysis has been performed from the root of four hairs, found inside the cardboard box containing S1 body, and from tissues (skin or muscle) belonging to the

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victims using "DNA IQ-Tissue and Hair Extraction kit" (Promega). Hair roots were incubated at 56° in an Incubation Buffer/Proteinase K solution containing DTT for 1 h, while about 1 mg of minced cadaveric tissues was incubated overnight in the same buffer. Then all samples were treated by different solutions (lysis, washing and eluition) and a special paramagnetic resin according to the manufacturer's protocol [1]. DNA was extracted from oral swabs belonging to the alleged parents of the victims by "Instant Gene Matrix"(Biorad) treatment. Samples were incubated at 56 °C for 30 min and then boiled for 8 min [2]. All extracts were then quantified by the ALU Quant-Human DNA Quantitation System (Promega) that uses a specific human DNA probe and a luciferase reaction to produce light proportional to the amount of human DNA in the samples analysed [3].

2.2. DNA amplification

DNA amplification was carried out in a laboratory different from the one dedicated to the extraction, so that amplified products never entered the extraction laboratory. STRs amplification was carried out by Gene Amp 9700 and 2400 thermal cyclers (Applied Biosystems), using the *AmpFLSTR Identifiler kit* (Applied Biosystem) that coamplifies the repeat regions of 15 STRs repeat loci plus Amelogenin. According to the kit protocol, positive and negative controls were enclosed during the amplification step [4]. To verify if the suspect and one of the male victims were cousins, we analysed six STRs localized on the chromosome Y: DYS388, DYS390, DYS391, DYS392, DYS393 and DYS319. Amplification has been performed using the GeneAmp PCR System 2400 (Applied Biosystems) according to Kayser et al. [5].

2.3. Electrophoretic analysis

Amplified products were analysed by capillary electrophoresis onto two ABI PRISM 310 Genetic Analyzer (Applied Biosystems) employing ABI softwares (DATA Collection, GeneScan Analysis, Genotyper Fragment Analysis).

For fragment length determination of the PCR products obtained by Amp*FlSTR Identifiler* kit, the orange-fluorescent internal lane DNA standard LIZ 500 was used for calibration, while for Y-STRs detection, we used the red-fluorescent internal size standard ROX 350.

3. Results and discussion

In homicide investigations, if there is a tissue sample from an unidentified deceased individual and investigators have an idea of who may be the sample's source, if the decedent's parents or children are available, it will be possible to compare the related survivors' DNA profiles with that of the deceased to conclusively determine whom the remains came from. There are two possible testing outcomes. An exclusion is a simple statement that the alleged father is excluded from the group of men who could be the father.

In the case of an inclusion, a statistical analysis is performed based on how common or rare are the obligate genes. These are the genes that the alleged father could have contributed to the child and that must have come from the child's biological father.

The inclusionary results are expressed as Probability of Paternity while the Exclusion power shows the probability, given the mother and child results, that a non-father would be

excluded from paternity by the set of analysed markers. In this casework, we performed the calculation using the Moroccan frequencies reported by Huckenbeck et al. [6].

1. DNA analysis of biological material belonging to the victim G1 and of her alleged relatives showed a compatibility among DNA profiles. The probability of paternity of the alleged father versus the victim G1 was *0.99999999* (*99.999999%*). The exclusion power of the examined systems was 0.99999999 (99.999999%). Vaginal swab analysis did not show any trace of seminal fluid (negative results for Florence, acid phosphatase and PSA tests) but only the presence of epithelial cells which showed the same DNA profile than the victim.

2. DNA analysis of biological material belonging to the victim S1 and of his alleged relatives showed a compatibility among DNA profiles.
The probability of paternity of the alleged father versus the victim S1 was *0.99971220* (*99.971220%*).
The exclusion power of the examined systems was 0.99982780 (99.982780%).

3. The comparison between DNA profile of H1 and the ones of S1 and of C1 showed that: (a) there was a compatibility, for the analysed loci, between DNA profile of H1 and the one of G1; (b) there were six incompatibilities, for the analysed loci, between DNA profile of H1 and the one of C1; (c) there was a compatibility, for the analysed loci, between DNA profile of H1 and the one of S1; (d) theprobability of S1 paternity of S1 (the alleged father) versus H1 was *0.99957940* (*99.957940%*). The exclusion power of the examined systems was 0.99986390 (99.986390%).

4. DNA analysis of biological material belonging to the victim T11 and of his alleged relatives showed a compatibility among DNA profiles.
The probability of paternity of the alleged father versus the victim T11 was **0.99971740** (**99.971740%**). The exclusion power of the examined systems was 0.99983220 (99.983220%).

5. The analysis of Y-STRs markers (that are inherited along the male offspring) showed that C1 and T11 had, for the analysed loci, an haplotype condivision, so presumably they belong to the same male line. This was compatible with the occurrence that they could be son of two brothers, that, in their turn, has inherited the same Y-STRs from the same father (the grandfather of the cousins).

6. DNA analysis of hairs showed that they were belonging to a man different from S1 and from C1.

DNA profile frequency was estimated on data from the population to which the individuals typed were belonged, since a different population study would probably lead to a different frequency estimate.
In particular, using the Moroccan frequencies reported by Huckenbeck et al. [6], the calculated value of profile frequency was 1:1,000,000,000.

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