



Identification of a carbonized body found inside a car

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Abstract

A man disappeared in a city of Southern Italy; later, a car was found burned and inside it was a burnt body. To identify the body, DNA typing was performed on a bone, a tooth and from partially carbonized brain tissue. For comparison, we obtained DNA specimen from the saliva samples of the parents of the missing man. The cumulative probability of paternity (P50%) was calculated as 99.999990%.

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1. Introduction

A man disappeared in the city of Cosenza (Calabria, South of Italy). A few days later, a burnt car was found in a brushwood containing a burnt body. The identification of the body was performed using DNA typing.

2. Materials and methods

2.1. DNA extraction

DNA extraction from the unidentified body was performed by means of a bone (after pulverization) [1], and a tooth [2] using phenol–chloroform procedure and from a partial carbonized brain tissue by Biorad Instant Gene Matrix (Chelex) treatment. This is a special chelating resin that absorbs cell lysis products, which interfere with the PCR

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amplification process [3]. A piece of brain tissue was exposed to mechanical lysis and then resuspended in $1 \times$ PBS and autoclaved water. A small quantity of the cell suspension was added to Instant Gene Matrix and then incubated at 56° for 8–12 h. The sample was boiled for 8 min in a 6% Chelex solution and then amplified directly [4]. For comparison, we extracted by Instant Gene Matrix, DNA from saliva samples belonging to the parents and the brother of the missing man. All extracts were quantified using the Quantiblot-Human DNA Quantitation kit [5].

2.2. DNA amplification

Amplification was carried out in a separate laboratory so that the amplified products would never enter the extraction laboratory. STR amplification was carried out by GeneAmp 9700 thermal cycler (Perkin Elmer), using the PowerPlex16 kit (Promega), which coamplifies the repeat regions of the following 16 STRs repeat loci: D3S1358, D5S8118, D8S1179, vWA, TH01, D13S317, D21S11, D7S820, FGA, TPOX, D16S539, D18S51, CSF1PO Penta E Penta D. A segment of the X–Y homologous gene Amelogenin is also amplified. Based on the kit protocol, positive and negative controls were included in the amplifications [6].

2.3. Electrophoretic analysis

Amplified products were analyzed by capillary electrophoresis on an ABI PRISM 310 Genetic Analyzer (Applied Biosystems) employing ABI software (DATA Collection, GeneScan Analysis, Genotyper Fragment Analysis) [7]. For fragment length determination of the products, internal lane DNA standard ILS 600 (Promega) was used.

Table 1

	Body (brain–bone–tooth)	Father	Mother	Brother
D3S1358	16/17	16/17	16/16	16/17
D5S8118	11/12	9/12	11/13	12/13
D8S1179	14/15	13/14	15/15	14/15
vWA	14/15	15/16	14/16	15/16
TH01	9/9	6/9	9/9.3	9/9
D13S317	8/12	12/12	8/8	8/12
D21S11	28/29	29/32.2	28/32.2	29/33.2
D7S820	12/12	10/12	9/12	9/12
FGA	21/23	22/23	21/21	21/23
TPOX	8/8	8/11	8/8	8/11
D16S539	9/10	9/10	10/14	9/14
D18S51	13/14	13/14	14/14	13/14
CSF1PO	10/11	8/11	10/13	8/10
Penta E	11/17	7/11	15/17	7/17
Penta D	10/12	12/12	10/10	10/12

3. Results

DNA typing results obtained from the carbonized body, from the parents and the brother of the missing man are shown in [Table 1](#).

4. Conclusion

The Amelogenin test confirmed that the body found inside the car was a male while DNA typing confirmed that the body was actually that of the missing man. The paternity test performed on the DNA samples from the missing man's family showed that the DNA profile from the unidentified body was compatible with the DNA profiles from the parents. The cumulative probability of paternity (P50%) calculated using special software and using allele frequencies of Cosenza was 0.99999990 (99.999990%). P(Ex) was 0.99999940 (99.999940%) [8].

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