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DNA typing in missing persons in Ecuador (South America)

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Abstract. The identification of missing persons and unidentified body remains by DNA analysis is currently a routine method in Forensic Genetics laboratories. DNA extraction from bones and teeth involves a degree of complexity that varies depending on the methodology used and the quality of the samples. Each laboratory should establish its own protocols on the basis of its local needs. The success of the DNA study of bones and teeth for human identification is not absolute and a 100% success rate is hardly ever achieved; the greater the number of individuals to be identified and the larger the number of samples, the greater the number of problems. © 2006 Published by Elsevier B.V.

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1. Introduction

This treatise succinctly explains the importance of DNA and its application in the identification of missing persons, by means of the analysis of the bone remains and teeth. Body identification [1,2]. In many situations in which, due to the circumstances, the possibility of identification is very complex, such as when using old biological samples, remains or human fragments, etc., DNA testing will be the only means of investigation.

2. Materials and methods

We report the cases analysed by our laboratory for the identification of missing persons. *DNA extraction*: For the bone remains, phenol–chloroform–isoamyl alcohol and proteinase K were used. For the victims' family members, we used the Wizard Genomic DNA

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Purification Kit System© (Promega) method. DNA was quantified by means of UV absorbance. *PCR*: Amplification was carried out in a Genius© thermocycler, in accordance with the manufacturer's recommendations. *Typing*: An ABI310 sequencer was used. The fragment sizing and the allelic designation of the different loci were carried out by comparing them using the kit PowerPlex 16 allelic ladders and they were subsequently interpreted using the Gene Scan Analysis Software® programme. We followed the recommendations of the *DNA Commission of the International Society of Forensic Genetics* for STR analysis. *Procedure control*: For internal quality control purposes, target controls were processed in the extraction and amplification of each reaction. A positive human DNA control of cell line 9947 A, previously typified for all the systems, was used. For external quality control, the laboratory carries out an annual proficiency test with the *International Society for Forensic Genetics* (GEP-ISFG). The examination was carried out in duplicate, with a DNA control of known genetic information. 15 autosomic microsatellites and the marker Amelogenin were studied. *Data analysis*: Previously published classical Bayesian methods were used to calculate the likelihood ratio in which two mutually exclusive hypotheses are taken into account [3,4]. A spreadsheet was used for likelihood ratio analysis [5]. The system we use integrates 13 microsatellites that form part of the CODIS system, used in genetics databases the FBI (USA) works with.

3. Results and discussion

3.1. Case 1: individual identification of missing persons

In Table 1, eight cases of DNA extraction from bones and teeth are sampled in order to identify missing persons. Only 75% of the identifications were DNA positive; in the

Table 1
Cases of missing persons analysed

Internal code	Remains analysed	Family member contrasted	Offence	Origin	Result
For 01-99	Long bone: tibia, ribs Flat bone: cranium	Presumptive parents	Kidnapping plus homicide	Burial	Positive (inclusion)
For 02-99	Long bone: right femur foetal remains	Presumptive father	Homicide plus paternity identification	Burial	Positive
For 10-02	Long bone: left humerus	Presumptive parents	Missing (kidnapping)	Putrid remains	Negative (exclusion)
For 06-03	Short bone: charred lower maxilla fragment Teeth: two lower molars	Presumptive parents	Identification of charred body following homicide	Charred	Positive
For 19-03	Long bone: right femur	Presumptive daughter	Kidnapping plus subsequent homicide	Burial	Positive
For 19-03	Long bone: left femur	Presumptive daughter	Kidnapping plus subsequent homicide	Burial	Positive
For 2-04a	Short bone: different bone fragments	No family members	Identification of missing person's body	Putrid remains	Negative
For 2-04b	Short bone: lower maxilla Teeth: six different dental pieces	Presumptive daughter	Identification of missing person's body	Putrid remains	Positive

Table 2
Correlation with presumptive family members

Family	Missing person	STR Amel	Contrasted family members	Samples studied (remains found)	Likelihood rate (LR)
1	Individual 1	XY	Presumptive mother	Right humerus head; flat bone, right ileum fragments; distal end epiphysis fragments of left ulna; another unidentified bone	39,794.122
2	Individual 2	XY	Presumptive father	Fragments of left talus and calcaneus	38.494
3	Individual 3	XY	Presumptive mother	Fragment of rib; patella; cranium; parietal bones; temporal bone fragment; femur fragment (exhumation); muscle fragment (exhumation); left calcaneus; left carpus fragment; left upper maxilla with six teeth	396.497
4	Individual 4	XY	Presumptive mother	Distal end right fibula epiphysis; distal end right femur epiphysis	1537.512
5	Individual 5	XY	Presumptive son	Distal end left femur epiphysis	1236.270

others, it was not possible to establish appropriate identification using DNA or any other method. According to the Public Prosecutor's Office and other NGOs, it is calculated that, in Ecuador, an average of 100 people disappear per month, 10% of whom are presumed to have died of violent causes. In all the cases analysed, the Public Prosecutor's Office pronounced judgment following the report issued and closed the case.

3.2. Case 2: catastrophes

On Wednesday, November 20th, 2002, there was an explosion inside one of the ammunition holds of the Brigada de Caballería Blindada Galápagos, in the city of Riobamba, Province of Chimborazo, leaving 7 individuals dead, over 100 wounded and 4 individuals missing. 20 bone-remains samples and 1 soft tissue sample (bloodstain on piece of fabric) from the explosion 'ground zero' were analysed.

The results found were in Table 2.

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