

CASE REPORT

Personal identification from skeletal remain by D1S80, HLA DQA1, TH01 and polymarker analysis

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Abstract: The completely reduced skeleton was found in a mountain stream. We presumed that the skeleton was a woman from a morphology feature of the skull and the pelvis bone. The level of the suture of the skull indicated that the age was the first half of the 70-years old from the 60-years old. As a result of the police investigation, the possibility of 66 years old woman who was missing for about six months was suspected. To inquire into her identification, a skull and left thighbone were cut off, and blood was collected from the suspect's daughters. We examined blood (ABO) and DNA types (D1S80, HLA DQA1 TH01 and polymarker system) for the skeleton and the suspect's families. Blood and DNA types analysis of two daughters revealed that their patients have 19 alleles in 9 blood and DNA types. Twelve alleles were admitted in the skeletal remain among presumed 19 alleles. There was no blood and DNA type to deny the mother and daughter relation all of nine types. The skeletal remain was not contradicted from the above-mentioned result though thought daughters' mother. *J. Med. Invest.* 49 : 83-86, 2002

Keywords : forensic casework, personal identification, skeletal, DNA analysis

INTRODUCTION

DNA analysis demonstrates large powers in forensic caseworks, especially the personal identification (1-3). The completely reduced skeleton was found. From the morphological examination, the possibility of a missing woman of 66 years old had been suspected. In this paper, we report a personal identification by DNA analysis from this skeletal remains.

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CASE REPORT

The completely reduced skeleton was found in a mountain stream. The bones were a skull, a pelvis bone, thighbones (right and left), and a finger bone. These bones were morphologically thought to be the one of the same individual. We presumed that the skeleton was a woman from a morphology feature of the skull and the pelvis bone. The level of the suture of the skull indicated that the age was the first half of the 70-years old from the 60-years old. It was impossible to presume her height, because the thighbone had been damaged. The postmortem duration was suspected to be about one year from half a year.

As a result of the police investigation, the possibility of 66 years old woman who was missing for about six months was suspected.

MATERIAL AND METHODS

To inquire into her identification, a skull and left thighbone were cut off, and blood was collected from the suspect's daughters (relative no. 1 and 2). The suspect's pedigree is shown in Fig. 1.

We examined blood and DNA types for the skeleton and the suspect's families.

Blood type examination

Absorption-elution method (4) and enzyme-linked immunosorbent assay (ELISA) method (5) were carried out for ABO blood typing from the skeletal samples. ABO blood type was detected from each blood by slide method (6).

DNA type examination

DNA was extracted from each material by phenol-chloroform method. DNA typing of D1S80 (MCT118), HLA DQA1, TH01 and polymarker (PM) system were carried out according to the manual of the National Research Institute of Police Science, Japan.

1) D1S80 type

D1S80 typing was carried out according to the previous report (7, 8) using D1S80 Primer Mix (LifeCodes, USA) and 2 ng of extracted DNA as template for amplification.

2) HLA DQA1 type and PM system

HLA DQA1 type and PM system were carried out according to the manufacture's recommendation using AmpliType PM+DQA1 PCR Amplification and Typing kit (Perkin-Elmer, USA) (9) and 4 ng of extracted DNA as template for amplification.

3) TH01 type

TH01 typing was carried out according to the manufacture's instruction using GenePrint STR System-TH01 (Promega, USA) and 2 ng of extracted DNA as template for amplification.

RESULTS

In blood groups ABO blood typing carried out. DNA types of D1S80 (Fig. 2), HLA DQA1 (Fig. 3), TH01 (Fig. 4) and PM system (Fig. 5) were analyzed. Table 1 shows the results of blood and DNA typing.

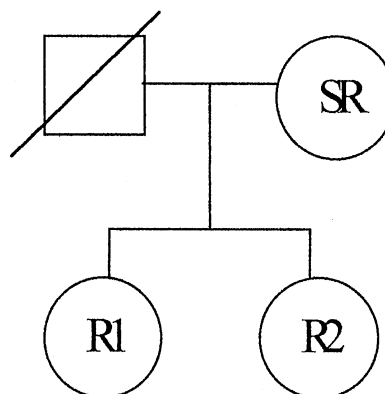


Fig. 1 Pedigree of the suspect's family
SR ; skeletal remain, R 1 ; relative no. 1, R 2 ; relative no. 2

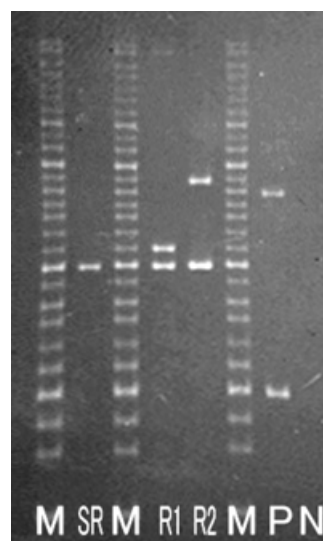


Fig. 2 Electrophoretic pattern of D1S80
SR ; skeletal remain (24-24type), R 1 ; relative no. 1 (24-25), R 2 ; relative no. 2 (24-30), M ; D1S80 allelic ladder, P ; positive control (18-29), N ; negative control

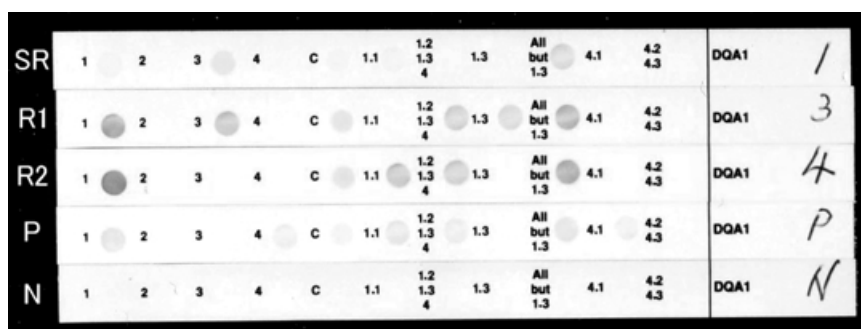


Fig. 3. Dot-blotting pattern of HLA DQA1
SR ; skeletal remain (1.1-3type), R1 ; relative no. 1 (1.3-3), R 2 ; relative no. 2 (1.1-1.2), P ; positive control (1.1-1.4), N ; negative control

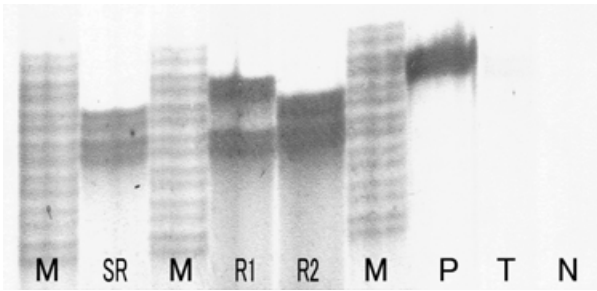


Fig. 4 Electrophoretic pattern of TH01
 SR; skeletal remain (8-9 type), R1; relative no. 1 (8-10⁻¹), R2; relative no. 2 (1.1-1.2), M; TH01 allelic ladder, P; positive control (10⁻¹-10⁻¹), T; typing control (10⁻¹-10⁻¹), N; negative control

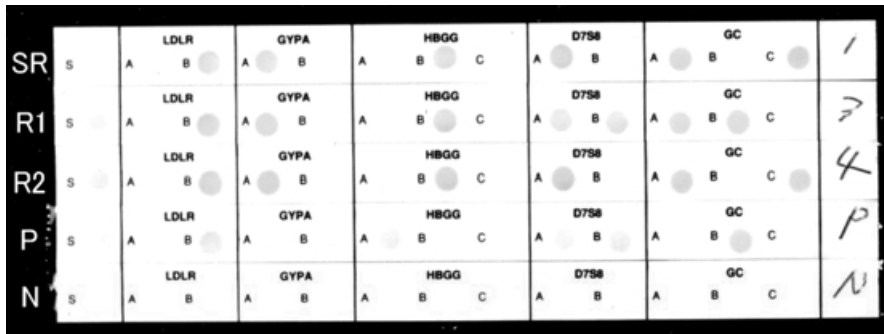


Fig. 5. Dot-blotting pattern of PM system
 SR; skeletal remain (LDLR; BB, GYPA; AA, HBGG; BB, D7S8; AA, GC; AC type), R1; relative no 1 (LDLR; BB, GYPA; AA, HBGG; BB, D7S8; AB, GC; AB), R2; relative no 2 (LDLR; BB, GYPA; AA, HBGG; BB, D7S8; AA, GC; AC), P; positive control (LDLR; BB, GYPA; AB, HBGG; AA, D7S8; AB, GC; BB), N; negative control

Table 1. Summary for blood and DNA typing

Blood and DNA types	samples			suspected allele
	skeletal remain	relative no 1	relative no 2	
<i>Blood type</i> ABO	A	O	A	A, O
<i>DNA type</i> MCT118	24-24	24-25	24-30	24, 25, 30
HLA DQA1	1.1-3	1.3-3	1.1-1.2	1.1, 1.2, 3
TH01	8-9	8-10 ⁻¹	8-9	8, 9, 10 ⁻¹
PM LDLR	BB	BB	BB	B
GYPA	AA	AA	AA	A
HBGG	BB	BB	BB	B
D7S8	AA	AB	AA	A, B
GC	AC	AB	AC	A, B, C

DISCUSSION

DNA analysis is the useful technique in forensic caseworks, especially the personal identification (1-3) and various body fluids (10-12).

D1S80 type is mini-satellite polymorphism, variable number of tandem repeat (VNTR), and its length is 16 base pair repeat. Since D1S80 population data are well characterized, D1S80 locus is used widely for forensic analysis (7, 8, 13). TH01 type is micro-satellite polymorphism, short tandem repeat (STR), and its length is 4 base pair repeat. TH01 locus is becoming more widely used for genetic characterization of forensic biological evidence (14, 15). From the reasons

above, routinely, we carried out DNA analysis with combination of D1S80, TH01, HLA DQA1 and PM system.

In this case, a skull and left thighbone were cut off for DNA sampling. DNA extracted from thighbone-marrow was highly fragmented because of putrefaction (16). So, we used for analysis DNA from a skull. The suspect's husband had already died. Materials from her brothers (sisters) were not obtained. Then, the individual was identified from the blood only of two daughters in the human race genetics. As a result, the personal identification was possible.

Daughters' ABO type were A and O type, and their mother's ABO allele was suspected to be A and/or

O. Skeletal remain was corresponding by A (AA or AO) type.

Daughters' D1S80 types were 24-25 and 24-30 type, respectively. Skeletal remain were 24-24 type, and 24 alleles were corresponding. When making it from daughters' HLA DQA1 types to mother, it was considered to have three alleles, such as 1.1, 1.2 and 3. Skeletal remain had two (1.1, 3) of three. It was suspected to have three alleles (8, 9, 10¹) when making it from daughters' TH01 types to mother. Skeletal remain had two (8 and 9) of three. The types of skeletal remain and daughters' types were completely corresponding to the LDLR, the GYPA, and the HBGG type among PM system. Daughters' D7S8 types were the AA, AB types, and their mother was presumed to have the allele of A and B. The D7S8 type of skeleton had A allele in the AA type. The GC type of the skeleton was AC type, and two were possessed among three (A, B and C) alleles having been presumed by the relatives.

Nine blood and DNA types were examined, therefore the mother and daughter relation was admitted in all types above.

In the personal identification by DNA analysis, the analysis of the relatives is important. In this case, the identity was suspected from the result of morphological examination of the skeleton. In addition, because two daughters' types were able to be analyzed, the identification can have been done in genetics.

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