CASE REPORT

Application of AmpFlSTR Profiler™ PCR Amplification kit for personal identification of a putrefied cadaver

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Abstract : A putrefied cadaver of q middle-aged woman was found drifting in the "Kii" water course. Autopsy findings indicated that the postmortem duration was about one week, and the cause of death was assumed to be drowning. In this case, a nail was collected as a sample for personal identification. After five months of police investigation, persons thought to be her family, husband and child, were found. A combination of D1S80 and the short tandem repeat (STR) typing system using an AmpFlSTR Profiler™ PCR Amplification kit was performed for identification. Nine STRs (D3S1358, vWA, FGA, TH01,TPOX, CSF1PO, D5S818, D13S317 and D7S820) and Amelogenin were analyzed by this kit. Those DNA typings successfully confirmed the family relation for personal identification of the cadaver. This analysis system may be useful for identification of a decomposed cadaver. J. Med. Invest. 51 : 243-246, August, 2004

Keywords **:** *forensic casework, personal identification, putrefied cadaver, DNA analysis, short tandem repeat*

INTRODUCTION

DNA typing of specimens obtained from a cadaver is extremely useful for identification of an unidentified body(1-9). Until 2003, four typing systems consisting of D1S80(10), TH01, and HLA DQA1and Polymarker (PM) system (LDLR, GYPA, HBGG, D7S8 and GC) had been used in the forensic laboratory of Japanese police (11, 12). From 2003, a Short Tandem Repeat (STR) typing system using an AmpFlSTR Profiler™ PCR Amplification kit (Applied Biosystems, Foster, USA) became applicable to routine casework (13, 14). In this report, we describe the personal identification of a putrefied cadaver using the STR typing system.

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CASE REPORT

A fisherman found a cadaver of a middle-aged woman drifting in the "Kii" water course. An inspection boat of the Japan Cost Guard retrieved the cadaver. The cadaver was only wearing a sock on the right foot. Chip wounds were observed in the left occipital region of head, the left dorsal area, and left hip. The left lower extremity was amputated at the left hip. These injuries were considered postmortem damage inflicted by the propellor of a ship while drifting. It was suspected that the woman had fallen from a height to the water-surface, because many rib fractures were observed. Autopsy findings indicated that the postmortem duration was about one week, and the cause of death was assumed to have been drowning. After five months of police investigation, a person regarded as a possible family member was located. DNA analysis was applied for the identification.

MATERIALS AND METHODS

Because blood could not be collected from the cadaver, a nail and skeletal sample were gathered for personal identification. From the potential family, husband and child, blood was collected with consent.

We examined the blood and DNA types of the cadaver and the potential family members.

ABO blood type examination

The absorption-elution method (15, 16) was carried out for ABO blood typing from the nail and skeletal samples. From each blood specimen, the ABO blood type was detected by the slide method (18).

DNA TYPE EXAMINATION

DNA was extracted from each material by the phenol-chloroform method. Eleven types of DNA polymorphisms, such as D1S80 (MCT118), D3S1358, vWA, FGA, TH01, TPOX, CSF1PO, D5S818, D13S317, D7S820, and Amelogenin, were investigated.

1) D1S80 type

D1S80 typing was carried out according to the previously described method (19) using D1S80 Primer Mix (LifeCodes, USA) and 2ng of extracted DNA as a template for amplification. D1S80 allele bands were separated by conventional electrophoresis.

2) Short tandem repeat (STR) and Amelogenin types

For typing of 9 STRs (D3S1358, vWA, FGA, TH 01, TPOX, CSF1PO, D5S818, D13S317 and D7S820) and Amelogenin, multiplex polymerase chain reaction (PCR) was performed using an AmpFISTR Profiler Plus™ PCR Amplification Kit (Applied Biosystems, Foster, USA) according to the user's manual. PCR product was separated using ABI PRISM 310 Genetic Analyzer, and then analyzed with GeneScan Software Ver.2.1. Internal size standard and allelic ladder marker (Applied Biosystems, Foster, USA) were also analyzed with the sample for typing.

RESULTS AND DISCUSSION

Table 1 shows ABO blood type, and summary of D1S80 (Fig.1), 9 STRs and Amelogenin typing systems (Fig.2).

Between 1996 and 2002, we successfully identified more than 20 cadavers using a combination of D1S

Table 1. Summary of ABO blood type and DNA types

	cadaver	potential husband	potential child
Blood type **ABO**	**O**	**B**	**B**
DNA typing			
D1S80	**30-30**	**24-31**	**30-31**
D3S1358	**15-17**	**15-15**	**15-17**
vWA	**16-16**	**18-15**	**16-18**
FGA	**21-24**	**22-24**	**24-24**
TH01	**6-9**	**9-9**	**9-9**
TPOX	**8-11**	**8-8**	**8-8**
CSF1PO	**12-12**	**10-13**	**12-13**
D5S818	**10-12**	**10-10**	**10-10**
D13S317	**8-10**	**9-11**	**9-10**
D7S820	**8-11**	**12-12**	**8-12**
Amelogenin	**XX**	**XY**	**XY**

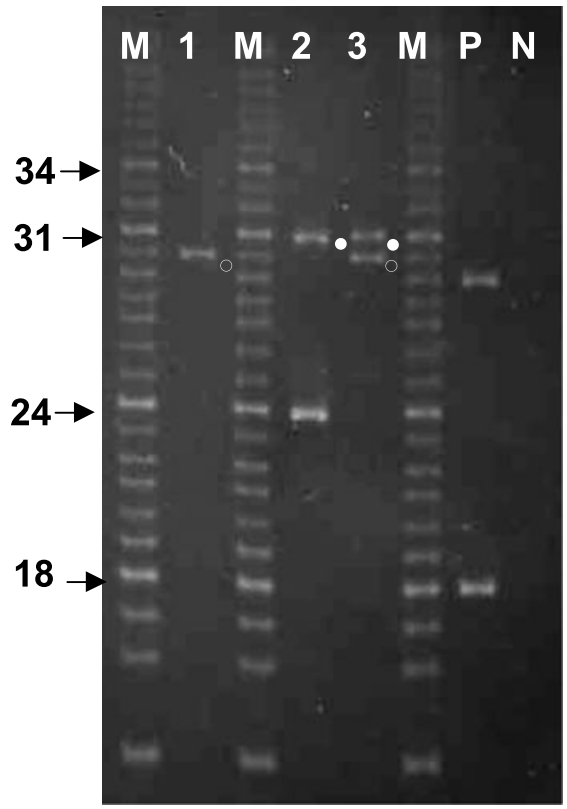

Fig.1 Allelic band pattern of D1S80
1 : cadaver, 2 : suspected husband, 3 : suspected child,
M : D1S80 Allelic Ladder, P : Positive Control (2.0 ng),
N : Negative Control
open circle : allele 30, closed circle : allele 31

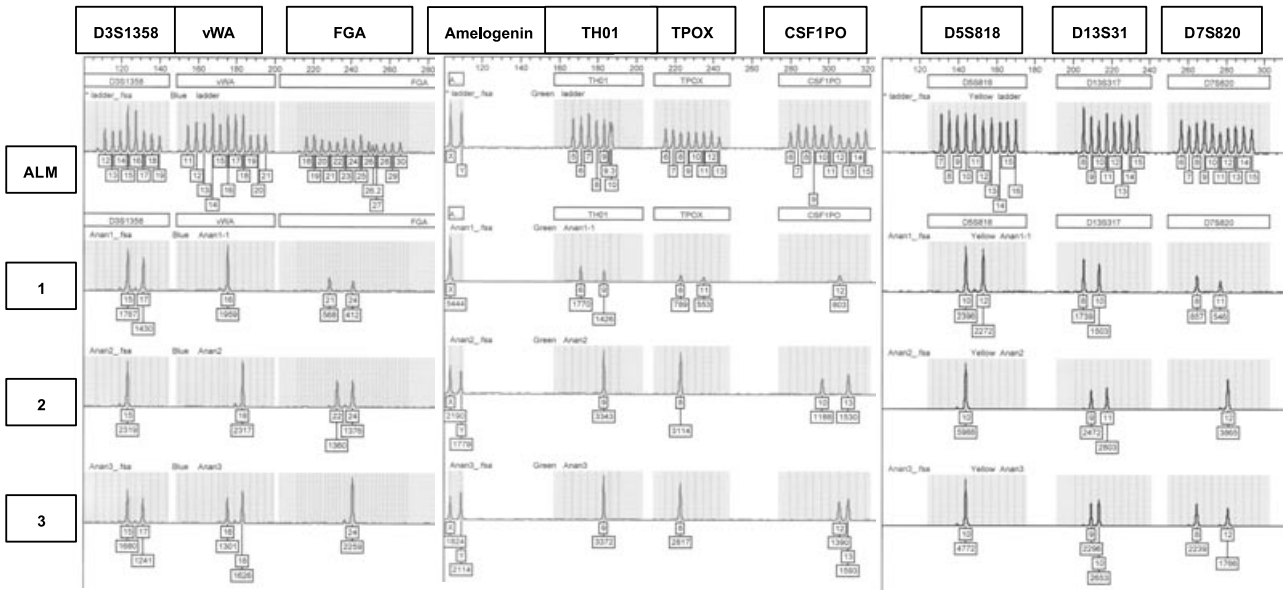

Fig.2 Electropherogram of examined STRs ALM : allelic ladder marker, 1 : cadaver, 2 : potential husband, 3 : potential child

80, TH01, HLA DQA1 and PM system (LDLR, GYPA, HBGG, D7S8 and GC) for routine casework. Verification of these personal identifications confirmed that DNA typing could be judged by DNA obtained from a nail within one month postmortem (20). From 2003, the combination of D1S80, 9 STRs and Amelogenin typing system was routinely applied for forensic casework. In this case, the nail was applied as the sample, and use of this new combination system was attempted for personal identification. DNA types of the potential husband, D1S80, D3S1358, vWA, FGA, TH01, TPOX, CSF1PO, D5S818, D13S317 and D7S820 were 24-31, 15-15, 18-18, 22-24, 9-9, 8-8,10-13, 10-10, 9-11 and 12-12, respectively. DNA types of the potential child, D1S80, D3S1358, vWA, FGA, TH01,TPOX, CSF1PO, D5S818, D13S317 and D7S820 were 30-31, 15-17, 16-18, 24-24, 9-9, 8-8, 12-13, 10-10, 9-10 and 8-12, respectively. Therefore, the cadaver's D1S80 allele was suspected to be allele 30. STR alleles of the cadaver were expected to be allele17(D3S1358), 16(vWA), 24(FGA), 9(TH01), 8(TPOX), 12(CSF1PO), 10(D5S818), 10(D13S317) and 8(D7S820). Each of the cadaver's alleles corresponded to the suspected alleles listed above. Those DNA types indicated a high probability of a relation between the cadaver and the potential family.

Previously, we reported relationships among detection rates achieved by four DNA typing systems (D1S80 typing, TH01 typing, HLA DQA1 typing, and PM system), the post-mortem interval, types of specimens (bone, nail, and blood), post-mortem changes, and the site at which the corpse was found (indoors, outdoor, or in the sea). The success of typing was highly influenced by the post-mortem interval. Using blood, almost all DNA types were detected, while the nail showed comparatively higher detection rates than bone. The detection rate decreased in the order of indoor, outdoor, sea, and soil as the site at which the corpse was found. We clarified that it was important to select the specimen corresponding to the postmortem interval and the site where the corpse was found (20).

In this case, the cadaver was found in sea-water, and the postmortem interval was assumed to have been about 1week. The nail was used as a sample for personal identification by the STR typing system using an AmpFlSTR Profiler™ PCR Amplification kit. As a result, personal identification was successful. This DNA analysis system may be applicable to the decomposed cadaver.

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