

K. Minakata · H. Nozawa · K. W. Suzuki · O. Suzuki

Paternity diagnosis by using umbilical cords preserved for periods ranging from 9 months to 44 years

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Introduction

Forensic DNA analysis has sometimes been made concerning aged human specimens [1, 2]. This report describes four cases of paternity diagnosis [3, 4] by using DNA extracted from preserved umbilical cords. The preservation of umbilical cords is rather rare in the world. In Egypt the umbilical cord is kept until the 1-month birth celebration, and then it is discarded. In some rural areas of The Philippines, the umbilical cord is preserved for a long time as an amulet and sometimes it is taken as a medicine. In Japan, however, most maternity clinics give the umbilical cord to the mother placed in a small box of paulownia wood as shown in Fig. 1. Many families keep it in a chest as a sacred material for a long time. Recently we examined four paternity cases where some persons were dead and only their umbilical cords were available for testing.

Materials and methods

DNA was isolated from dried material of five umbilical cord and seven blood samples using phenol-chloroform extraction. The purity and content of DNA were determined by the QuantiBlot Human DNA quantitation kit (ABI) as well as the UV absorptions at 230, 260 and 280nm. PCR amplification was carried out using up to 18 STRs (CSF1PO, F13A01, F13B, LPL, TH01, TPOX, vWA, D11S554, FGA, HPRTB, D18S849, D3S1744, D12S1090, D7S820, D9S304, D3S2459, D8S1132, DYS393) [5, 6, 7]. Allele frequency data used in this study can be obtained from the authors. The allele detection was performed by silver staining.

K. Minakata (✉) · H. Nozawa · K.W. Suzuki · O. Suzuki
Department of Legal Medicine,
Hamamatsu University School of Medicine,
1-20-1 Handayama, Hamamatsu 431-3192, Japan
e-mail: kminakat@hama-med.ac.jp,
Tel.: +81-53-4352233, Fax: +81-53-4352233



Fig. 1 The umbilical cord preserved for 44 years. The name of the midwife, body weight, body length, birth place, birth date, mother's name and father's name are written on the box from left to right

Results and discussion

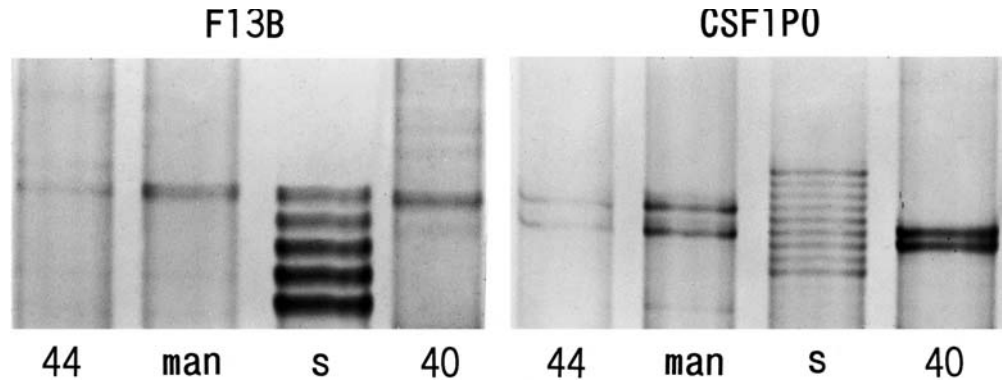
Hemolysis was not observed in all five umbilical cords immersed simply in water overnight. By using the cell lysis solution of DNA extraction kit (Wako, Japan), however, hemolysis was observed only in the 9-month-old sample. Treatment with proteinase K for 18–48h was needed for the other umbilical cords to solubilize DNA.

Case examples

Case 1. CSF1PO, F13B, TPOX, vWA, FGA, D18S849, D3S1744, D12S1090, D7S820 and D9S304 systems were examined for the putative father (30 years old), mother (30 years old) and a child (umbilical cord preserved for 9 months). Essen-Möller's paternity probability [8] was 99.9968% and the exclusion probability [9] was 99.989%.

Case 2. CSF1PO, F13A01, F13B, LPL, TH01, TPOX, vWA, D11S554, FGA, HPRTB, D18S849, D3S1744, D12S1090, D3S2459 and D8S1132 systems were examined for the putative father

Fig. 2 Two examples of DNA typing in case 4 (*man* indicates the putative father, *44* and *40* the umbilical cords preserved for 44 and 40 years, *S* alleles of the system)



(55 years old) and a child (umbilical cord preserved for 7 years). The paternity probability was 99.984% and the exclusion probability was 99.911%.

Case 3. CSF1PO, LPL, TPOX, vWA, D11S554, D18S849, D3S1744, D12S1090, D3S2459, D8S1132 and DYS393 systems were examined for the putative father (umbilical cord preserved for 38 years), mother (29 years old) and a child (4 years old). The paternity probability was 99.99975% and the exclusion probability was 99.9960%.

The relationship between the umbilical cord and the true mother (70 years old) who was still alive was also examined using the systems CSF1PO, LPL, TPOX, vWA, D11S554, D18S849, D3S1744, D12S1090, D3S2459 and D8S1132. The maternity probability was 99.9905% and the exclusion probability was 99.89%, indicating the legitimacy of the umbilical cord.

Case 4. Some results of electrophoresis are shown in Fig. 2. CSF1PO, F13B, TH01, TPOX, vWA, D11S554, HPRTB, D18S849, D3S1744, D12S1090, D7S820, D3S2459 and D8S1132 systems were examined for the putative father (70 years old) and two children (umbilical cords preserved for 44 years and 40 years). The paternity probability was 99.999910% and the exclusion probability 99.933% concerning the umbilical cord preserved for 44 years, and the corresponding values were 99.99947% and 99.958% for the umbilical cord preserved for 40 years.

These cases show unambiguous results to solve paternity disputes when persons involved were deceased. Furthermore, comparison of DNA genotypes between the umbilical cord and the true mother (or the true child) provided a basis for the legitimacy of the umbilical cord. The human DNA content per weight was comparable to that of dried blood experimentally prepared, even in the umbilical cord preserved for 44 years.

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