SNP Typing of Human Earwax Type

Human earwax type was determined by single nucleotide polymorphisms (SNPs) through analysis with the MCE-202 MultiNA. Compared with SNP typing using gel electrophoresis images, use of the MultiNA goes further by permitting quantitation.

Introduction

It has been found that there are some differences between individuals that can be explained using SNP analysis. For example, in the case of earwax, some visual determinations can be made from its shape, etc. using Mendelian heredity traits, and it can be classified as to whether it is of the sticky (wet) or rough (dry) type. There is also a report[1] that human earwax typing can be determined by SNPs. Here the SNP typing of earwax is introduced using the direct PCR method (Ampdirect) on blood and the ARMS-PCR Method[2] technology with the MCE-202 MultiNA for rapid analysis.

Results

Reaction products were analyzed using the MultiNA. Fig. 1 shows the data as a gel image. The base size in lanes A3 and B4 was indicated by bands at about 220bp and about 160bp. These showed the SNP base of the analytical target to be A and G heterologs, which is a wet type of earwax. A band was also observed at 160bp in lane A5 that was the SNP base G homology, a wet type. Another band was also observed at 220bp that was the SNP base A homology, a dry type.

Furthermore, numerical analytical results were obtained as are shown in Fig. 2. The numerical analysis results were exported from the MultiNA, and the peak analytical data (DNA fragment size and peak area), were applied in a simple algorithm to produce Fig. 3.

While almost all Europeans and Africans exhibit the wet type of earwax, in the regions of China and Korea, 100% are of the dry type. Yoshiura, et. al., have postulated that this dry type propagated from Northeast Asia. All of the people who submitted samples for the present study were Japanese, and 82% (14/17) were of the dry type. Thus, the results agreed with previous reports on the dry type being the major type among Japanese.

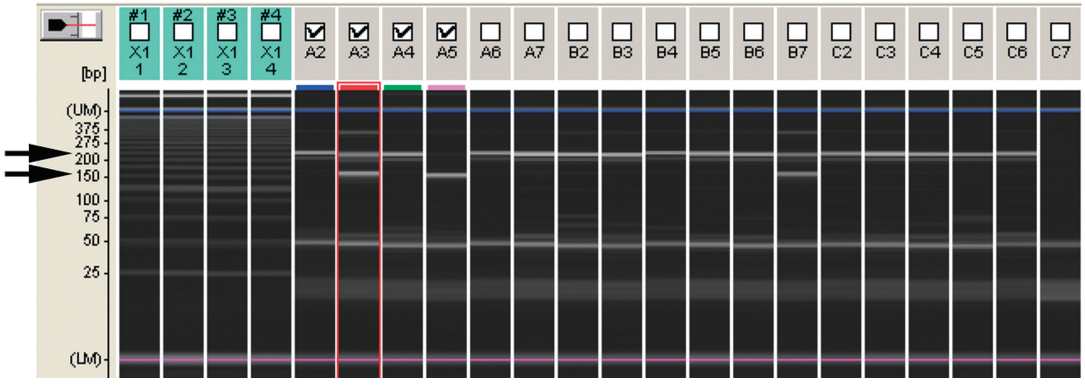

Fig. 1 MultiNA Representation of Gel Image of rs17822931 (ABCC11) PCR Products

[1] Yoshiura, K, *et. al*. A SNP in the ABCC11 gene is the determinant of human earwax type. *Nat Genet.* 38, 324-30 (2006).
[2] Ye S, *et. al.* An efficient procedure for genotyping single nucleotide polymorphisms *Nucleic Acids Res.* 29, E88-8 (2001)

Well name	A2	A3	A4	A5	A6	A7	B2	B3	B4	B5	B6	B7	C2	C3	C4	C5	C6	C7
Base size 156±3bp Peak Area (S1)	–	231.99	–	231.93	–	–	–	1.17	–	–	–	131.15	–	–	–	–	–	–
Base size 222±3bp Peak Area (S2)	174.97	203.15	272.41	2.4	212.39	434.13	316.6	359.36	161.69	291.74	273.25	116.8	189.53	374.89	238.81	158.71	211.07	–
Area ratio (S1/S2)	–	1.14	–	96.62	–	–	–	0.0033	–	–	–	1.12	–	–	–	–	–	–
Type	A homo	hetero	A homo	G homo	A homo	A homo	A homo	A homo	A homo	A homo	A homo	hetero	A homo	A homo	A homo	A homo	A homo	N.C.

Fig. 2 SNP Typing by Peak Analysis Data

Analytical Procedure

Analytical Device: MCE-202 "MultiNA"
Analytical Mode: DNA-500 on chip mixture
Primer array:

- FA: TTCTGCATTGCCAGTGTACTCA
- FB: GTCTGCCACTTACTGGCCC
- RA: CTTCACCGCCTTTGGGAAGAA
- RB: TGGCTACAGGGCCACTCCTTGG

Reagents:

- Ampdirect semi-refined for DNA
  (by Shimadzu Corporation) P/N 241-08800-97
- DNA-500 Reagent Kit for MultiNA
  (by Shimadzu Corporation) P/N 292-27910-91
- SYBR® Gold nucleic acid gel stain
  (by Invitrogen Corporation) S-11494
- 25bp DNA ladder
  (by Invitrogen Corporation)10597-011

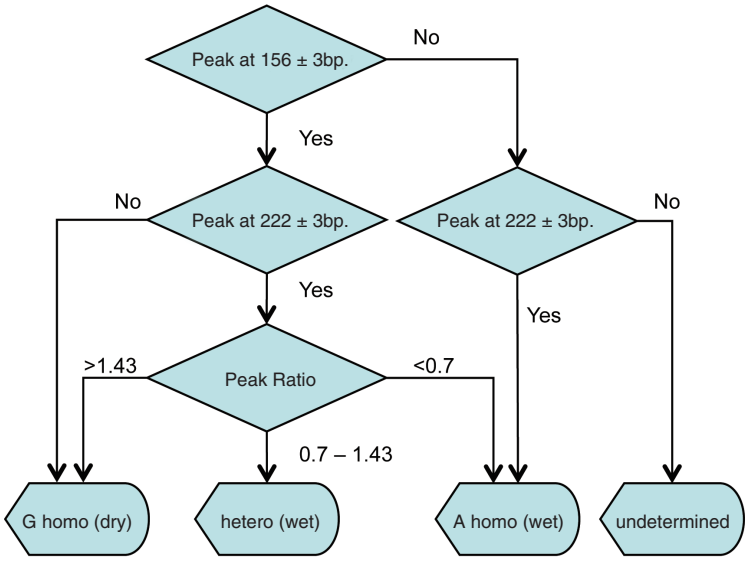

Fig. 3 SNP Typing Algorithm

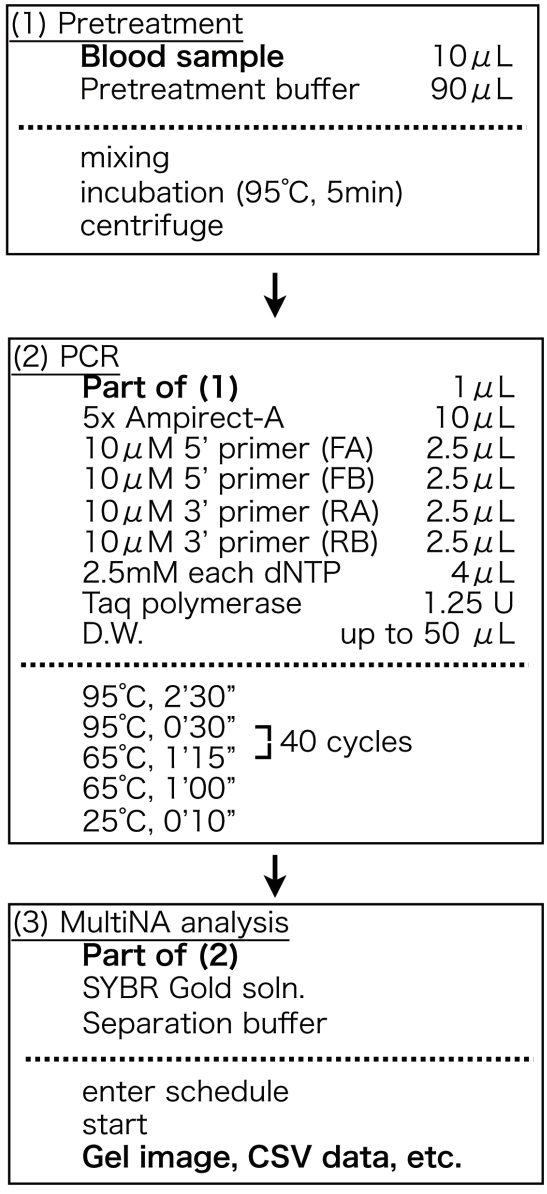

Fig. 4 Experimental Protocol for SNP Typing

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