A143 Unusual Variations in Mitochondrial DNA Sequences Between Three Maternally Related Siblings

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After attending this presentation, attendees will learn about the intricacies of developing guidelines for exclusion for mitochondrial DNA analysis.

This presentation will impact the forensic science community by demonstrating the importance of collecting multiple mitochondrial DNA references for the identification of skeletal remains.

One mission of the Armed Forces DNA Identification Laboratory (AFDIL) is to aid the Joint POW/MIA Accounting Command (JPAC) in the identification of missing U.S. service members from previous military conflicts. This is accomplished by extracting mitochondrial DNA (mtDNA) from samples of unknown origin collected in the field and comparing the haplotype to a database of mtDNA profiles obtained from family reference samples (FRS). The sequence comparison is used to either include or exclude potential missing service members in the remains identification. AFDIL requests at least two FRS per missing service member to resolve any potential differences between the missing individual and their maternal relatives (e.g., heteroplasmy or any single base mutations that may have occurred), although this may not always be possible.

Recently, two family reference submissions from persons listed as full siblings to an individual missing from the Korean War were found to differ by two polymorphisms in their respective mtDNA sequences. The sister had eight polymorphisms when compared to the rCRS and the brother had the same eight polymorphisms along with two additional polymorphisms, at nt146 and nt152. Examination of the electropherograms showed that both nucleotides exhibited complete homoplasmic changes and were free of any low level heteroplasmy or contamination. According to AFDIL's standard operating procedures and the current SWGDAM guidelines for mtDNA interpretation (SWGDAM, 2003),[1] these two individuals would be excluded from being maternally related.

A maternal half-sister to the siblings submitted a reference sample upon request for additional samples from maternal relatives. The half- sister's sequence was found to have the same eight polymorphisms common to the sister and brother as well as one additional polymorphism (nt146 C). AFDIL confirmed the relationship of the three siblings by testing autosomal STRs as well as an experimental assay analyzing chromosomal X-STRs.

In the 18-year history of AFDIL, 13,243 FRS have been received from 8,146 families and this is the first instance of siblings showing two distinct polymorphic differences in the mtDNA control region (when excluding the HV2 C-stretch and nt16093). Unlike autosomal STRs which allow for discrimination between individuals, mtDNA is typically used to discriminate among maternal familial lineages. The variability among the siblings in this case is explained given the positions at which the sequences are dissimilar. Positions 146 and 152 have relatively high mutation rates as both positions are within the top ten fastest mutating sites in the control region. It can be hypothesized that the mother of the three siblings was likely heteroplasmic at both nt146 and nt152. Unfortunately, the mother is not available for contribution of a sample to verify this hypothesis.

Given the infrequency of such an event, it is unnecessary to modify the current reporting guidelines; however, this can be seen as support for collecting more than one mtDNA family reference for the purposes of identifying unknown remains.

Reference:

[1] SWGDAM (Scientific Working Group of DNA Analysis Methods). Guidelines for Mitochondrial DNA (mtDNA) Nucleotide Sequence Interpretation. Forensic Science Communications 2003, April; 5(2).

Mitochondrial DNA, Sibling Variability, Identification

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