A133 Making a "Dentin" – The Use of Teeth for Human Identification: Modifications in Extraction Protocol Allow for Vast Improvements in Mitochondrial DNA Results

Carla D. Paintner, MS, MFS, Jennie G. McMahon, MS, and Suzanne M. Barritt, MS, Armed Forces DNA Identificatiom Lab, 1413 Research Boulevard, Building 101, Rockville, MD 20850; Brion C. Smith, DDS, 11663 Fairmont Place, Ijamsville, MD 21754; and Louis N. Finelli, MD, Armed Forces DNA Identification Lab, 1413 Research Boulevard, Rockville, MD 20850*

After attending this presentation, attendees will have step by step knowledge on the implementation of a new protocol for the extraction of mitochondrial DNA from highly degraded tooth samples, a greater understanding of handling difficult dental samples for DNA processing, and knowledge of the improvement in the success rate of samples processed at AFDIL.

This presentation will impact the forensic science community by explaining how the modifications to this protocol have radically changed how tooth samples from highly degraded cases are processed, and will illustrate how laboratories can implement this protocol. The AFDIL has documented an increase in reported tooth samples from a historic success rate of 78% to a rate of greater than 96% with the implementation of this simple protocol.

The primary mission of the mitochondrial DNA (mtDNA) section of the Armed Forces DNA Identification Laboratory (AFDIL) is to aid the Joint POW/MIA Accounting Command (JPAC) in the identification of United States service members lost in previous military conflicts. Mitochondrial DNA testing is frequently used to obtain genetic information in cases where nuclear DNA or direct reference material is limited. Obtaining DNA from skeletal remains exposed to harsh environmental conditions such as acidic soils, extreme heat or severe fragmentation is commonly a challenge. Often, a single tooth is the only element remaining after decades of degradation. Fortunately, tooth enamel offers a greater level of protection against environmental conditions than that provided to other osseous elements. A modified extraction protocol that yields reproducible results from a smaller sample with improved reliability while preserving the tooth for comparison to dental records is the goal of this study. The protocol utilizes increased cleaning stringency, along with a previously published demineralization extraction buffer (Loreille et al FSI:Genetics 2007) which dissolves a majority of the tooth powder. Dentin masses as low as 0.01 grams have been used to obtain fully reproducible HV1/HV2 mtDNA sequences, depending on the level of degradation. The implementation of this protocol has enabled the mtDNA Section at AFDIL to regularly generate multiple extracts from one tooth achieving our requirement for reproducibility. This protocol also improved the mtDNA testing success rate from a historical average of 78% to a current average of over 96%. Since less material is required for this protocol, it is not necessary to pulverize a complete tooth, allowing the odontologist to conduct subsequent comparisons to antemortem dental records that may become available at a later date. This protocol will benefit the forensic community in DNA identification of skeletal remains using bone and tooth material in a wide variety of circumstances including missing persons and mass graves.

mtDNA, Tooth, Extraction

 ** Presenting Author*