B51 Results of the SWGDAM Inter-Laboratory Exchange Study on Bone Extraction for Mitochondrial DNA Analysis

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The goal of this presentation is to present the results of the round- robin DNA typing study of skeletal remains conducted by the forensic mitochondrial DNA community including commercial, academic, and government forensic mitochondrial DNA laboratories in the United States and abroad.

This presentation will impact the forensic community by lending assurance to the forensic community that subtle differences in extraction, amplification, and sequencing protocols will still yield reproducible results.

With the implementation of the National Missing Persons DNA Database, the forensic DNA analysis of remains consisting of bone evidence continues to increase. Since the DNA found in forensic samples is frequently limiting and/or degraded, mitochondrial DNA (mtDNA) analysis is often the analysis method of choice. Obtaining mtDNA sequence from calcified tissue is particularly challenging. Laboratories employ several different approaches to obtain mtDNA of sufficient quantity and quality from skeletal remains. In addition, no proficiency test is currently commercially available with bones as the evidentiary material.

The Mitochondrial DNA Subcommittee of the Scientific Working Group on DNA Analysis Methods (SWGDAM) has conducted an inter- laboratory study comparing the extraction methodologies and sequencing results obtained from a single source of bone sample. For the purposes of this study, a tibia was obtained from an anthropological research facility. This tibia had been buried for a period of approximately three years prior to dry storage at room temperature at the facility for an unknown period of time. Prior to distribution, the tibia was assessed for suitability and verification of mtDNA sequence. The tibia was sectioned by the organizing laboratory and distributed to the twenty-one participating laboratories. Extraction, amplification, and sequencing of the bone sections were performed according to each laboratory's standard protocols. Results were submitted from nineteen of the participants with concordant results for mtDNA sequencing. For laboratories submitting results for autosomal and Y STRs, concordant results were also obtained.

In addition to submitting typing results, participating laboratories submitted their standard operating procedures which contained details of their extraction methodologies as well as amplification and sequencing strategies. These details are presented. Despite variation in the cleaning methods of these bone portions, as well as variations in extraction methods (including decalcification, if applicable), quantity of sample used, amplification parameters, post-amplification quantification, sequencing chemistries and instrumentation, all methods proved reliable and the results obtained were concordant. Comparison of these results highlights the robust nature of forensic typing methodologies.

Although the results obtained from the current study demonstrated the reliability of forensic testing, the next generation of this inter- laboratory bone exchange study will include a more environmentally challenged sample to more closely mimic the type of samples encountered in a forensic context.

This study also displays the willingness of the forensic community to advance the knowledge of the field through collaborative studies.

Mitochondrial DNA, Skeletal Remains, Inter-Laboratory Study

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