



B184 The Success of DNA Testing of Skeletonized Human Remains and the Comparison of Organic vs. Inorganic Extraction Protocols

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After attending this presentation, attendees will understand not only the success of skeletal elements for DNA testing as a whole, but will also increase their knowledge regarding different extraction techniques.

This presentation will impact the forensic science community by providing information on different extraction techniques for DNA testing of skeletonized human remains. This presentation will also encourage attendees to be flexible in protocols and procedures to allow for greater success.

Since 1992, the Armed Forces DNA Identification Laboratory (AFDIL) has processed more than 16,000 skeletal elements received from the Defense POW/MIA Accounting Agency (DPAA) for multiple DNA testing platforms. These skeletal elements have typically been exposed to a variety of environmental insults postmortem and range in time of death from 40 years to slightly more than 80 years. While the initial testing strategy focuses on Sanger sequencing of the mitochondrial DNA (mtDNA) Control Region (rCRS), in the past ten years, autosomal Short Tandem Repeat (STR) testing has become an increasingly prevalent testing option. Currently, more than one-third of the casework reported each month is some form of STR analysis. These include Low-Copy Y-chromosomal Short Tandem Repeat (Y-STR) Analysis (LCN-Y) and a variety of STR testing kits.

With the shift in focus to STR analysis, in addition to a desire to use less dangerous chemicals, AFDIL implemented an inorganic DNA extraction procedure in 2013. Validation studies indicated inorganic extractions provided a cleaner DNA extract, which is more suitable for STR analysis; however, there was some concern that the extract was too clean for the existing mtDNA protocols which were adapted specifically for extracts that included low-quality DNA as well as some inhibitory materials.

Over the subsequent 3.5 years, sample types have been tracked to determine the efficacy of inorganic extractions. While the general trend in STR analysis has seen a marked improvement, from ~20% success to a current 44% success, mtDNA success has slipped, from 92% to 85%. Initial evaluation of the samples indicates that skeletal materials exposed to inhibitory materials, such as formalin or fuel, were less successful overall when extracted using an inorganic protocol. In such instances, samples were re-extracted using a Phenol-Chloroform-Isoamyl Alcohol (PCIA) -based organic extraction and generated successful mtDNA results. An organic extraction protocol has been re-incorporated into day-to-day testing for severely compromised skeletal remains.

Frequently, the forensic community is eager to adopt a single strategy for processing of casework samples. A workflow with minimal decision points is simpler, especially when faced with a large number of samples or complex cases. This is seen to be one of the great appeals in the move toward automation; however, this evaluation of extraction protocols used for skeletonized remains indicates that a careful evaluation of the sample upon receipt and a selection of a workflow based on downstream needs may be valuable. It is also indicative of a need to maintain multiple protocols within a laboratory, rather than to suffer loss of success or capabilities by eliminating “outdated” protocols.

DNA, Extraction Techniques, Skeletonized Remains

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