## A130 Quantitative Ancient and Forensic DNA Techniques for Maximum DNA Recovery From Thermally Altered Bones and Teeth

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**Learning Overview:** The objective of this presentation is to convey the success of obtaining highly degraded DNA from thermally altered skeletal material. Here, the goal is to educate the attendees on the implications of applying ancient DNA methods to traditional forensic cases, and the possibility of reconstructing partial Combined DNA Index System (CODIS) Short Tandem Repeat (STR) profiles using mini-STR multiplexes, as well as reconstruct whole mitochondrial genomes using Next Generation Sequencing (NGS) technologies and bioinformatics pipelines.

**Impact on the Forensic Science Community:** This presentation will impact the forensic science community by offering the best DNA methods used to test the quality and quantity of DNA in highly burned skeletal remains.

The recovery and analysis of DNA from burned human remains represent some of the most challenging cases in medicolegal investigations. The rapid degeneration of DNA often impedes the ability of forensic examiners to obtain adequate DNA profiles for subsequent genotyping and identification. In this study, the goals were to assess the utility of two different extraction methods used by forensic and ancient DNA researchers to obtain degraded DNA, as well as to establish the effects of heat-induced color change and type/location of element sampled on DNA quality and quantity.

Over a period of two years, this study obtained and documented 62 samples from 26 fire death and cremation cases in collaboration with the Maricopa County Office of the Medical Examiner. Samples were chosen with the goal of obtaining different skeletal tissues showing different levels of burning (levels I–V). After processing the bone to powder, each sample was divided for extraction using either the ancient DNA protocol or the forensic protocol. The total nuclear DNA concentration of these extracts was qualified with an Applied Biosystems® Real-Time Polymerase Chain Reaction (RT-PCR) using the Quantifiler® Trio DNA Quantification Kit, and STR detection was performed using the PowerPlex® ESX 17 Fast Systems STR kit by Promega®. In addition, sample extracts were also converted into double-stranded DNA libraries and enriched for mitochondrial DNA and nuclear Single Nucleotide Polymorphisms (SNPs) for next-generation sequencing.

The results show that average DNA yields recovered from fire-related victims show a progressive decrease in DNA concentration across burn categories I–V. Both extraction protocols work equally well for burn categories I–III; however, highly incinerated remains (categories IV–V) produced better CODIS STR results using the ancient DNA protocol. Overall, the data confirm the difficulties in obtaining DNA from burnt skeletal material using conventional RT-PCR and STR genotyping technologies. Confounding limitations inherent in both ancient and forensic DNA research, such as small sample sizes, intra- and inter-sampling differences in material input, low DNA yields paired with high degrees of DNA degradation, continue to restrict the amount of genetic information gleaned from the most challenging forensic cases. All these factors were recognized in this study. However, integration and adoption of optimized ancient DNA protocols, such as the Dabney DNA extraction protocol, in forensic case analyses may improve the success rate of acquiring adequate STR profiles to identify fire victims in circumstances in which traditional applications have failed.

**Forensic DNA, Ancient DNA, NGS Analysis**



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