NEXT-GENERATION SEQUENCING OF MITOCHONDRIAL DNA EXTRACTED FROM HISTORIC HUMAN REMAINS

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DNA extracted from human skeletal remains is often of poor quality and/or low quantity.However, extracted DNA is often vital to determining the identity of an individual, particularly in forensic or missing persons cases. The goal of this research was to examine low quality DNA extracted from degraded, historic human remains recovered from a Maryland gravesite using mitochondrial next-generation sequencing (NGS) to learn about the family of the remains. The historic human remains were recovered in the 1970s from the Calverton site (ca. 1600s), locatedin Calvert County, Maryland.

Despite the degradation commonly observed in nuclear DNA extracted from historic or degraded human remains, the more resilient mitochondrial DNA can often be relied upon to provide relevant information and data that can assist in determining the identity of the remains. NGS has been found to accurately and efficiently examine mitochondrial DNA, even from old ordegraded remains, meaning that the results of this research should offer immediate benefits for missing persons identification efforts and cold case investigations involving degraded remains.

Because the maternally inherited mitochondrial DNA can be highly indicative of ancestry and kinship, it offers immense utility when attempting to identify an individual. As such, this research provides further insight into forensic genealogical techniques for human identification through maternal kinship analyses using mitochondrial DNA as well as ancestryestimation.

In this research, a bone sample obtained from one set of skeletal remains from the Calverton site was extracted prior to DNA quantitation and amplification. Initially, capillary electrophoresis (CE) was performed to produce an STR profile and examine the nuclear DNA extracted from the remains, which showed significant degradation. After the nuclear DNA was analyzed and separated using CE, the sample was submitted for NGS. Library preparation was performed using the ForenSeq mtDNA Control Region Kit, and the sample was sequenced usingthe MiSeq FGx. Preliminary results have shown that NGS can be utilized to successfully analyze degraded mitochondrial DNA extracted from historic human remains, increasing the yield of data available for analysis when compared to CE data alone. Mitochondrial DNA sequencing of the degraded remains produced high-quality results, making haplogroup determination possible. The haplogroup is indicative of the individual’s ancestry and is used in kinship analyses.