MITOCHONDRIAL DNA ANALYSIS OF HUMAN HAIR: DECONTAMINATION STUDY & APPLICATION TO FORENSIC CASEWORK

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The Armed Forces DNA Identification Laboratory's (AFDIL) previous protocol for the extraction of DNA from hair, used proteinase K digestion in the presence of DTT. This procedure occasionally produced an incomplete digestion or one that required incubation for over twenty-four hours, with repeated additions of proteinase K. Incomplete digestion can produce lower DNA yield leading to poor quality sequencing data. A new protocol similar to and adapted from one developed by the Federal Bureau of Investigations (FBI), includes the addition of four twenty minute Terg-a-zyme washed in a sonicating water bath, and the use of microtissue grinders to facilitate the digestion of the hair shaft. Additionally, Centricon 30s (C30's) are used instead of C100's in post extraction purification in order to minimize loss of what may be highly fragmented DNA. The objective of this presentation is to: 1) demonstrate that this extraction procedure is effective in removing contaminating DNA from hair, and 2) demonstrate the new protocol as it applies to forensic casework involving baby hairs.

Three unrelated individuals of known mtDNA sequence each donated head, body and pubic hair; all hair shafts were cut to approximately 2cm in length (roots were not included). A sample of head, body, and pubic hair from each individual was contaminated with either blood, semen, saliva or handling by an individual whose mtDNA sequence was known. A total of 36 samples were cleaned by Terg-a-zyme sonication washes, ground in microtissue grinders, extracted with a standard organic protocol, and analysed by mtDNA amplification and sequencing. Results support the finding of a previous pilot experiment that four Terg-a-zyme washes are highly effective at removing contamination even from hairs that are grossly contaminated by body fluids rich in DNA.

The second part of this presentation demonstrates application of this protocol to forensic casework involving baby hairs. AFDIL routinely performs mtDNA analysis on ancient skeletal remains from the Vietnam War, Korean War, and World War II. Often a maternal relative is not available for comparison to the remains and an alternative reference source may be needed. A common reference source in these cases is baby hair from the service member's first haircut that may still be in a family scrapbook. AFDIL's previous experience indicated that it can be difficult to obtain mtDNA from these samples. However, data generated from two different baby hair samples, using the above extraction protocol, produced high quality mtDNA sequences. These results support the use of this new protocol for the extraction of baby hairs.