EFFECT OF ANIONIC DETERGENT SELECTION ON STR PROFILE DEVELOPMENT FROM BONE-DERIVED DNA EXTRACTS

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DNA analysis is often essential to make positive associations in cases of unidentified persons, missing persons, and mass fatality incidents. In cases such as these, human skeletal remains are frequently the only source of genetic material available, but bone-derived DNA characteristically provides lower quantities of DNA and lower quality short tandem repeat (STR) profiles than that of other sample types.

Conventional DNA extraction methods were developed based on the biochemical composition of soft tissues or body fluids. DNA from these unmineralized sample types is more readily extracted and contains fewer inhibitors than DNA found in bone; furthermore, skeletal remains encountered in casework may be aged or subjected to environmental factors that reduce the quality of DNA obtained. These sample-specific issues support the development of specialized extraction techniques for bone in order to obtain the highest quantities of DNA and improved quality STR profiles.

Prior work has demonstrated that increased DNA quantities are obtained from human skeletal remains when using an SDS detergent-based buffer in conjunction with Collagenase Type II (CLSII) enzyme. It has also been determined that metals, particularly calcium, copurify with DNA when processing bone samples. These copurified metals have been shown to inhibit PCR amplification of STR markers. Building upon these findings, a protocol was designed to determine whether use of an anionic detergent stronger than SDS would continue to improve upon current methods for DNA extraction from human bone. DNA was purified on the EZ1 Advanced XL System (Qiagen, Hilden, Germany) after employing this modified digestion step. The DNA isolate was quantified using the Investigator Quantiplex HYres Kit (Qiagen), then STR markers were amplified with the Investigator 24Plex QS Kit (Qiagen) and fragment analysis was performed with the 3500xL Genetic Analyzer (Thermo Fisher Scientific, Inc., Carlsbad, CA). STR profiles were analyzed using GeneMapper® ID-X v1.4 (Thermo Fisher Scientific). Results indicate that using increased strength anionic detergent in conjunction with CLSII enzyme will improve quantities of DNA obtained and increase the quality of STR profiles produced from human skeletal remains.

Results indicate that full profiles were recovered for all concentrations of SDS tested and Buffer ATL. Peak height ratio was maintained at or above 0.6 at 1-2% SDS. This was a qualitative improvement compared to samples digested using CLSII enzyme with Buffer ATL. These results will be compared to samples digested using CLSII and buffers containing Sodium N-Lauroylsarcosinate and Triton™ X-100.