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SODIUM PHOSPHATE ENHANCED DNA EXTRACTION FROM BONE

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The forensic community frequently uses bone tissue for DNA typing the remains of missing persons, crime victims and victims of mass disasters (Hochmeister et al. 1991, Hagelberg et al. 1991, Gill et al.1994, Holland et al. 2003). Extraction of DNA from bone can be difficult due to a compact calcified matrix combined with a relatively low cellular content. Consequently, improved methods of dealing with bones' calcified matrix during DNA extraction should prove beneficial to assist forensic practitioners in human identification. The predominant inorganic component of bone is hydroxyapatite (HA), a stable calcium phosphate compound ($[Ca_5(PO_4)_3OH]_2$) that is the major contributor to bones' structure and compact matrix. Several current forensic protocols include lengthy incubations in a high concentration of EDTA to decalcify skeletal remains and interfere with the interaction of DNA with HA. In this study, we have pursued faster, alternative methods of disrupting the interaction between DNA and HA .By incorporating high concentrations of sodium phosphate (NaP) in the extraction buffer, the binding of DNA to the endogenous HA of bone may be blocked or reversed. Our preliminary work showed that this extraction method is suitable for downstream mitochondrial DNA (mtDNA) analyses and can be completed in as little as two hours. More in depth studies included optimization of NaP concentration and DNA purification conditions. Depending on the bone, this extraction method yields at least a 2,000-fold increase in mtDNA copy number concentration when compared to protocols that do not disrupt the HA-DNA interaction. In addition, initial work indicates that STR profiles can be obtained from these extracts by optimizing PCR conditions. In summary, application of this protocol to forensic science has the potential to dramatically reduce the amount of time required to isolate DNA from bone samples, increase mtDNA yields and possibly improve chances of obtaining STRs from skeletal remains. Keywords: mtDNA, extraction, skeletal remains