AN OPTIMIZED PROTOCOL FOR EXTRACTION OF MITOCHONDRIAL DNA FROM HAIR SHAFTS

Erin S. Burnside, M.S., Brittania J. Bintz, M.S., Mark R. Wilson, Ph.D.
Western Carolina University, Chemistry & Physics, 111 Memorial Drive NSB #231, Cullowhee, NC 28723, USA

Forensic scientists are often faced with the challenge of limited or degraded samples, where a full nuclear DNA profile may be difficult to obtain. In these instances, mitochondrial DNA (mtDNA) analysis can be particularly useful, as mtDNA is more easily recoverable from challenging sample types such as hair shafts and bone. Current extraction protocols generally yield enough mtDNA from two centimeters of hair shaft to reliably sequence two hypervariable regions (HV1 and HV2) of the mtGenome. However, HV1 and HV2 comprise only about 4% of the entire mtGenome. This, coupled with the fact that mtDNA is, by nature, less discriminatory than nuclear DNA, limits the utility of mtDNA analysis.

In this study, we have combined traditional extraction methods with two kit-based extraction methods (Qiagen® QIAamp® DNA Investigator and Applied Biosystems® PrepFiler® Forensic DNA Extraction Kits) to optimize the mtDNA yield recovered from two centimeters of hair shaft. Preliminary results indicate a consistent ten-fold increase in mtDNA copy number / µl over methods currently used in crime laboratories, as measured by a custom real-time qPCR assay (Kavlick *et al.* 2011). With this improvement in extraction efficiency, we have also seen promise in the ability to use whole genome amplification (WGA) to further increase the analyzable amount of mtDNA for downstream applications. Our goal is to maximize the mtDNA extracted from two centimeters of hair shaft so that more mtGenome sequence information may be obtained for comparison to a reference sample, leading to a higher discriminatory power of mtDNA analysis. ⌘