B118 The Utility of Whole Genome Amplification in Forensic DNA Analysis

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After attending this presentation, attendees will understand the theoretical and practical limitations of two methods of whole genome amplification (WGA) for use with forensic samples will be presented, specifically focusing on the effects of poor DNA quality and/or quantity.

Although techniques used to analyze DNA have progressed greatly in the past decade, limited DNA template can still hinder analysis. An ideal solution to this problem would be the amplification of the entire genome, prior to PCR, so that any locus could be examined as needed. This presentation will impact the forensic community and/or humanity by raising awareness about the advantages of using whole genome amplification in forensic DNA laboratories.

The theoretical and practical limitations of two methods of whole genome amplification (WGA) for use with forensic samples will be presented, specifically focusing on the effects of poor DNA quality and/or quantity.

Forensic biologists are often confronted with issues of limited DNA that may restrict their ability to generate a complete genetic profile, while reserving sample for future analysis or repeat analysis by the defense. The polymerase chain reaction (PCR) is commonly used to amplify specific loci, effectively alleviating some issues of low quantities of starting DNA. However, limited DNA template can still hinder analysis, especially if addi- tional assays are requested such as Y-chromosome analysis, mtDNA sequencing, or private defense examination.

An ideal solution to this problem would be the amplification of the entire genome so that any locus could be examined as needed. WGA is a process which is capable of replicating an entire sample, reducing the chance of entirely consuming evidence. WGA of high quantity clinical material has been studied extensively, but few investigations characterizing WGA for forensic use have been reported. Specifically, WGA has not been widely tested on forensically relevant samples, most importantly those con- taining DNA that is limited in quantity or degraded. The objective of this project was to characterize well-developed WGA methods—Improved Primer Extension Preamplification (I-PEP) and Multiple Displacement Amplification (MDA)—on samples commonly seen in forensic labora- tories including those with low DNA quality and quantity.

For this research, control and artificially degraded DNA, as well as DNA from hair, fresh and aged blood, and aged bone, was amplified using both WGA methods. Differences in the suitability of untreated and WGA treated samples were compared for use in downstream analysis. Nuclear and mtDNA were PCR amplified to test maximum amplicon lengths and to examine WGA yields. Amplification of the single copy nuclear gene amel- ogenin was undertaken using primers designed to generate 200 or 400bp product. Multilocus STR profiles were generated using the ABI AmpFLSTR® Identifiler® PCR amplification kit. A variety of mtDNA control region amplicon sizes were also tested.

For high molecular weight DNA, product yield could be increased 20 to 2000 fold by I-PEP and 1000 to 10,000 fold by MDA. However, sub- stantial differences were observed in the maximum PCR product length of untreated and whole genome amplified product, particularly from degraded material. MDA tests on low quality samples were generally unsuccessful and often resulted in extensive non-target DNA amplification. Overall, I- PEP and MDA increased the product yield of high quality DNA, but these methods had limited success on highly degraded samples.

Whole Genome Amplification, DNA Amplification, Degraded DNA

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