B168 Application of Multiple Displacement Whole Genome Amplification to Forensic DNA Analysis

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The goal of this presentation is to convey research findings evaluating the fidelity of whole genome amplification by multiple displacement amplification from DNA extracted from aged bloodstains.

This presentation will impact the forensic community and/or humanity by describing research on the forensic applicability of a technique which may allow forensic scientists to overcome the problem of limited sample sizes and open the door for many additional types of forensic DNA analysis that can be useful in the investigation of crime.

The recent completion of the human genome sequencing project has created many opportunities for further elucidation of human genetics. Such advances continue to fuel the explosive growth of forensic DNA technology. The bottleneck in forensic science is that the quantity and condition of forensic evidence samples frequently limit the extent of analysis. Recent improvements in whole genome amplification may allow forensic scientists to overcome the problem of limited sample sizes and open the door for many additional types of forensic DNA analysis that can be useful in the investigation of crime.

Multiple displacement amplification (MDA) is a method of whole genome amplification (WGA) which is capable of providing large quantities of human genomic DNA from limited sample sources. It is a relatively new method which, although validated for various types of clinical samples (whole blood, cheek cells, etc.), has yet to be tested on the type of samples commonly encountered as forensic evidence. This research seeks to determine whether WGA/MDA is a suitable means of amplifying human genomic DNA from samples of a forensic nature. Current forensic DNA methods amplify targeted areas of the human genome in order to produce a genetic profile for the sample of interest. However, forensic evidence samples are frequently limited to that which is collected from a crime scene. A method that could accurately and representatively amplify the entire sample genome would provide sufficient DNA for multiple analyses and for archival storage. Minute samples could be amplified to ensure a sufficient quantity of DNA for both prosecution and defense teams to analyze. Amplification of the entire genome would also create sufficient DNA to analyze genomic loci that provide information useful in determining phenotypic characteristics of the unknown source of biological evidence from a crime scene.

This research will determine whether WGA/MDA can provide accurate STR profiles from DNA extracted from dried bloodstains. Blood will be drawn from anonymous donors for the preparation of control DNA and bloodstains. Control DNA will be extracted from fresh blood. Known volumes of blood will be stained onto a white cotton substrate, dried, and stored at room temperature. Stains will range in size based on the original volume of fresh blood used to stain the substrate: 1µl, 10 µl, and 50 µl. As an extraction control, the same volumes of blood will be aliquoted directly into microfuge tubes and stored under the same conditions as the bloodstained swatches. DNA will be extracted from the stains and extraction controls at various time points, ranging from Day Zero to 10 weeks, using Gentra's PureGene kit. Extracted DNA will be quantitated using PicoGreen fluorescence. DNA extracted from aged bloodstains and extraction controls will be amplified using Amersham's GenomiPhi Kit. Control DNA from the freshly drawn blood will also be amplified to compare the aged versus control DNA. The amplified DNA will be quantitated using PicoGreen fluorescence. The whole genome amplified DNA will then be analyzed for 7 of the 13 short tandem repeat (STR) loci required by the FBI for inclusion in the CODIS database system. The seven STR markers are D5S818, D13S317, D7S820, FGA, D3S1358, D21S11, and D18S51. Control DNA that has not been amplified via GenomiPhi will also be analyzed in these 7 loci. STR profiles from control DNA will be compared to the STR profiles obtained from the GenomiPhi amplified aged bloodstain samples to determine if the latter samples are capable of providing accurate DNA profiles.

Forensic DNA Analysis, Multiple Displacement Amplification, Whole Genome Amplification

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