DEVELOPMENT AND EVALUATION OF A RAPID PCR METHOD FOR THE POWERPLEX®S5 SYSTEM FOR FORENSIC DNA PROFILING

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After attending this presentation, attendees will have a brief understanding of how using fast chemistry and rapid PCR methods can increase sample throughput using a miniSTR system. This presentation will impact the forensic community and/or humanity by providing a new rapid PCR protocol for a miniSTR system. This will allow laboratories to significantly reduce backlogs in cases involving low amount DNA template or degraded samples that may have become more difficult to analyze.

Forensic DNA profiling is a multi-step process taking approximately 10 h to complete, with PCR amplification requiring 3 to 4 h. A reduction in the amount of time required for the amplification step would allow for faster human identification and increase laboratory throughput. This can be accomplished using fast chemistry and rapid PCR cycling instrumentation. Fast chemistry involves the application of new polymerases and buffers that maintain the levels of sensitivity and fidelity of standard polymerases but increase the extension rate and are activated more quickly. These fast chemistries are designed to be paired with rapid PCR cycling instrumentation where temperature transitions between annealing and extension PCR steps and holding times are shortened. The goal of this work was to optimize and evaluate a rapid PCR method for the PowerPlex®S5 system for forensic DNA profiling.

By pairing fast chemistries with a fast thermal cycler, the authors were able to reduce the amplification time for the PowerPlex®S5 system by 70% (1 h). Sensitivity and heterozygous peak height ratios were comparable between the fast and standard protocols. However, a substantial decrease (5%) in peak height ratio was noted at the D18S51 locus with the fast cycling method. An increase in average mean stutter for combined loci of 2.6% was observed in profiles amplified using the fast protocol compared to the standard system.

The results of this study demonstrate that a fast amplification protocol for a miniSTR system can be used for forensic DNA profiling. Further optimization and validation of the fast PowerPlex®S5 protocol are required before it can replace the standard amplification protocol in forensic laboratories.

Keywords: Short Tandem Repeats, Rapid PCR, DNA Typing