

DATABANK APPLICATIONS OF DIRECT AMPLIFICATION USING SWABSOLUTION™ AND THE POWERPLEX® FUSION AMPLIFICATION KIT: A VALIDATION STUDY

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Due to recent expansion of the Combined DNA Index system (CODIS) database in Wisconsin, the DNA Databank Unit will be implementing several techniques for the collection and processing of reference DNA samples that will allow a significant reduction in cost and an increase to efficiency while maintaining a high success rate. The purpose of this study was to validate the direct amplification procedure using SwabSolution™ and the PowerPlex® Fusion amplification kit. For use in our laboratory, the procedure from the SwabSolution™ Technical Manual was modified to reduce the amount of substrate needed (¼ of a swab head instead of a whole) as well as the amount of reagent needed (300µL of SwabSolution™ Reagent per sample instead of 1.0mL). Setup for amplification and genetic analysis via gel capillary electrophoresis (CE) was validated manually as well as robotically on the Tecan Freedom Evo. An optimization study demonstrated that 3.0µL of swab extract and 25 cycles on a GeneAmp 9700 thermal cycler generated quality DNA profiles on an ABI 3500xL Genetic Analyzer. All samples were analyzed using Gene-MapperIDX v1.4. The Limit of Detection Threshold (analytical threshold) and Laboratory Interpretation Threshold (stochastic threshold) were determined through a sensitivity study that utilized target amounts of organically extracted DNA applied to cotton swabs. The remaining lysate from the direct amplification procedure was also evaluated for its use in DNA IQ and organic extractions. In conclusion, direct amplification will allow the Wisconsin State Crime Laboratory to streamline sample processing as reference sample intake continues to increase without creating a backlog and maintaining a high level of integrity.