AN EVALUATION OF DIRECT PCR AMPLIFICATION

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The objective of this research was to generate complete autosomal STR profiles from body fluids using direct amplification and various commercially available STR amplification kits. Attempts were also made to detect the Y-profile from male body fluids using 1.2mm punches and AmpFℓSTR® Yfiler™ kit following direct amplification procedure.

The following nine collection media were used for this study: proPRIME *i* Indicating Micro, 705 Micro, Blood Direct #1 and #2, Collection Card, CEP swab from FITZCO, EasiCollect from Whatman, FTA Indicating Micro and Bode DNA Collector. Blood from two deceased individuals and saliva from three living donors were used in this study. The three single source saliva samples and two single source blood samples were deposited on each of the 45 collection devices.

A 1.2mm punch of each of the 45 substrates containing one body fluid was amplified with PowerPlex® 18D, PowerPlex® 16 HS, PowerPlex® 16, and PowerPlex® 21 Systems from Promega Corporation, and AmpFℓSTR® Identifiler® Direct, Identifiler® Plus, and Identifiler® PCR Amplification Kits from Applied Biosystems following each manufacturer's recommended conditions. Similarly, 1.2mm punches of the substrates containing male body fluids were amplified with AmpFℓSTR® Yfiler™ PCR Amplification Kit.

Results from the eight kits mentioned above were compared. Both blood and saliva samples appeared to yield complete DNA profiles. Two different reaction volumes were attempted with substrates that yielded complete profiles from single source samples, the first using the manufacturer's recommended volume, and the second using half of the reaction volume suggested in the protocol. For some of the substrates thermal cycling conditions were modified as necessary to generate complete DNA profiles.

Another goal of this research was to demonstrate that direct PCR amplification can be applied to commercially available kits not intended for direct amplification in the forensic community. The results indicate that it is possible to do so.