STR TYPING FROM BLOODSTAINED FABRIC WITHOUT DNA EXTRACTION

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A concordance study between the Ampf/STR Profiler Plus™ and Ampf/STR COfiler™ DNA typing system™ (Perkin Elmer Biosystems), and the GenePrint® PowerPlex™ 16 System (Promega Corporation) was carried out using samples of bloodstained fabric and without conventional extraction procedure.

Highly polymorphic short tandem repeat (STR) loci or microsatellites offer great promise for forensic analysis, paternity testing and gene mapping. STR loci consist of simple tandemly repeated sequences 1-6 base pairs in length.

The goal of this research was to determine if the current primer sets, GenePrint® PowerPlex™ 16 System and the Ampf/STR Profiler Plus™ and AmpF/STR COfiler™ DNA typing system™ from the two commercially available manufacturers, Promega Corporation and Perkin-Elmer Biosystems respectively, produce concordant results using bloodstained fabrics without organic or inorganic extraction procedure.

Whole blood from various individuals was spotted on 100% white cotton fabric. Once the bloodstains were dry, approximately 1 mm or less of the bloodstained thread was cut from each sample and placed in an autoclaved reaction tube containing either TE buffer or sterile distilled water. The tubes were then subjected to a simple preincubation period as described by the author previously (1). After the preincubation was completed, a mixture of respective PCR Reaction Buffer, Primers and AmpliTaq Gold Polymerase was added into each of the tubes. The bloodstained threads now submerged in the mixture were then amplified.

PCR multiplex amplification was performed using the Ampf/STR Profiler Plus™ and Ampf/STR COfiler™ kits (Perkin Elmer) following the manufacturer’s recommended protocol. Thermocycling, for the two separate reactions, was carried out according to the protocols recommended by the manufacturers. Fifteen STR loci and the Amelogenin locus co-amplified using the three-color detection system provided by the GenePrint® PowerPlex™ 16 system. All sixteen loci were amplified simultaneously in a single tube in the Perkin-Elmer GeneAmp® 9700 thermal cycler.

To test the validity of the results, bloodstained fabric samples were also extracted using organic extraction. The quantity of extracted DNA was determined by using the slot blot hybridization method and the Perkin Elmer QuantiBlot® kit. The extracted DNA samples were also subjected to amplification and detection.

All loci using the three kits were detected when analyzed by the ABI Prism® 310 Genetic Analyzer. Amplified STR alleles were separated by capillary electrophoresis which is capable of resolving amplification products varying in size by a single base pair. This allows for accurate allele identification and sizing.

Most of the bloodstained fabric amplified and alleles were called correctly. While some of the bloodstained samples did not amplify in all of the loci, the alleles detected using the three kits were consistent with results obtained from DNA extracted organically. Results using both of the kits were similar.