PCR Volume Reduction Study Using Bloodstained FTA™ Collection Cards and Capillary Electrophoresis

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The technology of STR DNA analysis has enabled the establishment of cost-effective and efficient investigative offender databases. With the enactment of Bill C-3 (The DNA Identification Act) anticipated in the fall of 1998, Canada will begin banking an annual estimate of 28,000 to 98,000 samples obtained from individuals convicted of primary and secondary designated offenses. The undertaking of such a project required consideration of many factors involving the scientific technology, security and privacy, quality assurance, as well as legislative realities. From a practical perspective, collection, cost, processing time and long-term storage of the biological materials become important issues.

A study was conducted to examine whether a reduction in PCR amplification volume from 25 µl to 5 µl would produce valid and efficient results. Blood samples from 10 individuals were collected on FTA™ collection cards and processed for DNA. Amplification was carried out using the AmpF*l*STR™ *Profiler Plus*™ kit (Perkin Elmer) in a 25 µl or 5 µl reaction for 23 cycles with a 1.2mm diameter processed FTA™ sample (Harris Micropunch). The amplicons were analyzed by capillary electrophoresis using the ABI PRISM® 310 Genetic Analyzer (Perkin Elmer).

Results showed that the yield of amplified product was enhanced by as much as six-fold when a 5 µl reaction volume was utilized. The quality of the amplicons was assessed for consistency and balance of peak heights, trailing, non-templated A-addition, stutter, and heterozygote ratios. The issue addressed in this study is to asses whether any potential negative effects due to the volume reduction would outweigh the benefits of the increased product yield when using a 5 µl reaction volume. In addition, yield variability based on allele peak height was evaluated using both 5 µl and 25 µl amplification volume formats. Our studies demonstrate that a reduced amplification volume and the FTA™ extraction procedure provide a rapid method for reliable and reproducible STR profiling, with potential implications for both databasing and casework.