B27 A Rapid and Simple Elution Method of DNA From Whatman® FTA® Classic Cards

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After attending this presentation, attendees will understand how 1.2mm FTA® punch disc is used for direct amplification using ABI Identifiler™ in the Singapore National DNA Database Laboratory. A rapid, inexpensive, and simple elution method was developed to elute DNA from the FTA punch disc so that amplification volume of 10ul or less can be achieved.

This presentation will impact the forensic community and/or humanity by demonstrating how 1.2 mm FTA™ punch disc is used for direct amplification using ABI Identifiler™ in the Singapore National DNA Database Laboratory. A rapid, inexpensive, and simple elution method was developed to elute DNA from the FTA™ punch disc so that amplification volume of 10 ul or less can be achieved. This method of elution will benefit the forensic community in being informed of a simple, cost-effective, and rapid methodology to elute DNA from FTA™ classic cards.

The Singapore National DNA Database Laboratory performs DNA typing using ABI® Identifiler™ through direct amplification on 1.2 mm FTA® punch disc. FTA® punch discs are washed using the Beckman Coulter® Biomek® 2000 robotic workstation on 96-well PCR plates and dried in oven before the amplification reagents are added.

Using direct amplification on 1.2 mm punch discs results in amplification failures when amplification volumes of 10 ml or less are attempted. Isolation of DNA from FTA™ cards can be achieved by extraction techniques using organic extraction or commercial DNA extraction kits. Alternatively, alkaline conditions or use of restriction enzymes such as Pst I can be used to elute the DNA. However, with the exception of the use of alkali, these methods previously described are both costly and require long incubation time.

Described here is an inexpensive, rapid, and simple method to elute DNA sufficient for DNA typing. Briefly, 10 ml of deionized water are added onto two 1.2 mm FTA® punch discs that have been washed and dried in each well in the PCR plate. The PCR plate is then sealed and heated for 95ºC for 10 minutes, flash vortex, and centrifuged at 3000 rpm for 15 minutes. 3.5 ml of the FTA® eluate from the PCR plate is added to a PCR plate, pre-aliquoted with 4 ml of Identifiler™ PCR master-mix on the Beckman Coulter® Biomek® 2000 robotic workstation, sealed before amplification using 28 PCR cycles.

Using this method of elution, the DNA concentration estimated using ABI® Quantiblot® is approximately 0.48 ng/ml with the average total yield about 3.84 ng, which is optimal for amplification using Identifiler™. Out of the 82 samples genotyped using the ABI 3100 Genetic Analyzer, 81 returned a full DNA profile with the exception of one which returned a partial profile and required a re-injection using a higher voltage in order to obtain a full DNA profile. Full concordance was returned when the DNA profiles were compared to direct amplification of FTA® punch approach.

In conclusion, the elution of DNA from FTA® punch discs allows the amplification volume to be amendable to smaller PCR volumes of 10 ml and below. Excess DNA from the FTA® eluate can also be stored and used for other DNA analysis work without processing additional FTA® punches.

FTA™ Cards, DNA Database, Forensic DNA

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