Biomek®-3000 and GenPlex in Forensic Genetics

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INTRODUCTION

SNP genotyping provides a supplement for conventional STR-based kits currently used for human identification. GenPlex (Applied Biosystems (AB)) is a SNP genotyping kit based on a multiplex of 48 informative, autosomal SNPs from the SNPforID Consortium [1] plus the amelogenin gender marker [2]. Our objective was to setup, implement and validate a small and affordable, automated liquid handling robot for forensic case work samples (buccal swabs on, FTA® paper and Qiagen purified blood).

MATERIAL AND METHODS

The reaction scheme consisted of numerous steps (Figure 1) and was difficult to perform consistently without an automated liquid handler. All steps were performed in 96-well micro titre plates. Automation was accomplished with a Biomek-3000 (Beckmann Coulter) automated liquid handler using five in-house developed methods. All methods allowed the user to select the number of subsequent injections on the capillary electrophoresis instrument (AB 3130xl, AB) enabling processing of both partial and full plates.

RESULTS

The results obtained with 15 manually processed samples were compared to results obtained with the same samples processed on the Biomek-3000 (Figure 2). Full concordance between manually processed and automated processed samples were obtained in all genetic systems. Subsequently, a total of 286 samples were analyzed in duplicates with the GenPlex reaction using the Biomek-3000. Of the total of 572 samples, 97.6% resulted in a full profile. Full concordance was obtained between the two investigations of each sample. The results were further compared to those obtained from the same samples using a 49-plex PCR in combination with an ISO 17025 accredited SNPlux® (AB) single base extension assay [3]. Full concordance of the results was obtained in all but one sample resulting in 99.99% concordance.

Figure 2. A representative sample manually processed and processed on the Biomek-3000.

SNPlex Hybridization Plate

CONCLUSIONS

Overall, the results demonstrate that the Biomek-3000 can perform a series of complex reactions leading to highly consistent forensic genetic SNP typing results. Further key findings:

- Complete SNP profiles are reproducibly obtained using 250 pg input DNA.
- The performance of the GenPlex reaction is unreliable when the amount of DNA is below 100 pg.
- Some miscalled alleles were observed in samples with 50 and 20 pg of DNA.
- Cluster analysis can be used to identify mixtures.

REFERENCES