

sbeadex[®] forensic kit for automated processing

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Introduction

LGC Genomics' magnetic bead technology uses in-house developed special magnetic particles and kits to separate nucleic acids and is well suited for the complete automation of the DNA extraction process because it avoids manual interaction such as centrifugation. The purpose of this study was to assess the performance of the sbeadex[®] forensic kit in recovering DNA from forensic reference samples in line with the technical standards for loading profiles to the UK National DNA Database (UK NDNAD).

Experimental design

Three sample types routinely submitted as forensic reference samples were processed during the evaluation study. Buccal swabs (n=225), whole blood samples (n=18) and hair roots (n=18) were collected from staff donors with known STR profiles. A liquid handling robotic platform was used to carry out the sbeadex[®] DNA purification. The system is able to manipulate volumes up to 200 µL, therefore the standard sbeadex[®] protocol was adapted to suit the specification of the robotic workstation. The concentration of the resulting DNA extracts was measured by fluorescence using PicoGreen[®] dsDNA reagent. Samples were amplified using SGM Plus[®] PCR kits (Life Technologies) and were run on capillary electrophoresis genetic analysers (Life Technologies) before analysis using GeneMapper-ID analysis software (Life Technologies).

The following parameters were measured to assess the quality of the resulting DNA profiles:

- Concordance of allelic designations
- Success rates and full profile rates
- Profile quality including stutter peaks, n-peaks, background, artefact peaks, and heterozygote balance.

Assessment against evaluation criteria

All samples resulting in full acceptable profiles (measured against criteria for loading to UK NDNAD) produced concordant results with the known donor profile. Success rates were comparable to the success rates achieved when using the chemistry previously validated for the robotic system (validated kit A). Table 1 describes the percentage of samples by type that passed the technical standards required to enable loading to the UK NDNAD after one pass through the system.

Of the 27 samples that failed to meet the acceptance criteria 13 required re-electrophoresis and 14 required re-amplification. After necessary reprocessing the overall pass rate for the data set was 99.2 %. Two hair samples were poor quality and therefore full acceptable profiles could not be obtained.

The average peak heights for the sbeadex[®] data sets were higher than the average peak heights achieved from the validated kit A for all loci 1250 rfu compared to 719 rfu. Comparison of the average DNA yields showed that the sbeadex[®] protocol outperformed validated kit A protocol, 32.7 pg/µL compared to 25.4 pg/µL respectively.

Heterozygote balance between the sbeadex[®] data set and the validated kit A data set were compared by calculating the heterozygote peak area ratio (PAR), Figure 1.

Sample type	n	Number passed	Pass rate (%)
Hair	18	12	66.7
Blood	18	16	88.9
Buccal swab	225	206	91.6
Total	261	234	89.7

Table 1: First time pass rates by sample type

Heterozygous balance

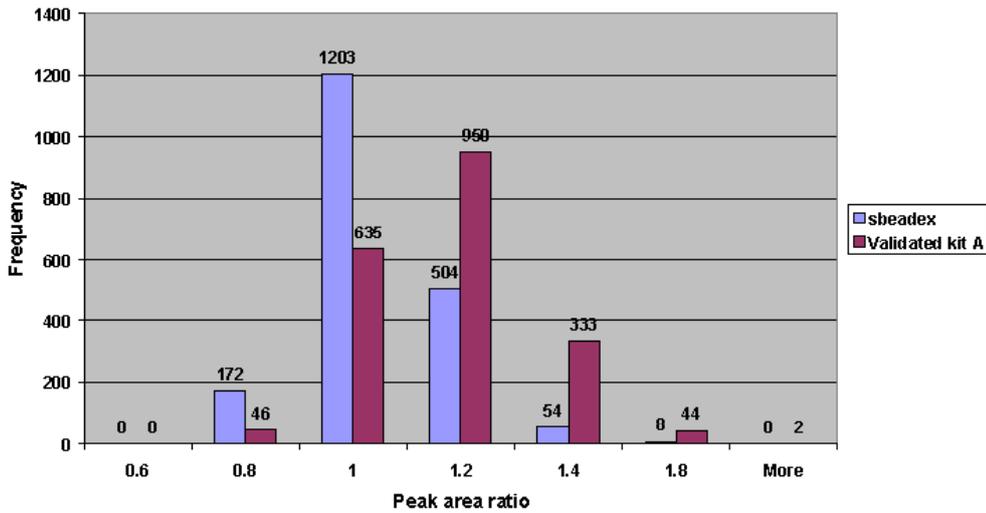


Figure 1: Shows the distribution of peak area ratio for heterozygous loci in the two data sets

The sbeadex® data had an average PAR of 1.0 compared to the validated kit A data set which had an average PAR of 1.1. No samples within the sbeadex® failed to meet the technical standard for heterozygous balance.

There were no incidences of samples failing for the presence of artefact peaks, excessive stutter and n-peaks or for poor background (Figure 2).

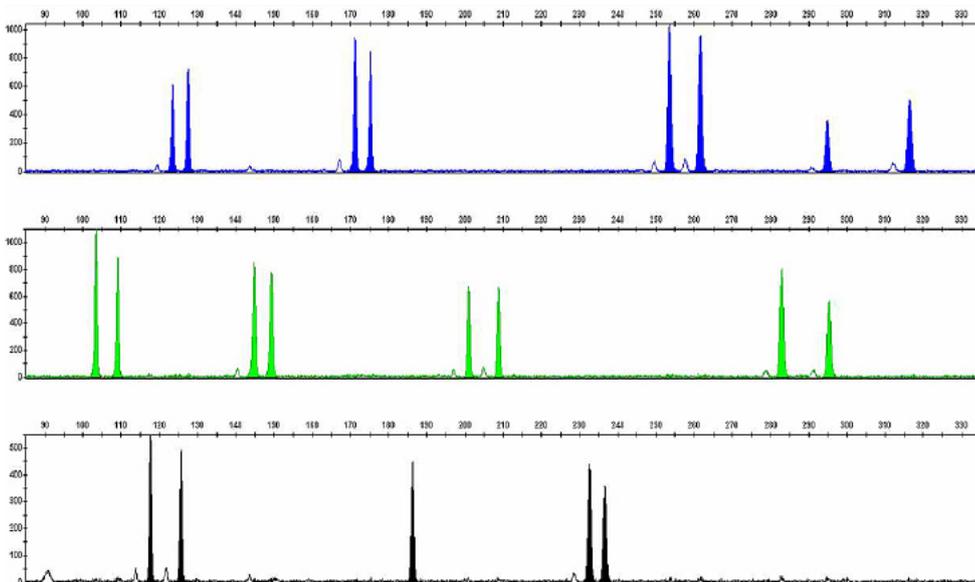


Figure 2: Shows a typical STR profile generated from template DNA prepared with the automated sbeadex® forensic kit protocol

Conclusion

The results show that the sbeadex® protocol is comparable to the existing validated kit A currently being used. The performance of the sbeadex® forensic kit and

protocol at recovering DNA from forensic reference samples in line with technical standards for loading profiles to the UK NDNAD is acceptable.



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