In 2010, the FBI CODIS Core Loci Working Group recommended expansion of the CODIS Core Loci in the United States (Harex, 2012). Reasons given for expanding the CODIS Core Loci from the usual 13 loci to 24 loci included the need for international data compatibility and increased power of discrimination in missing person cases. Both Promega PowerPlex® Fusion and Applied Biosystems® GlobalFiler™ Express PCR systems have incorporated these changes.

Sensitivity, precision, concordance, reproducibility and contamination studies were performed as part of an internal validation study of these systems performed on an Applied Biosystems® 3500 Genetic Analyzer. The goal of this study was to validate both commercial kits produce reliable and robust results, in addition to identifying a single thermal cycling parameter and single injection time for both systems using FTA cards (blood and saliva) and buccal samples.

METHODS AND INSTRUMENTATION

Materials and Reagents:
- FTA Cards (blood and saliva)
- Buccal Swabs
- Prep-n-Go™ Buffer
- SwabSolution

Instrumentation:
- Applied Biosystems® GeneAmp® PCR System 9700
- Applied Biosystems® 3500 Genetic Analyzer
- GeneMapper® ID-X Software Version 1.4

RESULTS

PowerPlex® Fusion:
- Incubation room temp for 20 min
- Reagents need to thaw before use
- Uniform reaction mix
- DNA Control 2800M: needs dilution
- Approx 90 min Amplification

GlobalFiler™ Express:
- Incubation room temp for 20 min
- Reagents ready for use
- Uniform reaction mix
- DNA Control 2800M: no dilution needed
- Approx 80 min Amplification

CONCLUSIONS

Both amplification kits produce robust and reliable results. The following parameters are recommended for the Department of Forensic Sciences:

PowerPlex® Fusion
- 26 cycles with a 12 second injection for blood and saliva on FTA cards, as well as buccal swabs samples. Amplification will involve one 1.2mm punch of saliva or blood on FTA cards and 2 μl of swab DNA. If drop-out or single peaks below the stochastic threshold are observed, these samples may be improved with a 24 second injection.

GlobalFiler™ Express
- 25 cycles with a 15 second injection for blood and saliva on FTA cards, as well as buccal swabs samples. Amplification will involve one 1.2mm punch of saliva or blood on FTA cards and 3 μl of swab DNA. If drop-out or single peaks below the stochastic threshold are observed, these samples may be improved with a 30 second injection.

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http://www.ingams.org/techguide/calculator/index.html


ABSTRACT

In 2010, the FBI CODIS Core Loci Working Group recommended expansion of the CODIS Core Loci in the United States (Harex, 2012). Reasons given for expanding the CODIS Core Loci from the usual 13 loci to 24 loci included the need for international data compatibility and increased power of discrimination in missing person cases. Both Promega PowerPlex® Fusion and Applied Biosystems® GlobalFiler™ Express PCR systems have incorporated these changes.

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INTRODUCTION

Direct amplification eliminates the need to perform DNA extraction and DNA quantification of reference samples. Two of the most novel direct PCR kits are the PowerPlex® Fusion and the GlobalFiler™ Express Amplification Kits. These kits were developed as part of the effort to extend the Combined DNA Index System (CODIS) core loci.

The expansion allows an increase in international data compatibility and discrimination power to aid in missing person cases. It also reduces the likelihood of adventitious matches due to the rapid increase in numbers of profiles stored in National DNA Index System (NDIS).

The internal validations were necessary before implementing a procedure for the analysis of blood and saliva samples on FTA cards, as well as buccal swabs. The side-by-side validation allowed Department of Forensics to properly evaluate the advantages and disadvantages of both kits to choose the one better suited for their needs.

VALIDATION STUDIES

- Sensitivity
- Precision
- Concordance
- Reproducibility

*Not discussed in this paper

Do not hallucinate.


DISCLAIMER

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