Detection of Fluorescent Labeled STR with Silver Staining

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Introduction

Short Tandem Repeat (STR) sequences are one of the most important systems for paternity and forensic DNA typing. Simultaneous amplification of different loci is possible and several commercial kits are available. We examined the feasibility of fluorescent labeled primers using a standard silver staining technique.

We used the GammaSTR[™] system (Promega Corp., Madison, WI). This system has four STR loci, D16S539, D7S820, D13S317 and D13S818, which enables high discrimination and do not have overlapping allele size ranges.

Materials and Methods

Whole blood from ten unrelated individuals was obtained from the Ecuadorian Red Cross Blood Transfusion Services. DNA was extracted by the Chelex method. The amplification of the four STR loci was performed using the GammaSTRTM system. A twenty five µl reaction was prepared according to the manufacturer's recommendations. Electrophoresis of PCR products was performed under standard conditions in 4% and 6% polyacrylamide gels. The electrophoresed STR fragments were visualized by traditional silver staining. Alleles were assigned by comparison with the respective ladder.

Results and Conclusions

The silver staining technique works well with commercial fluorescent STR systems using STR loci with non-overlapping allele size ranges. The fluorescent labeled method does not interfere with the interpretation. There were some weak non-specific amplifications but these did not interfere with the analysis.

The 4% polyacrylamide gel had some double denatured fragments and this was solved with 6% gel. The GammaSTR[™] kit has a high discrimination in different populations. Furthermore the silver staining is economic and easy to perform. Therefore, GammaSTR[™] using silver staining is a good alternative for paternity and forensic DNA typing. It can be used with other fluorescent kits.

In summary, the silver staining method would be useful for small laboratories for fluorescent-labeled primers without fluoremetric technology.