



B134 Optimization of Mini Plex Primer Sets for STR Testing of DNA Extracts From Bone Samples 10 – 13 Years Postmortem

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The International Commission on Missing persons has developed a DNA led identification system for identification of victims of the conflicts resulting during the breakup of the former Yugoslavia. As a first step in the identification effort bone samples were taken from whole skeletons and tested using a silica-based extraction method and the Promega PowerPlex® 16 system. With the PP16 system results were obtained in approximately 85% of the samples tested. The 15% that were considered failures, during the first phase, had between 0 and 11 loci amplified with most of the failed loci being the longer PCR products.

To better understand why these samples were failing STR analysis extracts were assessed using the Applied Biosystems Quantifiler™ Human DNA quantification system. Most of the samples displayed relatively low amounts of DNA (<10 pg/μl) and/or the presence of PCR inhibitors. The results of both the quantification as well as the allelic drop outs in the longer loci indicate that the DNA in these extracts may be degraded to the point where commercially available STR kits are not effective.

In an attempt to get more STR data out of the DNA extracts, that failed the initial phase of this testing, the possibility of using reduced amplicon primer sets has been investigated. Initially a concordance study was performed to ensure that the alleles amplified by the new primer sets would reflect the same alleles designated by the PP16 system and the Applied Biosystems AMPF/STR® SeFiler™ kits. Reduced amplicon primer sets were also optimized for amplification of DNA isolated from a number of bone samples. The optimizations tested include modifications to the PCR buffer, Mg⁺⁺, and different *taq* enzymes.

The results of the optimizations will be presented and compared to optimizations of the PP16 and SeFiler kits. Additionally the effectiveness of the mini primer sets for STR testing of DNA extracts from bone samples will be discussed in detail.

STR, DNA, Mini Plex