ANALYSIS OF THE UK, EUROPEAN AND CODIS CORE STR LOCI BY PCR / ELECTROSPRAY IONIZATION MASS SPECTROMETRY

D. D. Duncan, K. M. Boles, S. A. Hofstadler, T. A. Hall
Ibis Biosciences, a subsidiary of Abbott Molecular, Inc., Carlsbad, CA

We have developed an assay to genotype the loci within the UK, European, and CODIS STR marker sets. Amplicons were generated for twenty one STR loci plus amelogenin in an 8-well PCR panel. PCR products were then desalted and analyzed on a fully automated electrospray ionization-mass spectrometry (ESI-MS) platform. The resulting highly accurate mass measurements were used to determine the base composition of the PCR products, specifying the number of their constituent dA, dG, dC, and dT residues; allele assignments were derived from the base compositions. Single nucleotide polymorphisms (SNPs) have been observed in many of the STR loci, but SNP variants are undetectable with standard electrophoretic analysis of amplicon length. In contrast, the accuracy of base composition analysis by ESI-MS supports the detection of SNPs with this assay.

Here we describe the preliminary evaluation of the assay. The marker panel was comprised of amelogenin, CSF1PO, D1S1656, D2S441, D2S1338, D3S1358, D5S818, D7S820, D8S1179, D10S1248, D12S391, D13S317, D16S539, D18S51, D19S433, D21S11, D22S1045, FGA, SE33, THO1, TPOX, and vWA. The 8-well PCR panel was arrayed in 96 well prefabricated plates containing all reagents required for amplification, accommodating the analysis of up to 12 samples per plate. The profiles of a panel of over fifty human DNA samples were determined and compared to results obtained with the NGM Select™ and Identifiler® kits (Applied Biosystems). The nominal alleles determined with the ESI-MS assay, ignoring the SNP determinations, were concordant with profiles obtained with the Applied Biosystems kits. Additional parameters for evaluation included species specificity, sensitivity, reproducibility, accuracy, allelic balance, and interlocus balance, and were typical of the forensic ESI-MS assays previously developed. SNP-based variant alleles were observed in most STR loci. Overall, the results indicate that the assay is generally concordant with current methods, but the detection of SNPs provides additional diversity within most loci, thus increasing the discriminatory power of the assay.