Validation of Identifiler PCR Amplification and Typing Kit

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A validation project targeting the amplification of forensic casework with the Applied Biosystem AmpFISTR Identifiler PCR Amplification kit was analyzed on both the 310 and 3130 capillary electrophoresis instruments. The validation consisted of various studies including precision, stutter, threshold, sensitivity, reproducibility, injection time, mixture samples, degraded samples, inhibited samples, and adjudicated case samples. Full (25 mL) and half (12.5 mL) volume amplification reactions were compared side by side with samples of various DNA concentrations. Half volume reactions proved to be more sensitive by exhibiting greater peak heights. Therefore, half volume amplification reactions were utilized for the validation of Identifiler. Identifiler was found to be comparable to Profiler Plus and Cofiler amplification kits in the majority of the studies. Identifiler proved to be more sensitive than Profiler Plus and Cofiler by repeatedly detecting complete DNA profiles down to concentrations of 0.1ng of high quality DNA template. Amplifications using 0.05ng of DNA produced partial profiles above 150 RFU, but exhibited allele dropout and imbalance at heterozygous loci due to stochastic effect. The minor contributors in mixtures were clearly detected down to mixtures of 10:1 of extracted samples. It is unclear if there is a definite advantage for amplification of degraded samples with Identifiler compared to Cofiler and Profiler Plus. It appears that Identifiler is comparable to Cofiler and Profiler Plus for degraded samples, but more research needs to be conducted on degraded and adjudicated case samples prior to the implementation of Identifiler in forensic casework.