

PCR OPTIMIZATION OF STR SYSTEMS ON MULTIPLE THERMAL CYCLERS

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Selection of appropriate instrumentation for the forensic typing workflow requires protocol optimization and verification that the system is reliable and fit-for-purpose. Selection of an optimal thermal cycler for a given laboratories' workflow depends on demonstration of different STR kit chemistries on multiple PCR instruments. Demonstration of this compatibility is not trivial as reliable STR analysis of forensic DNA evidence depends on multiple factors, including the efficiency and reproducibility of the PCR cycling. All these findings need to be sufficiently documented.

Here we present optimized cycling parameters for Promega PowerPlex® STR Systems on several different thermal cyclers, including Eppendorf and Applied Biosystems cyclers. Peak heights and peak height ratios for heterozygous autosomal loci of samples analyzed with PowerPlex Systems using the Eppendorf Mastercycler® pro S and Eppendorf Mastercycler® nexus SX1 were compared with data generated for the developmental validation with the Applied Biosystems GeneAmp® PCR System 9700. Comparable data with low input amount (50 – 100pg) samples as well as humic acid-inhibited samples also demonstrate good performance of these thermal cyclers.

Additional stressing by cycling at annealing temperatures that are lower and higher than the optimal annealing temperature for each PowerPlex System gave the expected results, indicating that these thermocyclers are suitable for use with the PowerPlex Systems. By demonstrating excellent performance of multiple thermal cyclers with optimized protocols, laboratories can choose the cycler that fits their workflow and budget.