HIGH-PERFORMANCE PCR HUMAN IDENTIFICATION SYSTEMS

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In this study, we report on novel human identification systems that combine the following three unique technologies to achieve significant reductions in STR genotyping PCR protocol times and to enable multiplexing of STR loci directly from whole blood. (1) Phusion DNA Polymerase: we have utilized a novel DNA polymerase having a covalent linkage of Sso7d DNA binding protein onto the polymerase domain. The resulting Phusion DNA Polymerase has over 10-fold processivity in comparison to a normal Taq polymerase ensuring robust amplification of all loci and alleles even with short PCR extension times. Phusion also exhibits 3'-'5 exonuclease activity eliminating 'plus A' –products and eliminating the need for the conventionally used long final extension step. Phusion is extremely tolerant to PCR inhibitors present in whole blood and blood anticoagulants and the High-performance PCR Human Identification Systems provide complete STR profiles with up-to 10% of whole blood of the PCR reaction volume. (2) Piko Thermal Cycler: the High-Performance Human Identification Systems have been optimized to run in the Piko Thermal Cycler that has a maximum ramp rate of 10ºC/sec. (3) Ultra Thin Wall reaction vessels: we have made use of the UTW plastic technology with >50% reduction in the wall thickness of the reaction vessels, facilitating rapid thermal transfer between the PCR block and the reagents.

This tripartite solution results in less than 60 min total PCR protocol times for co-amplification of 17 STR loci. The High-Performance PCR Human Identification Systems will allow significantly faster throughput of samples in a forensic laboratory than has been previously possible. The possibility for PCR directly from blood may result in further time and cost savings for the laboratory by eliminating the need for DNA extraction with some samples.