

## **VALIDATION OF POWERPLEX 16HS AND POWERPLEX 18D ON THE APPLIED BIOSYSTEMS 3500 GENETIC ANALYZER AND AN EXAMINATION OF SIGNAL NORMALIZATION IDENTIFILER PLUS**

Erica L. R. Butts, Carolyn R. (Becky) Hill, David L. Duewer, John M. Butler, Peter M. Vallone  
National Institute of Standards and Technology, Biochemical Science Division, Gaithersburg, MD

The need to internally validate new technology is an on-going process within the forensic DNA community. Prior to being implemented into a forensic laboratory, a new product used in forensic DNA analysis must be validated internally within each laboratory to ensure the technology generates reliable and reproducible results. The ABI 3500 Genetic Analyzer is the newest capillary electrophoresis instrument available to the forensic DNA community. An internal validation of the Applied Biosystems 8-capillary 3500 Genetic Analyzer was performed using two commercial short tandem repeat (STR) multiplex kits (PowerPlex 16 HS and PowerPlex 18D). Additionally, the signal normalization feature of the 3500 was evaluated with use of Identifiler Plus due to specific size standard limitations within this feature.

Validation experiments to evaluate performance of the 3500 platform consisted of a precision study, sensitivity study, genotype concordance, and a two-person mixture study. Injection parameters were also varied to identify the optimal injection time for both PowerPlex 16 HS and PowerPlex 18D on the 3500. Results from these studies were used in setting analytical and stochastic thresholds for both PowerPlex 16 HS and PowerPlex 18D kits.

The 3500 instrument provides optional signal normalization intended to provide more consistent signal intensity across injections and between instruments. Normalization requires the use of LIZ 600 v2.0 size standard and therefore can only be used with Applied Biosystems STR multiplex kits. Experiments designed to evaluate the effects of normalization included a sensitivity study and a two-person mixture study. Evaluation of signal normalization will be discussed and the impact of normalization on data interpretation (including analytical and stochastic thresholds) will be presented.