

## High-Throughput STR Typing Using a 96-Lane Microfabricated Capillary Array Electrophoresis Device

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Microfabrication technology produces high-density microfluidic circuits on a single wafer, allowing rapid, low-volume capillary electrophoretic (CE) analyses of genetic samples in a highly parallel fashion. A 96-lane radial microfabricated capillary array electrophoresis ( $\square$ CAE) device coupled with a rotary scanning confocal detection system has previously been developed, demonstrating high-quality electrophoretic separations of sequencing samples in less than 30 minutes with single-base resolution.<sup>1</sup> The  $\square$ CAE system has also been integrated with other on-chip sample preparation steps such as PCR<sup>2</sup> and sample pre-concentration<sup>3</sup> to develop completely integrated and portable sample preparation and genetic analysis systems.<sup>4,5</sup> Here we combine the high discriminatory power of STR analysis with microfabricated capillary array electrophoresis technology to dramatically advance the field of forensic identification. This technology provides the high speed and throughput, reliability, sensitivity, reduced cost, and automation needed to address the increasing case backlogs in forensic laboratories

We have explored the forensic applications of these microchip technologies these possibilities by performing STR analyses using the PowerPlex<sup>®</sup> 16 and Profiler Plus<sup>®</sup> systems with a variety of real-world samples.<sup>6</sup> This 96-sample Micro-CE system produced rapid, parallel sample separations in < 30 min with single-base resolution. These separations are at least 10 min faster than the single-capillary ABI 310 and 30 min faster than the 16-capillary 3100 instruments. The resolution obtained based on the TH01 9.3 and 10 alleles is 0.76 (Rb = 1.3),<sup>2</sup> which favorably compares to resolutions reported using conventional CE instruments.<sup>7</sup> Forty-eighty single-source population samples (28 Hispanic, 3 Asian and 17 African American) amplified with the PowerPlex<sup>®</sup> 16 system were analyzed on the  $\square$ CAE system and showed consistent allele calls for all microsatellites and microvariants compared to the expected results (supplementary materials). This demonstrates the capability of the  $\square$ CAE system to accurately perform high-speed, high-volume, parallel STR sample analyses. We evaluated the performance and reliability of the  $\square$ CAE system in resolving mixtures using PowerPlex<sup>®</sup> 16 samples consisting of male and female DNA amplified at defined ratios (10:0, 9:1, 3:1, 3:2, 2:3, 1:3, 1:9 and 0:10). The 3:1 and 1:3 samples are the lowest ratios in which all minor components were successfully detected and typed. This limit is the same as that achieved using commercial CE instruments. A sensitivity study was performed by typing

PowerPlex® 16 and Profiler Plus® samples amplified using serially diluted DNA templates (22, 11, 5.5, 2.75, 1.38, 0.69, 0.34, 0.17, 0.08, 0.043, 0.021, 0.011 and 0.0054 ng). STR alleles were successfully and completely called from the 0.17 ng sample, a somewhat higher sensitivity than found for the ABI 310. We have assessed the capabilities of the □CAE device to type real-world forensic DNA samples by typing 17 samples from case evidence previously processed and analyzed by the Palm Beach County Sheriff's Office. The DNA samples were extracted from a variety of common sources encountered in forensic analysis, including saliva, semen, single and mixed blood stains from sexual assault, paternity, burglary, armed robbery as well as homicide cases. The DNA profile results generated on the □CAE system were completely consistent with previous results.

Our demonstration that high quality STR separations can be effectively performed on the 96-lane microfabricated capillary array electrophoresis system using commercial reagent kits, establishes the feasibility of using this apparatus for high- throughput forensic identification. This accomplishment also provides a platform for the development of fully integrated automated low-volume sample preparation systems to improve reliability and throughput of forensic typing at lower cost.

#### References

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