PORTABLE MICROCHIP GENETIC ANALYZER FOR "REAL TIME" FORENSIC DNA ANALYSIS

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Microfabrication technology offers great potential for the integration of all the steps of forensic DNA analysis on a single microdevice. This integration should enable rapid, low-volume and low-cost PCR sample preparation[1], sample purification[2], and fast capillary electrophoresis (CE) separation.[3] We have previously demonstrated 4-plex mini-Y STR typing on a fully integrated PCR-CE microdevice instrument in the lab.[4] STR typing using such a fully integrated microsystem can also be performed outside of the laboratory producing rapid and reliable DNA analysis. Such advances should enable the possibility of conducting STR typing at a crime scene or other relevant point-of-analysis to help identify subjects of interest.

As a first step towards making portable STR typing possible, a microdevice has been developed that includes: (1) a 160-nL PCR reactor with a microfabricated heater and a resistance temperature detector (RTD) for precise temperature monitoring, (2) pneumatic valves for fluidic manipulation, (3) a micro co-injector for internal sizing standard injection, and (4) a 7-cm long microchannel for CE separation. The amplified STR products are first electrokinetically injected from the PCR reactor towards the CE channel simultaneously with a sizing standard from the co-injector. The microdevice is run on a portable instrument that contains 4-color laser fluorescence detection and all the necessary electronic, pneumatic/valve and optical components for chip operation.[4] To evaluate this portable microsystem for crime-scene investigation, a 9-plex autosomal STR typing system has been constructed using primer sequences employed in PowerPlex® 16. It consists of Amelogenin, and eight STR loci (D3S1358, THO1, D21S11, D5S818, D13S317, D7S820, vWA, and D8S1179) with a size range of 106–258 bp. Reproducible STR profiles for 9947A female and 9948 male standard DNA can be obtained successfully in 2.5 hrs using this PCR-CE microdevice with a standard deviation in size < 0.8 bp (n=3). The minimal input of DNA required for a complete DNA profile has been determined to be 100 copies by typing 0, 10, 20, 50 and 100 copies of 9947A standard DNA. To evaluate the ability of our system to analyze samples from commonly encountered stain sources in forensic investigations, we have successfully typed DNA samples extracted from liquid blood and swabs by the Palm Beach County Sheriff's Office (PBSO) using various commercial DNA extraction methods (DNA-IQ™, QIAamp™, and Ultraclean™). All the extraction methods yielded DNA samples with reasonable quality and quantity for full STR profiles on our PCR-CE microdevice. We are now exploring the feasibility of using our portable genetic analyzer to perform real time DNA analysis at a mock crime scene in collaboration with PBSO.

This integrated PCR-CE microsystem presents some interesting issues for forensic policy and practice. It could be used to aid crime scene investigators by providing initial typing information at the scene that is used to direct sampling choices. The pending field trials of this device at a mock crime scene will explore the concept of on-site DNA typing for aiding crime scene sampling or for early identification of subjects of interest. In addition, the ability to rapidly type an arrestee before release on bail might be useful for generating probable cause to hold the individual because of hits on other earlier criminal evidence.

References

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