

A131 Advanced Integrated and Portable Microfluidic Systems for Fully-Automated STR Analysis

James P. Landers, PhD*, University of Virginia, Department of Chemistry, McCormick Road, Charlottesville, VA 22904; Jessica V. Norris, PhD, Brian Root, PhD, and Orion Scott, PhD, MicroLab Diagnostics, 705 D Dale Avenue, Charlottesville, VA 22903; Annelise Barron, PhD, Stanford University, Department of Bioengineering, Palo Alto, CA; Abby Mackness, JD, Lockheed Martin, 9221 Corporate Boulevard, Rockville, MD 22407; and Joan M. Bienvenue, PhD, Lockheed Martin, 8801 General Griffins Court, Fredericksburg, VA 22407

After attending this presentation, attendees will have a complete understanding of the current state of development of automated, microfluidic systems for human identification. It will also highlight the advantages of microscale analytical systems for rapid generation of STR profiles from buccal swab samples.

This presentation will impact the forensic science community by illustrating next-generation DNA typing technology that will revolutionize how genetic analysis is performed both in the field and in the laboratory.

STR typing has become the accepted gold standard for human identification over the past two decades, and is now successfully employed in paternity testing, criminal casework, and missing person cases, as well as for databasing efforts. Although highly successful and reliable, current methodologies require 8-10 hours to complete under routine conditions, use large sample volumes, costly reagents, and are labor-intensive. Additionally, samples are open to the environment at multiple points during processing, making them susceptible to contamination. A translation of these sample processing and analytical methods to the microscale format will permit automation, miniaturization, and integration that will provide the end user with a system that provides expedited, cost-effective analysis in a closed system that reduces sample handling and possible contamination.

A variety of fully-integrated microfluidic biochemical analysis systems have been demonstrated recently for a variety of applications (e.g., Easley *et al.*).¹ Although integrating purification, PCR amplification, and electrophoretic separation/detection has been successfully demonstrated for pathogen detection, human identification using STR typing poses a number of new challenges for integrated systems, including: efficient miniaturized DNA purification, PCR amplification of the required thirteen core STR targets with commercial multiplexed kits, fine-tuning the use of commercial kits optimized for large volume amplification (25 µL) to function effectively at the microscale and, finally, rapidly separating the amplified target fragments with single base resolution and detection of 5-color fluorescence.

A system capable of the simultaneous and fully-automated processing and analysis of STR loci directly from buccal swab samples will be presented. Utilizing a single, integrated and disposable microfluidic chip, the multi-step sample processing and analysis that consumes 8-10 hours for conventional forensic STR analysis, can be carried out in less than forty five minutes. Exploiting novel DNA purification technology, DNA is purified from crude sample in less than fifteen minutes and guided into a chamber for a complex, STR PCR amplification using IR-mediated thermocycling. The PCR process, alone requiring ~3 hrs with conventional thermocycling, can be completed in less than twenty five minutes due to the excellent thermal properties of the microchip and the use of IR as a heat source, with efficient

amplification of all 16 STR loci in sub-microliter volumes. Separation is carried out using electrophoresis in a short channel (6 cm), using an optimized polymer, with baseline resolution and with 5-color detection based on acousto-optic filtering. Seamless integration of these methods on a single disposable microdevice provide a means long sought after in the forensic community for performing sample processing and fluidic manipulation entirely in sub-microliter volumes regime.

This presentation will provide a glimpse at the role that microfluidics will play in the future of forensic DNA analysis. The design and function of the integrated instrument capable of accepting the microfluidic device will be detailed, with data supporting the capability of the microfludic system for rapid, automated, end-to-end genetic analysis for human identification.

Reference:

Easley, C. J., Karlinsey, J. M., Bienvenue, J. M., Legendre, L. A., Roper, M. G., Feldman, S. H., Hughes, M. A., Hewlett, E. L., Merkel, T. J. Ferrance, J. P. Landers, J. P. Proceedings of the National Academy of Sciences of the United States of America 2006,

103, 19272-19277.

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