B144 Human Identification in Less Than 45 Minutes: A Rapid and Fully Portable DNA Solution

Nani M. Grimmer, BS, The Bode Technology Group, Inc, 10430 Furnace Road, Ste 107, Lorton, VA 22079; Michael N. Parsons, MS, 10430 Furnace Road, Ste 107, Lorton, VA 22079; Abigail S. Bathrick, MFS, 10430 Furnace Drive, Lorton, VA 22079; Katie Kennedy, BS, The Bode Technology Group, Inc, 10430 Furnace Road, Ste 107, Lorton, VA 22079; and Donia Slack, 10430 Furnace Road, Ste 107, Lorton, VA 22079*

After attending this presentation, attendees will be aware of an ongoing effort to develop a human DNA identification system that provides a discriminatory power of more than 98% and is performed on a commercially available, rapid, portable, and ruggedized instrument. Attendees will gain insight into the research and development of the system and the flexibility of using Single Nucleotide Polymorphisms (SNPs) for DNA identification.

This presentation will impact the forensic science community by introducing a system that can be used to develop investigative leads through the analysis of biological evidence on-site at crime scenes or areas of interests. The presented system will also support the applicability of SNPs to forensic DNA technologies and human identification.

In recent years, major progress has been made toward the development of fully integrated rapid DNA analysis devices; however, most of these instruments are not amenable to on-site DNA processing at the crime scene due to their size. Overall, the existing rapid DNA devices are large and mobile laboratories would be necessary to transport the systems to a crime scene. To address the need for a portable rapid DNA analysis system, The Bode Technology™ Group, Inc. has developed a human identification SNP assay for use with the RAZOR® EX instrument. The RAZOR® EX is a lightweight real-time thermal cycler typically used for the detection and identification of pathogens and biothreat agents. Commercially available RAZOR® EX assays are performed in under an hour using a rapid extraction method and a specialized pouch containing lyophilized amplification reagents. Additionally, the device has an on-board Liquid Crystal Display (LCD) screen that displays the assay results in real time.

To adapt the RAZOR® EX instrument for human DNA analysis, a multiphased approach was taken for development of a human identification system: (1) a simple, rapid, and efficient extraction method was created that is capable of lysing blood, semen, and epithelial cells in less than 15 seconds; (2) a customized TaqMan® allelic discrimination assay capable of amplifying forensic samples was developed using SNPs that were previously characterized for forensic use; (3) two amplification controls were designed to estimate the quantity and quality of the forensic sample; and, (4) the amplification reagents were lyophilized to ensure the stability of the assay at room temperature. While the assay results can be easily interpreted from the RAZOR® EX's LCD screen, a software application was also developed to provide more accurate data analysis and to allow population statistics to be applied to the sample. The entire process, from collection to result, has been performed on mock evidentiary samples in less than 45 minutes.

The current version of the rapid portable human identification system has generated robust results both in the laboratory and in mock field testing. The discriminatory SNP assays have been tested on over 400 forensically relevant samples. Prior to lyophilization, initial wet assay testing examined 25µL of blood (neat and diluted 1:50), semen (neat and diluted 1:50), and saliva (neat and diluted 1:10) deposited on glass, plastic, rusted metal, adobe brick, red brick, concrete, ceiling tile, MDF, duct tape, plywood, cotton fabric, denim, and paper. Full SNP profiles were generated from 100%, 86.5%, and 43.6% of the neat blood, semen, and saliva samples, respectively. As expected, fewer full profiles were generated from the diluted fluid samples, with a 30.8% success rate for diluted blood, a 23.1% success rate for diluted semen, and a 28.2% success rate for diluted saliva. Across all sample types, assay success rates were higher for samples deposited on non-porous substrates. Following lyophilization of the assay, 27 mock forensic samples were amplified with the lyophilized pouches on-site at mock crime scenes. The mock forensic samples consisted of blood, semen, and saliva (100µL) deposited onto metal, concrete, plastic, and cotton substrates that were exposed to uncontrolled indoor or outdoor conditions for a minimum of 12 months. Of these samples, 67% generated full SNP profiles. Additional controlled laboratory testing has been performed to examine the reproducibility and sensitivity of the lyophilized assay. A full developmental validation is anticipated to determine the system limitations.

Rapid, Portable, SNPs

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