A162 Validation of a New Robotic System to Automate High Throughput mtDNA Sequencing in a Forensic Laboratory

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After attending this presentation, attendees will understand the benefits and challenges encountered when validating a new robotic platform in a forensic laboratory for the mtDNA sequencing of high-quality reference samples.

This presentation will impact the forensic science community by providing information on how a robotic instrument can be successfully introduced into the laboratory and demonstrate how it can improve the high throughput processing workflow within the laboratory.

Currently, a number of forensic laboratories, including the Armed Forces DNA Identification Laboratory (AFDIL), are working toward developing novel automated solutions that allow cost effective, reproducible, and error-free processing of a high quantity of samples. Each year, the mitochondrial DNA (mtDNA) section receives and processes approximately 3,000 buccal swabs from maternal family members that are used for comparison to unknown samples from past U.S. military conflicts. Applying a high throughput approach toward reference processing is desired for increased efficiency and accuracy. For the last 12 years, AFDIL has used the Tecan Genesis® Workstation 200 to semi-automate all post-amplification mtDNA control region sequencing steps. This process, although accurate, required some user intervention due to a lack of fully integrated peripherals. In December of 2011, Tecan stopped supporting the older Genesis platforms. In preparation for this loss, AFDIL began the process to procure a new liquid handling instrument or face having to manually process family reference samples (FRS). It was vital that the new system continue to provide the necessary robotic capabilities while also incorporating enhancements that could further improve the workflow, increase overall sample throughput, and decrease user interactions.

Research and planning resulted in AFDIL acquiring and customizing the Hamilton's MICROLAB® STARplus liquid handling platform to allow for increased sample throughput and reduced user interactions during the automated mtDNA sequencing of FRS. The system includes eight independent pipetting channels, a 96-probe head, and two different gripper tools, as well as an external arm to integrate a plate sealer and eight thermal cyclers. Method development was an intensive process that included definition of labware, liquid class optimization, and the creation of a user-friendly workflow that allows parameters such as the number of samples or sequencing primers used to be easily changed. This flexibility ensures that the method can be utilized in a wide range of circumstances and by multiple sections at AFDIL without revisions. During method development, if any aspect of the automated post-amplification processing was unacceptable, the necessary modifications were made to the method or materials prior to the start of the validation. The development of a robust and reliable automated method was imperative to ensure efficient and accurate sample processing while meeting throughput requirements.

After five months of method development and optimization, an internal validation was performed to demonstrate that the Hamilton STARplus would produce sequencing products that were comparable to the results obtained when processed manually or on the Tecan. The validation included three experiments which were designed to meet applicable Quality Assurance Standards guidelines. Once the validation was complete, AFDIL's laboratory information management system and Standard Operating Procedures were updated for the integration of the new Hamilton robotic workflow. This post-amplification robotic system will enhance the high-throughput processing capabilities of reference samples and allow AFDIL to process at least 400 FRS per month, equating to over 6,000 sequencing reactions covering a 1,200-base pair region of the mtDNA genome. The implementation of the customized Hamilton STARPlus system into casework will decrease both processing time and required user interactions, greatly improving AFDIL's ability to process reference samples in real-time and ensure that the most up-to-date database is available for comparisons in missing persons cases.

The views expressed in this abstract are those of the authors and do not reflect the official policy of the Department of the Army, the Department of Defense, or the U.S. Government.

Mitochondrial DNA, Automation, Validation

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