## B142 Development of a Portable, Laminated Dynamic Solid-Phase DNA Extraction Method on a Rotationally-Driven Platform

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After attending this presentation, attendees will better understand the growing field of microfluidics and the design and development toward a novel, inexpensive, and portable alternative for extracting DNA from forensically relevant samples.

This presentation will impact the forensic science community by introducing a technique that adapts DNA extraction to a microfluidic platform for the potential of an inexpensive, on-site DNA extraction system in resource-limited areas with increased sample efficiency compared to current benchtop methods.

Solid-Phase Extraction (SPE), a critical step in genetic analysis, relies on the binding of DNA to silica-coated particles.[1] Dynamic SPE (dSPE) manipulates silica-coated magnetic particles during the extraction process to ensure optimized trapping and elution of DNA. Adapting dSPE to a microdevice enables advantages over benchtop methods including: increased sample efficiency, decreased reagent volumes, decreased assay cost, a closed system to prevent contamination, and the ability for integration with downstream processing via STR or next generation sequencing.

Polyester (Pe) can be patterned and bonded with Toner (T) to create multilayer microfluidic PeT devices for <$2 in <15 minutes.[2,3] Successful demonstration of dSPE via pressure-driven flow in rapidly fabricated PeT devices has been shown via manual pipetting.[2] Recent developments with open architecture PeT devices, hydrophobic valving, and centrifugally-driven fluid flow provide an attractive new platform for automated dSPE. The proposed dSPE system functions at low spin speeds (<1,500rpm), incorporates valving for non-aqueous solutions, is low cost ($0.25 per assay), and is amenable to rapid, simple fabrication. For the first time, DNA extraction from whole blood on a disposable plastic PeT microdevice using an automated, rotationally-driven platform is described.

The proposed PeT microdevice is composed of four layers and is fabricated using laser printer lithography. The device, when loaded with dSPE reagents and sample, can be run through a five-speed bidirectional spin program (0 to ~1,276rpm) on the home-built system. Binding of sample DNA to the particles is driven by an Alternating Magnetic Field (AMF). A novel combination of a hydrophobic valve and backpressure prevents the wash and elution buffers from entering the bound DNA chamber. After binding, IPA is released by centrifugal force (overcoming backpressure) at ~293rpm, followed by a TE wash through hydrophobic valve bursts at ~340rpm. After washing, the bound DNA is eluted and mobilized to a separate elution chamber at ~1,276rpm using "stop" valves positioned below the main DNA chamber. Approximately 10µl of purified DNA results from this process and is accessible by puncturing the Poly(DiMethyl) Siloxane (PDMS) covering the elution chamber. To demonstrate that the spin system effectively purifies PCR-ready DNA from 2µL of whole blood, the target β-globin was successfully amplified via microchip electrophoresis. In addition, samples extracted from FTA® cards were successfully amplified for STR analysis and were shown to display full profiles.

The proposed PeT microdevice has the potential to extract forensically-relevant samples when compared to the Qiagen® EZ1® instrument. Preliminary results suggest that DNA extracted from the microdevice and the EZ1® from buccal swabs were nearly equivalent in concentration, 0.28±0.1ng/µL and 0.27±0.7ng/µL, respectively. Furthermore, full STR profiles have been obtained with both methods. PeT-based peak heights were 1,619±1,054 Relative Fluorescence Units (RFUs) compared to the EZ1® peak heights at 1,774±705 RFU. Current efforts are underway to increase extraction efficiency and reproducibility. Overall, this is the first demonstration of a PeT microdevice used for chaotrope-driven dSPE on a rotationally-driven platform. The portable system has the potential to provide a cost-effective alternative to current SPE kits and once fully optimized, can be coupled to other pre- and post-processing DNA steps including preliminary DNA detection and amplification.

The opinions or assertions contained herein are the private views of the authors and are not to be construed as official or as reflecting the views of the Department of the Army or the Department of Defense.

**References:**

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**Microfluidics, Extraction, DNA**