

B77 Battling the Backlog: A Novel Bioanalytical System for the Separation and Collection of Intact Spermatozoa

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Learning Overview: The goal of this presentation is to demonstrate a novel technology that addresses the bottleneck separation of male from female DNA in rape kits for potential application to the forensic analysis of sexual assault evidence.

Impact on the Forensic Science Community: This presentation will impact the forensic science community by introducing a novel method utilizing a Capillary Zone Electrophoresis (CZE) system for the separation of intact cells collected from rape kits. The results presented will invite a meaningful dialog on the optimization and potential implementation of this technology that would significantly improve rape kit analysis speed and efficiency.

The national backlog in sexual assault cases is estimated to be between tens of thousands to half a million untested rape kits. The primary challenge crime labs face in analyzing these kits is the separation of purified male DNA from the mixture of primarily female DNA from gynecological swabs.¹ Standard protocols use Differential Extraction (DE), a manual separation technique that can take upwards of 12 hours to complete. The procedure incubates collected samples in detergents of increasing strength with varying time and temperature environments to differentially lyse fragile epithelial cells prior to stronger sperm cells. The final result contains, at best, a primarily male mixture of DNA that requires a trained analyst in Short Tandem Repeat (STR) mixture interpretation for perpetrator identification.^{2,3}

This study addresses the bottleneck challenge of DNA separation in rape kit processing through the use of a novel CZE system.⁴ CZE is a promising tool to perform the cell separation and has three major advantages over alternative technologies: a small amount of sample is consumed, which allows for replicate analyses of limited available evidence; rapid separation time compared to standard methods; and single cell detection and collection when interfaced with an automated fraction collector developed in-house. An electrokinetic injection of a simulated sexual assault sample is separated at a high voltage across a capillary with an inner diameter of 100 μ m. The CZE instrument is coupled with an automated fraction collector that deposits samples eluted from the distal end of the capillary into individual wells on a microtiter plate corresponding to a distinct migration time interval.

Quantitative Polymerase Chain Reaction (PCR) amplification of a Y-chromosome sequence is used to confirm the separation and collection of male DNA from sperm cells in a single well of the microtiter plate in fewer than 15 minutes, representing a significant improvement in separation time compared to current methods.⁵ Furthermore, the system was designed to integrate seamlessly within the current work flow of a standard crime laboratory to increase its potential for adoption and implementation in battling the rape kit backlog.

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Sexual Assault, Electrophoresis, Automation