B11 Absolute Quantitation of Semen-Specific Biomarkers From Post-Coital Samples

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After attending this presentation, attendees will better understand the use of a multiplex tandem mass spectrometry assay for the detection of seminal fluid protein biomarkers that will be of benefit in the determination of the post-coital interval for semen detection.

This presentation will impact the forensic science community by providing results from a quantitative study in an area of forensic science that is under-researched. The results of this study have the potential to aid in determining the post-coital interval, which can further assist sexual assault forensic examiners in the timely collection and processing of sexual assault evidence.

Sexual assault is a prevalent crime in today's society; however, the forensic testing utilized for sexual assault evidence is costly and time consuming, resulting in a backlog of untested evidence. The current enzyme-activated and antibody immunochromatographic tests used by forensic examiners for the identification of seminal fluid only provide presumptive results. This is due to false positives with non-biological material, cross-reactivity, and positive results with non-target fluids. Immunochromatographic tests rely on the tertiary structure of proteins in order to produce positive results; however, proteins may be subjected to undesirable conditions resulting in the unfolding of the tertiary structure, preventing enzyme or antibody interaction. A negative serological test may prevent the sample from receiving additional DNA testing, regardless of whether the protein was present in a degraded form. The presumptive nature of these tests has deterred scientists from reliably determining a post-coital interval. The development of a confirmatory technique for the identification of seminal fluid would allow for the post-coital detection of semen to be determined and would assist in the analysis of sexual assault evidence.

Prior experiments have identified, developed, and validated a qualitative, mass spectrometry-based assay for the confirmatory identification for human seminal fluid. A previously performed sensitivity study has shown that seminal fluid biomarkers (prostatic acid phosphatase, prostate specific antigen, and semenogelin) are readily detectable by mass spectrometry from a little as 1nL of semen, levels at which the currently employed presumptive tests begin to fail.

The goal of the current research was to validate a quantitative variation of the previously developed method in order to quantitate these semen-specific proteins in post-coital samples. The current method employs scheduled multiple reaction monitoring on an Agilent® 6430 triple quadrupole mass spectrometer coupled with a nanoflow chip cube High-Performance Liquid Chromatography (HPLC). Using synthetic peptide standards, a linear calibration model was developed and mock casework samples were used to analyze the multiplex assay's sensitivity and limits of detection in addition to a comparison of the specificity and selectivity of this approach to immunochromatographic assays.

In conclusion, this study provides evidence that mass spectrometry produces more sensitive results for the detection of seminal fluid, providing a more reliable method for the detection of semen in sexual assault samples. Furthermore, the use of mass spectrometry has the potential to enhance the serological screening process and aid in the determination of the post-coital interval.

Forensic Science, Proteomics, Seminal Fluid