B96 Novel Techniques for Identifying the Semen Donor in Extended Interval Post-Coital Samples

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The goal of this presentation is to aid caseworkers in understanding how to best recover the male profile of a post-coital sample, thus allowing attendees to develop semen donor DNA profiles from samples recovered >72 hours after intercourse.

This presentation will impact the forensic community and/or humanity by informing the forensic science community of various novel strategies to recover DNA profiles from extended interval post-coital samples using Y-STR technology. Moreover, detecting a Y-STR male profile with samples collected greater than 72 hours after intercourse is of value to forensic casework because in most instances it is unlikely that an autosomal STR profile of the semen donor would be obtainable with vaginal samples taken then.

In forensic casework some victims of sexual assault wait to provide vaginal samples more than 36 hours after the incident. In these cases the ability to obtain sperm diminishes as the post-coital interval is extended. Absence of significant numbers of spermatozoa in such specimens can be due to the victim's behavior after the assault (showering, douching, using spermicidal agents, and vaginal vault drainage), menstruation, and vaginal inflammation. Previous data demonstrated that it is possible to obtain Y-STR profiles from post-coital samples recovered up to 72 hours after intercourse regardless of external ejaculation or microscopic detection of sperm. In this work, the post-coital range of time points sampled was extended to include 72 hours through 168 hours and typed the extracted DNA using the both the Applied Biosystems AmpF*l*STR® Yfiler™ PCR Amplification Kit and Promega PowerPlex® – Y System.

Results indicated that complete Y-STR profiles were unable to be obtained after 72 hours post-coitus using a standard organic extraction technique without differential lysis and the input of 300 ng of total DNA. However complete Y-STR profiles were obtained at 96 and 120 hours post-coitus using a differential extraction method. To further extend the post-coital interval, several additional strategies were employed. Post amplification clean up was introduced which not only increases the peak height of observed alleles but permits the detection of alleles that were initially below the levels of detection. Indeed the use of post-amplification clean up produced full profiles where previously partial profiles were observed. Another strategy employed was to use enzyme cocktails incorporating proof reading enzymes to determine whether a further increase in detection sensitivity was possible.

A direct lysis of the swab contents was employed as an alternate method for post-coital analysis. Previous research in this laboratory has found that the direct lysis method is faster than standard methods and can produce reliable genotypes in samples recovered up to 96 hours after intercourse. Therefore, two modifications were made to the procedure to further extend the post-coital interval from which a semen donor profile could be obtained. The first was to clean up the extract before amplification and the second was to clean the direct lysis product after amplification. These clean up strategies remove contaminates that can interfere with downstream reactions, which in turn increases analytical sensitivity.

Post-coital samples in which one or more of these strategies have been employed dramatically improved the profile quality and increased the post-coital interval (>72 hours) in which sperm DNA can be recovered. Detecting a male profile greater than 48 hours after intercourse is of value to forensic casework because in most instances it is unlikely that an autosomal STR profile of the semen donor would be obtainable with vaginal samples taken then.

Extended Interval Post-Coital, Post Amplification Clean Up, Direct Lysis

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