B97 Using Microscopy to Predict Success or Failure of a Differential DNA Extraction

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Attendees will learn the results of a study examining the usefulness of microscopy in order to predict the success or failure of a differential DNA extraction.

This presentation will impact the forensic community and/or humanity by demonstrating that microscopic evaluation of sexual assault evidence is a valuable tool for determining whether or not the traditional differential DNA extraction method will be effective.

Differential DNA extraction is a commonly used method of examining sexual assault evidence in the forensic DNA laboratory. This extraction method allows the DNA analyst to chemically separate sperm donor DNA from epithelial cell donor DNA resulting in two fractions. A differential extraction failure is defined as incomplete removal of the epithelial cell portion of the sample from the sperm cell fraction resulting in a DNA mixture of the two donors or the presence of the epithelial cell donor only in the sperm fraction. A sperm cell fraction that results in a mixture of epithelial and sperm donor DNA can be problematic when performing statistical analysis of the sample; decreasing the weight of the DNA test results in court. In addition, sperm cell fractions resulting in mixtures can make productive searches of DNA databases, such as CODIS, more difficult.

Microscopic slides have been prepared from 325 vaginal swabs. These slides have been stained via the traditional Christmas tree staining procedure and examined microscopically. The number of epithelial cells and the number of sperm cells was rated for each slide. In addition, DNA from the vaginal swabs has been extracted using a traditional organic differential procedure. The sperm cell fractions of these extractions have been amplified using the AmpF*l*STR□ Profiler Plus□ and COfiler□ amplification kits and analyzed via the ABI Prism□ 310 Genetic Analyzer. The resulting sperm fraction DNA profiles were examined in conjunction with the microscopic data in order to determine the correlation, if any, between the microscopic data and the mixed or single source nature of the DNA profile results.

The findings of this study show that microscopic ratings indicating a large number of epithelial cells regardless of the number of sperm cells cannot predict a failure of the standard differential DNA extraction procedure. Likewise, microscopic ratings indicating a small number of sperm cells regardless of the number of epithelial cells also cannot predict a failure of the differential DNA extraction procedure. However, samples that contain both a small number of sperm cells and a large number of epithelial cells will result in a failure of the differential DNA extraction procedure in 70% of cases.

A modification of the standard organic differential method was utilized on a subset of 20 samples that originally resulted in a sperm fraction DNA mixture. However, adding an additional aliquot of Proteinase K during the epithelial cell digest and increasing the number of sperm cell washes only improved samples with microscopic ratings indicating larger numbers of sperm present. This modification did not increase the likelihood of obtaining a single source sperm fraction for samples with the characteristic microscopy (low sperm/high epithelial) indicating a mixed DNA profile is likely. Therefore, these results indicate that perhaps epithelial cell DNA is bound somehow to the sperm heads or is in some other way difficult to remove from the sperm fraction via the traditional procedures.

This study has demonstrated that microscopic evaluation of sexual assault evidence is a valuable tool for determining whether or not the traditional differential DNA extraction method will be effective. If an effective alternative differential DNA extraction method was available, samples identified via microscopic evaluation likely to result in sperm cell fraction mixtures using the traditional organic method could be diverted, thereby increasing the number of single source sperm fraction profiles.

DNA, Mixture, Microscopy

 ** Presenting Author*