

PRECISE ISOLATION OF SPERM CELLS FROM FFPE TISSUE SECTIONS SIMULATING A SEXUAL ASSAULT VICTIM

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Formalin-fixing and paraffin-embedding (FFPE) is a common practice to preserve tissues for pathological testing. DNA extracted from FFPE tissues may also provide alternative source of genetic material for forensic casework, such as solving paternity cases or identifying unknown embalmed human remains. In case of sexual assault victims, DNA can be extracted from FFPE tissue sections obtained from victim's vaginal epithelium, aiming to detect assailter profile. However, this often results in mixed or partial profiles, because of the low abundance of male component and its dilution in the more abundant female DNA.

DEPArray™ technology has been widely demonstrated to enable the isolation of pure cells in many fields, including forensic mixtures.

We report here preliminary data on the analysis of sexual assault-simulating FFPE sections, with DEPArray™ technology, for precise assailter genetic identification.

A slit was created within each of six samples of vaginal epithelium obtained at autopsy, where different quantities of semen (1-5-10-25-50-100 µl) were injected, then formalin fixed and paraffin embedded for histological analysis. From each block, two sections (50 µm each) were obtained, then dissociated and immunofluorescently labelled with anti-cytokeratin-FITC, to detect epithelial cells, and sperm-head antigen-APC for sperm cells.

With a single tube procedure, recovered cells were lysed and genotyped with PowerPlex Fusion 6C, followed by CE-separation and fragments analysis.

At DEPArray™ interface, sperm and epithelial cells were detected according to specific staining signals and to morphological criteria, such as cell size and shape; selected cells of each type were isolated in small pools of pure cells.

Genetic profiles were obtained from the isolated cells and matched with corresponding donors profile: the completeness of cells-derived profiles was proportional to the number of cells amplified: as an example for sperm cells, which are haploid, more than 20 alleles were obtained from as little as eight sperms.

For the first time we report the successful application of DEPArray™ technology to FFPE specimens for forensic purposes, demonstrating that pure genetic profiles can be generated from the different cell species isolated from FFPE sections.

This preliminary work suggests that FFPE on recent tissues obtained from sexual assault victims, could be successfully approached with the described method, in order to obtain consistent genetic profiles from the assailter.