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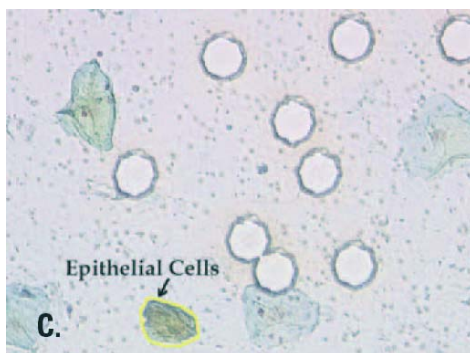
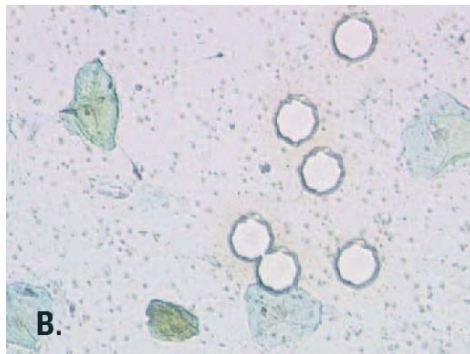
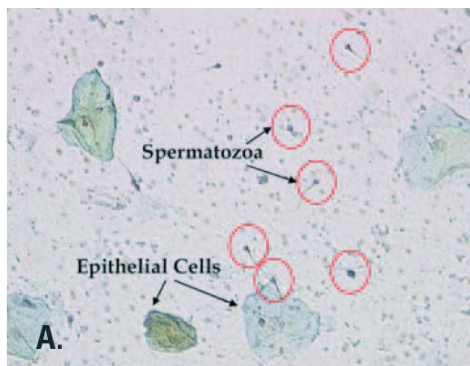
Leica Laser Microdissection  
Application Note

A vertical strip on the left side of the page contains three stacked microscopy images. The top image shows a dense field of green fluorescent spots. The middle image shows the same field with red outlines around individual spermatozoa. The bottom image shows a network of green fluorescent structures.

# reSOLUTION

Separation of  
Spermatozoa for  
Forensic Analysis

# Separation of Spermatozoa from Epithelial Cell Mixture for Forensic Analysis



Laser microdissection technology can be used to selectively separate specific cell types from mixtures of sexual assault evidence for forensic analysis. Sperm cells can be microdissected from sperm/epithelial cell mixture smears. DNA signals from female epithelial cells could then be clearly separated from those of male sperm.

The Leica LMD system provides an effective and quick method to separate sperm cells from a sperm/epithelial cell mixture. It facilitates low copy number analysis by precise dissection, minimizes sample manipulation, and reduces downstream analysis steps.

**Figure 1.** Images of spermatozoa and epithelial cell mixture before (A) and after (B) laser microdissection. Sperm cells (A and B) and female epithelial cells (C and D) were selected, microdissected, and collected individually for DNA analysis.

## Method:

Smears comprising of sperm and epithelial cell mixtures were prepared on membrane coated glass slides. Smears were stained with traditional "Christmas Tree" staining (Kernechtrot and Picroindigocarmine):

- Cells were heat affixed on the slide
- Covered with Kernechtrot solution for 20-30 minutes and then washed with water
- Covered with Picroindigocarmine for 15 seconds
- Washed with ethanol

Slides were observed in brightfield and fluorescence (fluorescence images not shown). All images were automatically captured during laser microdissection with the Leica LMD system and optional IM1000 imaging software. Sperm cells and epithelial cells were dissected respectively and collected into different PCR tubes.