Use of Automated Magnetic Levitation Technology in Sexual Assault Forensics

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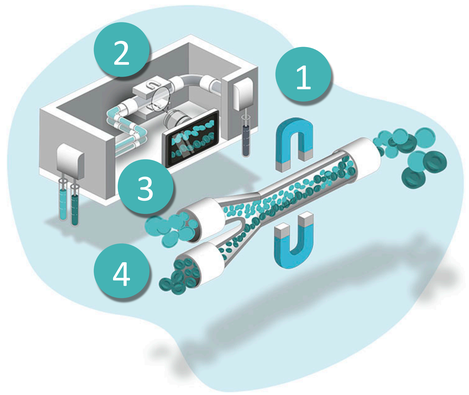
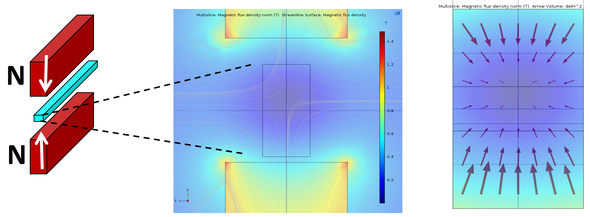
Abstract

Magnetic levitation is a technology that is capable of label free separation of cells based on their relative cell density. Here we demonstrate the separation of epithelial cells from sperm and blood from mock forensic swabs and cell mixtures, using this technology on our LeviCell Platform. This separation method has positive implications for rapid testing of sexual assault kits thereby addressing the DNA mixture problem. Exploiting the relative density difference between epithelial and non-epithelial cells, we were able to separate and collect epithelial cells from single swabs in less than 30min. We also demonstrated the compatibility of the collected cells for downstream PCR and STR analysis using Quantifiler and Innogenomics HY-Quant kits that are commonly used in forensic laboratories. Thus, our technology offers a simple, rapid, automated, and cost-effective platform for complete separation of epithelial cells from sperm to address the DNA backlog problem faced by Forensic Labs.

Technology Background



Step 1: A sample consisting of a mixture of cells, or cells to be analyzed, is introduced to the system through an inlet port.
Step 2: Levitas technology separates individual cells according to type or state based on the different levitation heights caused by the unique magnetic and density signatures (described in more detail below)
Step 3: The different cell types stabilize at different levitation heights. As the sample flows through the system, different cell types can be imaged, counted, and collected.
Step 4: By continuing to aspirate the sample, the levitated cells transition to a bifurcated tube that separates cells in the top chamber from the lower chamber.



Within the cross-section of the capillary, the resulting forces are directed inwards, pushing cells to the center of the flow cell and at a height determined by a combination of the density and magnetic properties of the objects in the flow cell.

Primary Advantages of Levitation

Simple & Efficient

SORT SMARTER, NOT HARDER

With Magnetic Levitation Technology, there are no complicated fluidics or preparation steps. Simply load the sample in and process it.

Undamaged Cells

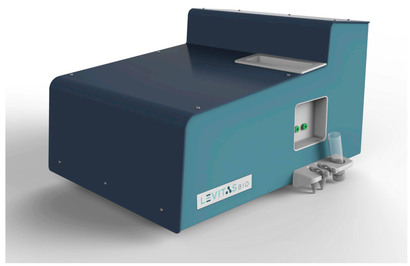
LIVE CELLS IN, LIVE CELLS OUT

Unlike all other methods, levitation is gentle on the cells and does not damage or injure them.

Unlabeled

SKIP THE MARKERS

Remove bias from cell analysis by eliminating labels and dyes. Catch cells based on what they are, not what marker they expressed.

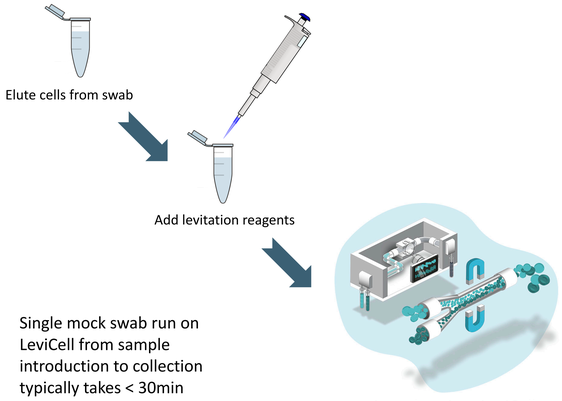


Applications of Levitation Tested for Forensics

- *Aged epithelial swabs (up to 10 years old)*
- *Epithelial (buccal) + blood cell isolation*
- *Epithelial cells (buccal and vaginal swabs)*
- *PBMC + Epithelial (buccal)*
- *Epithelial + sperm*
- *Epithelial + blood +sperm*

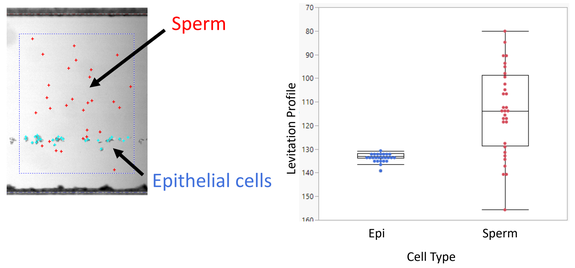
Same, simple workflow used for all sample types

Workflow for Forensics Samples



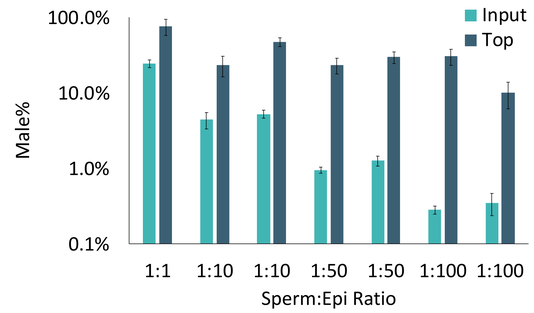
Single mock swab run on LeviCell from sample introduction to collection typically takes < 30min

Levitation of Epithelial Cells Compared to Sperm



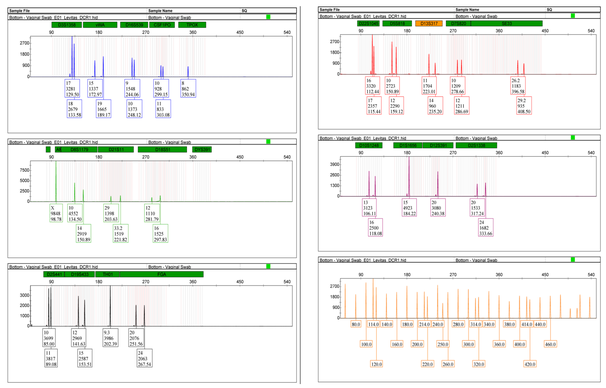
A mixture of sperm and epithelial cells eluted from swabs were introduced into the LeviCell. After ~3 min of equilibration, the epithelial cells formed a tight band, levitating near the bottom of the flowcell, while the sperm cells were dispersed throughout the flowcell. The formation of a tight band allows the user to pull the epithelial cells into their desired collection channel and enrich for sperm DNA.

Purification & Enrichment of Male DNA from Mixed Samples



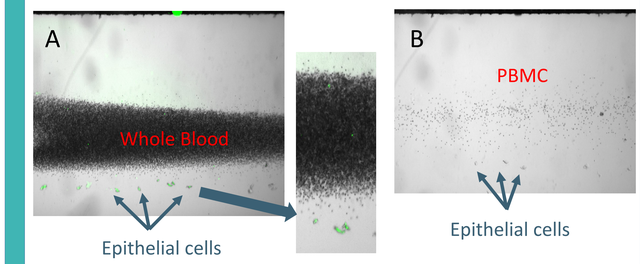
Mixtures of sperm and epithelial cells at different ratios, ranging from 1:1 to 1:100 (indicated on the x axis in this graph) were introduced into the LeviCell. The ratio of sperm to epithelial cells in the input as well as in the final, collected output channel were determined by qPCR using Y chromosome and autosomal targets. Male DNA was enriched in outputs for LeviCell processed samples.

Compatibility with STR Analysis



Outputs from LeviCell processed vaginal swab elute was subjected to STR analysis. The resulting outputs indicate that samples that have been processed through levitation are compatible with standard forensics analyses.

Separation of Epithelial Cells from Other Cell Types



Two additional examples that illustrate separation of epithelial cells from other cell types. (A) This panel shows the result of introducing a mixture of whole blood and epithelial cells into the flow cell. Similar to what was seen with sperm-epithelial cell separation, the epithelial cells form a distinct layer beneath the whole blood cells. (B) This panel shows the result of introducing a mixture of PBMC's and epithelial cells into the flowcell. Again, the epithelial cells form a distinct layer beneath the PBMC population.

Conclusions

Magnetic Levitation provides a simple and quick (< 30min) method to separate different cell types such as epithelial cells from sperm or blood based on cell density.

Magnetic Levitation based separated cells are compatible with downstream DNA analysis and forensic workflow.

Magnetic levitation offers a new way to address DNA mixture problem via upstream cell separation and can be used for SAK analysis.

Magnetic levitation offers

(A) Single instrument for a variety of sorting applications
(B) Simple one step-workflow with minimal hands-on time
(C) Alternate solution without use of expensive reagents or consumables
(D) Compatibility with small volumes and rare samples

References

Magnetic Levitation of Single Cells. Durmus et al, PNAS July 14, 2015 112(28)

Separation of sperm and epithelial cells in a microfabricated device: Potential application to forensic analysis of sexual assault evidence. Horsman et al, Anal Chem, 2005 Feb1; 77(3): 742-9

Acknowledgements

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