

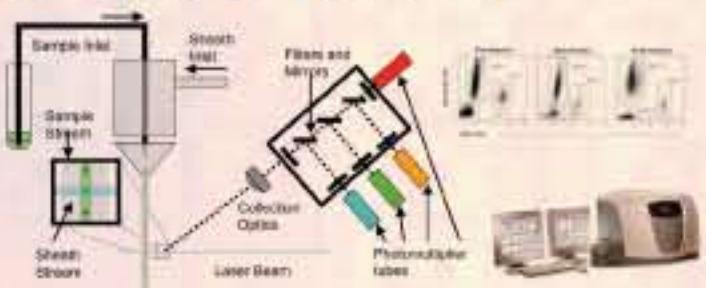
Miniature Flow Cytometer for Medical Diagnostics and Pathogen Detection



Sandia National Laboratories

Igal Brener (PI), Surendra Ravula, Karl Westlake, Darren Branch, Conrad James, Paul Clem,
Yun-Ju Lee, John Williams, Jennifer Sigman, Chris Bourdon, Steven Younghouse

Introduction to Flow Cytometry



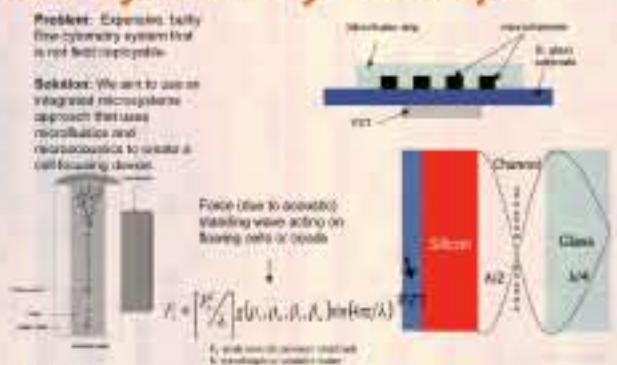
- An analysis tool for understanding & measuring biochemical and physical properties of cells, i.e., protein levels.
- Specific receptors or proteins are fluorescently labeled.
- Separate sheath flow focuses cells into a single-file line.
- Single or multiple laser beams excite fluorescence and/or scattering. Fluorescent intensity is then correlated with receptor levels or protein expression.

Why Miniature Cytometry at Sandia?

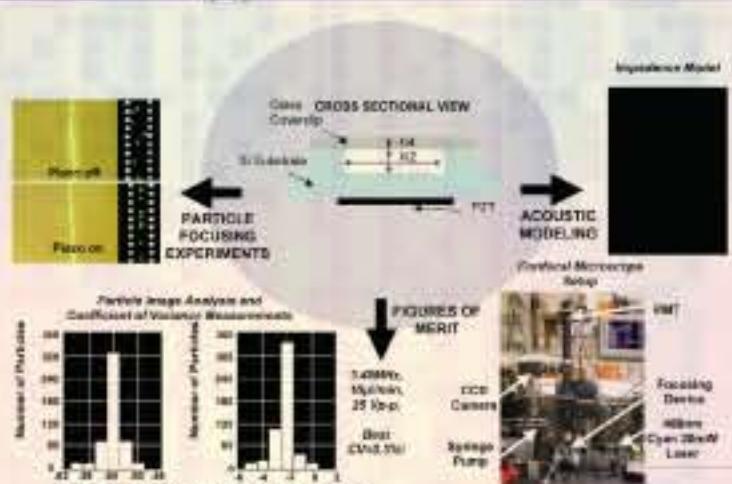
- National Security
 - It is the gold standard for microsphere-based assays.
 - Has been used for biothreat I.D. as the back-end detector in PCR-based devices.
- Clinical diagnostics in civilian and military clinics
 - It is the gold standard for infectious disease diagnostics.
 - Bio-attacks, AIDS, anemia, cancer, and many infectious diseases.
 - Assays can be multiplexed, saving time and sample size.
 - Credible replacement for ELISA.



Microacoustic Cytometry Concept



Overview of Approach and Results



Fabrication Paradigms for Microacoustic Focusing Platforms



Key Features

- Integration of acoustics with dielectrophoresis to achieve highly focused streams
- Use of glass-glass thermal bonding for optical access to channels

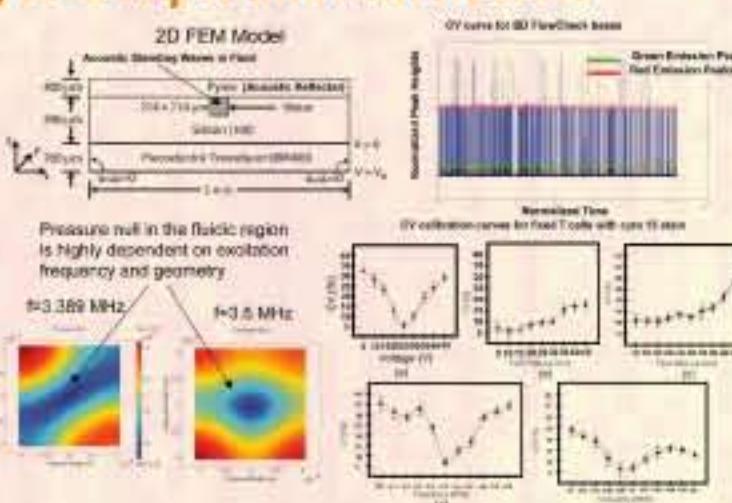
Key Features

- Particles focused hydrodynamically along the width and acoustically along the height of the microchannels
- Integration of an acoustic sorter along with the focusing
- Fabricated on a bulk piezo substrate and channels created out of a multilayer SU 8 photoresist process

Key Features

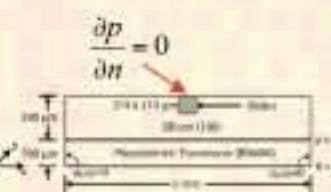
- Ease of fabrication
- 2-step Bosch etch process
- Anodic Bonding of glass reflector
- 2-D Focusing with a single PZT transducer

Modeling and Experimental Results



Impact of Reflector Boundary Conditions: Force on Microparticles

Perfect Reflector Case



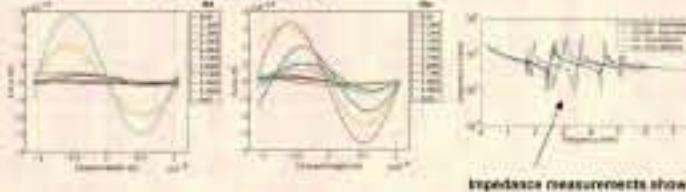
Perfect Reflector: To achieve the best possible acoustic reflection, this boundary condition requires that the normal derivative of the pressure is zero.

Pyrex Reflector Case



Pyrex Reflector: Conventional λ/4 reflectors may cause the normal derivative of the pressure to be non-zero. This causes a reduction in the acoustic reflection and hence standing wave field is weaker.

Perfect Reflector Case

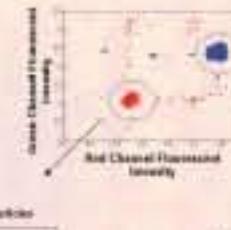


Impedance measurements show frequencies that are most efficient for coupling acoustic energy into the resonator

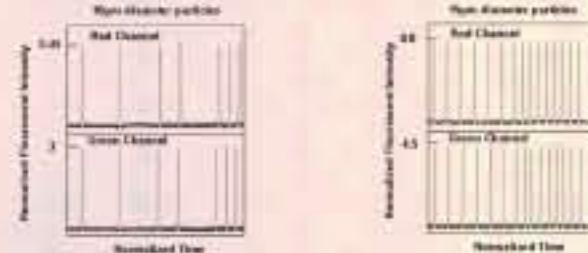
Pyrex Reflector Case



Force calculations suggest a strong node only at several frequencies



Experimental Results - Separation of Particles by Size



Summary and Conclusions

- Designed, fabricated, and characterized a microfluidic-acoustic focusing module for flow cytometry.
- Obtained good flow-focusing with microbeads and cells.
- Developed a detailed FEM modeling strategy to serve as a design and analysis tool
- Needs further optoelectronic integration for a complete cytometer-on-a-chip.

Partial List of References

- D. Hahn, R. C. L. Bremer, J. B. Griswold, and S. Gopalakrishnan, "Microfluidic flow cytometry: analysis of cells and particles," *Biological Measurement*, vol. 30, pp. 873-880, 2005.
- S. E. Nelson, K. Elson, G. J. Schatzki, and L. Brune, "Microfluidics and nanofluidics for medical flow cytometry," in *PITI Nanotechnology Conference Santa Clara, CA*, 2005.
- S. K. Jones, R. G. Sauer, R. L. Ulrich, L. M. Oliver, C. R. Jones, and L. Brune, "A microfluidic flow cytometry system for optical detection of cells and nanoparticles," in *Applied Optics Society Meeting Abstracts*, 2005.
- H. C. Courtney, L. L. Anderson, M. A. Anderson, C. M. Casner, and L. Brune, "A flow cytometry system using nanoparticle tracking for detection of cells and nanoparticles," *Optics Letters*, vol. 25, pp. 630-632, 2000.
- A. Hafezi and J. Shah, "Micro-manipulation of small particles for mode-purification of cold atomic clouds using manipulation of light and microreflections," *Optics Letters*, vol. 30, pp. 1247-1250, 2005.
- H. W. Hwang, M. H. Lee, S. Bae, Y. Choi, M. H. Park, J. J. Heo, and M. S. Cho, "A fiber-optic-coupled ultrasonic impinger," *Aerosol and Air Quality Research*, vol. 2, pp. 429-434, 2002.
- J. J. Heo and H. W. Hwang, "Fiber optic particle fluxes combining ultrasonic standing waves and Lorentz force," *Aerosol and Air Quality Research*, vol. 2, pp. 435-439, 2002.
- H. W. Hwang and J. J. Heo, "Ultrasonic acoustic particle fluxes for aerosol detection in high velocity," *Int. J. Environ. Res. Public Health*, vol. 1, pp. 111-117, 2004.
- S. K. Jones, D. H. Hahn, C. O. Jones, B. J. Townsend, M. H. Park, L. Brune, "A microfluidic focusing and selection system for optical microscopy and flow cytometry," *Journal of Microelectromechanical Systems*, vol. 15, pp. 775-778, 2006.
- H. W. Hwang, "On the focusing and trapping of small particles in an acoustical field in an ideal fluid," *Journal Physics Fluids*, vol. 18, pp. 775-778, 1996.