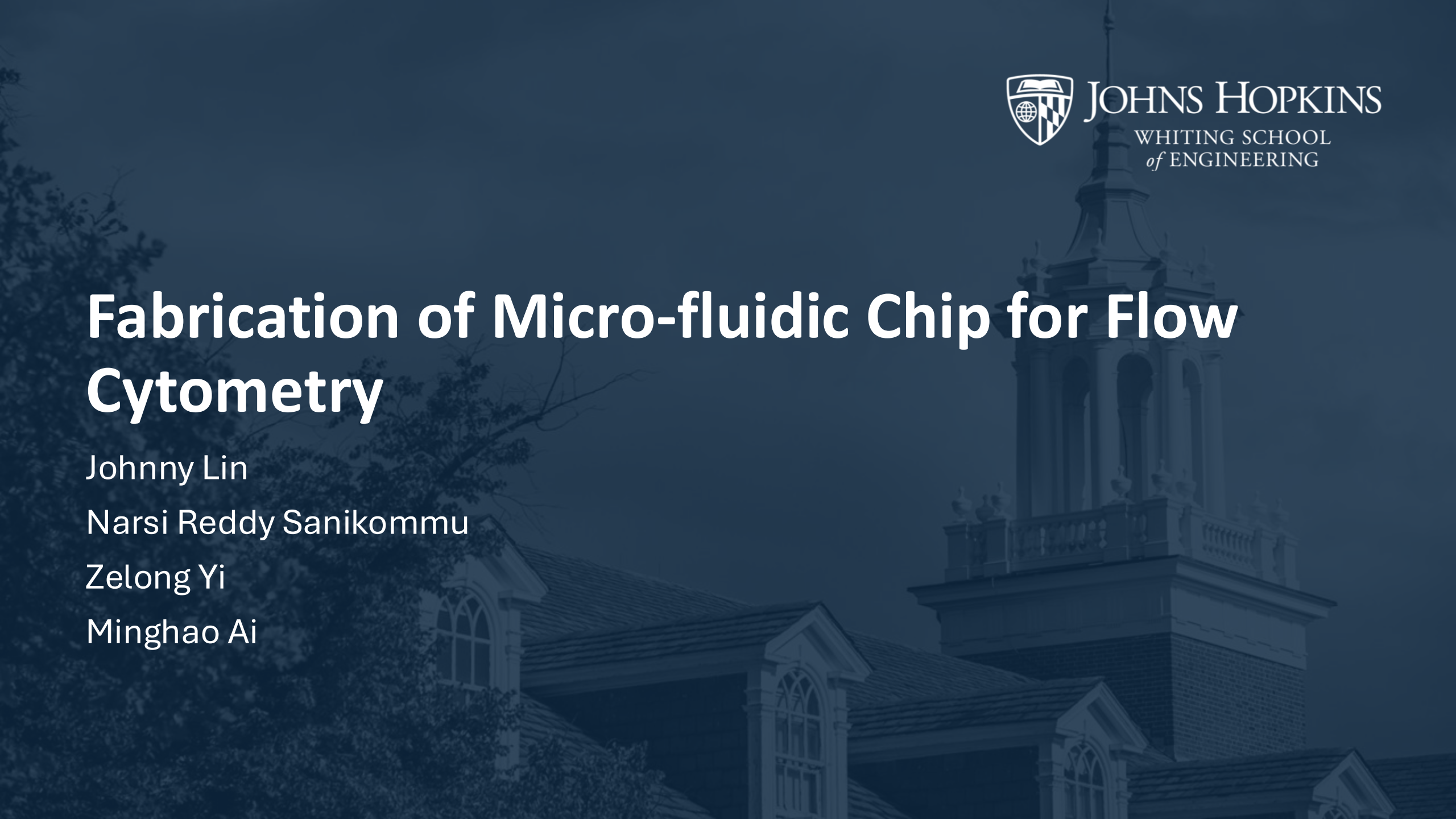

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Fabrication of Micro-fluidic Chip for Flow Cytometry

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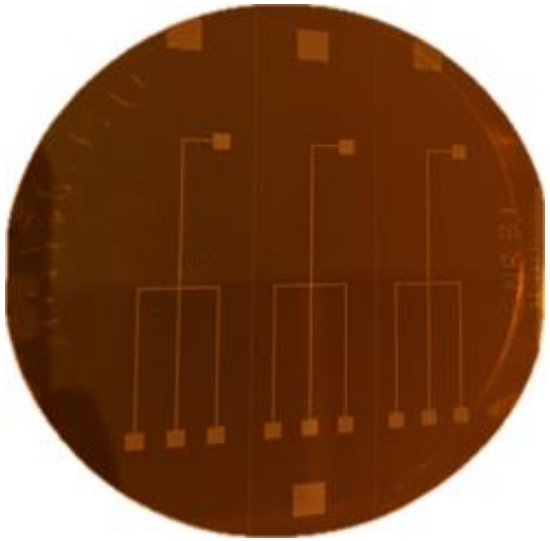
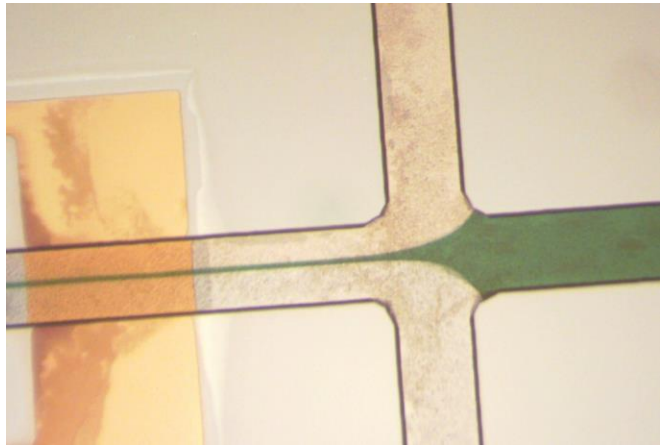
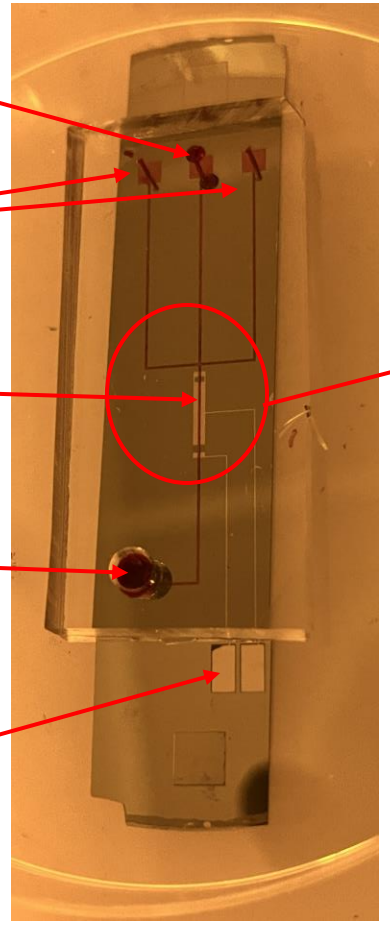
Introduction and Literature survey

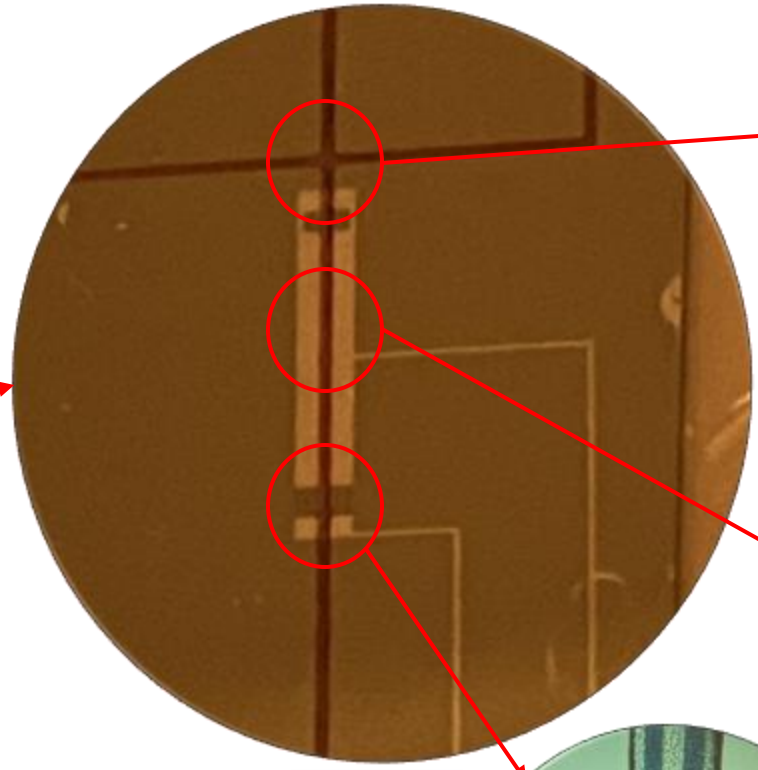
- **Flow cytometry** is a laser-based analytical technique used to measure and analyze multiple characteristics of single cells as they flow through a laser beam in a fluid stream.[1]
- A **Flow Cytometer on Chip**, also known as microfluidic flow cytometry, is a miniaturized version of conventional flow cytometers that integrates the functionalities of cell sorting and analysis into a compact, microfluidic device. This chip-based cytometry utilizes microfabricated channels and detection systems, significantly reducing the equipment's size, cost, and sample volume, while enhancing portability and integration for point-of-care diagnostics.
- Here, we are using flow focusing using buffer solution from the channels on the sides and further flow focusing using the electrokinetic principle.

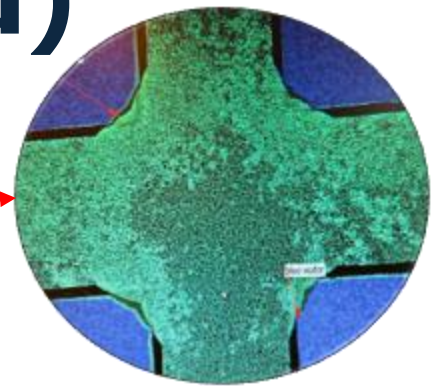




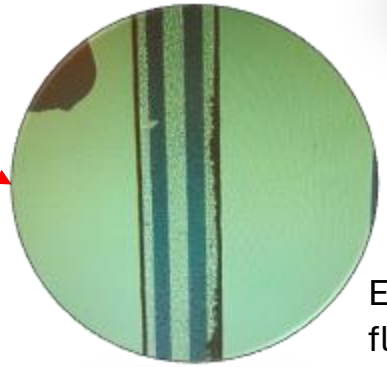
Features of the device (device related)



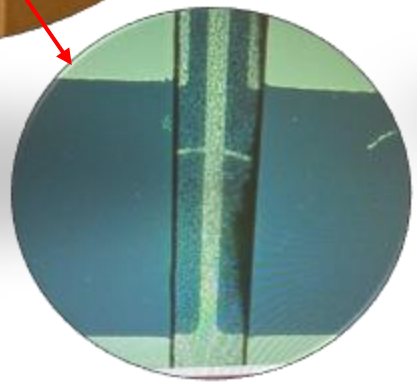




Hydrodynamic flow focusing junction

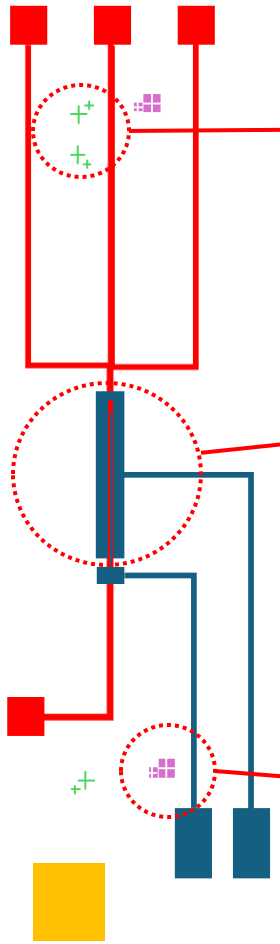


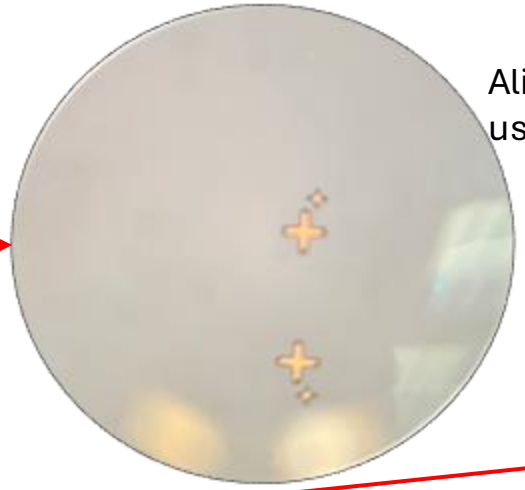
Electrodes in the flow channel



Layout of electrode that is inside flow channel

Features of the device (Fabrication Related)

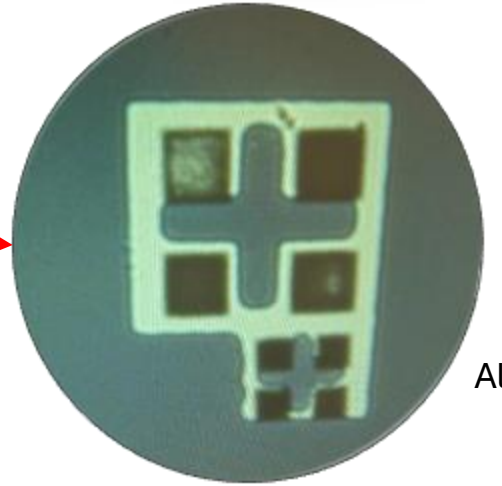




Alignment marks formed after Al etch, used for 3rd UV exposure

Layout of the electrode in the flow channel

This feature is used to measuring oxide thickness, metal thickness etc. during fabrication process

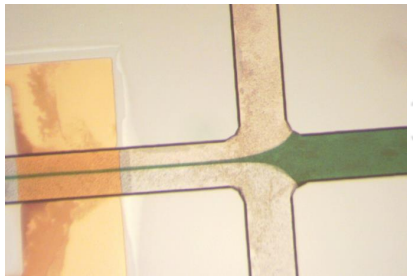


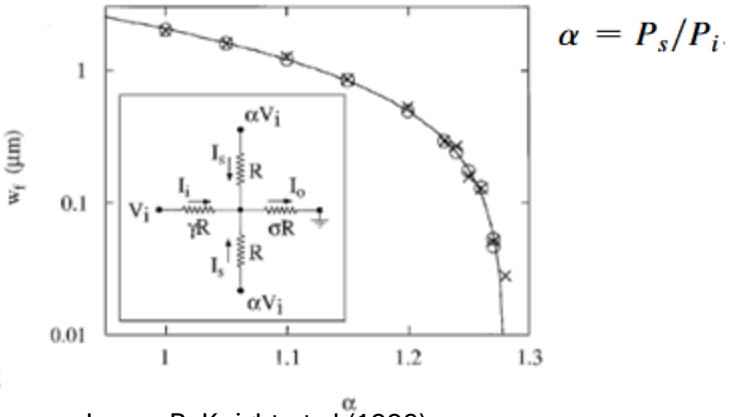
Alignment marks used for Al etch.

Mechanism: Flow focusing and Electro kinetics Focusing

Hydrodynamic flow focusing

- Flow in microfluidic channels is laminar based on Re and can be considered fluid circuits analogy to electric circuits.
- Channels CS is rectangular and hydraulic resistance can be calculated based on Brian Kirby[2]
- Another way to calculate the focussing width is by James B. Knight et al (1998)[3].
- Resistances of the side, inlet, and outlet arms of the circuit are R, γR, and σR, respectively, with the parameters γ and σ reflecting differences in channel geometry

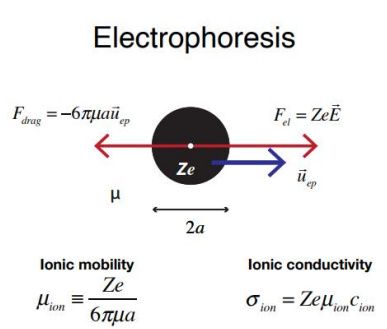


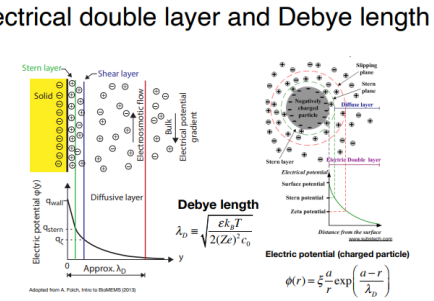


James B. Knight et al (1998)

Electroporation flow focusing

- Electrokinetics is the study of the motion of bulk fluids or selected particles embedded in the fluids when subjected to electric field dynamics [2].Electrophoresis (EP)
- describes the movement of charged particles in a liquid medium under an external electric field. Biological materials such as red blood cells (RBCs), DNA, and proteins carry charges.
- For example, red blood cells (RBCs) have a negative charge density of approximately 10 negative ionic groups per cell [4].





Professor Hur class, Brian Kirby[2]

Fabrication process

Oxidation of wafer

Spin coating of PR and patterning

Etching

Etching of sio2 from Si wafer

Patterning metal by Al etchant

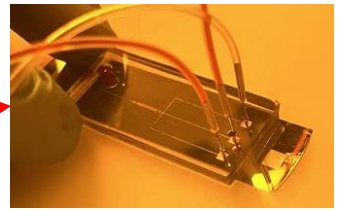
Oxidation of wafer

Thin film deposition of Al

Removal of oxide layer after electrode patterning

PDMS bonding with pre-punched holes

Insert catheter in holes



Anodic bonding with predrilled holes

Insert tubing in holes and cover with resin

Silicon

Initial SiO2 layer

SiO2 insulation layer

PR resist +/-

Al Electrode layer

PDMS

Glass

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Testing of the device

- Two Packaging Method: Anodic Bonding and PDMs
- Testing Method: Manual control of fluid pressure via syringes connected to tubes

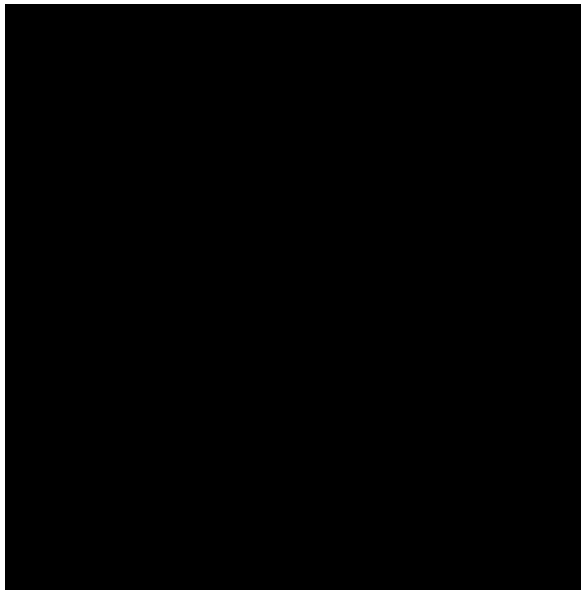
Anodic Bonding Characteristics:

- Using pre-drilled holes to connect tubes
- Using adhesive to seal the connection
- Able to withstand constant pressure with no leakage

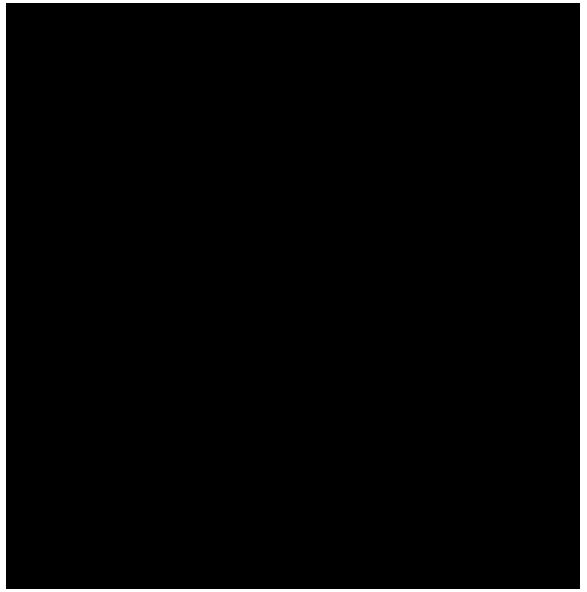
PDMs Characteristics:

- Using needles to insert tubes
- No adhesive to seal the connection
- Prone to leak due to insufficient connection strength. It is leaked midway through testing.

Anodic Bonding

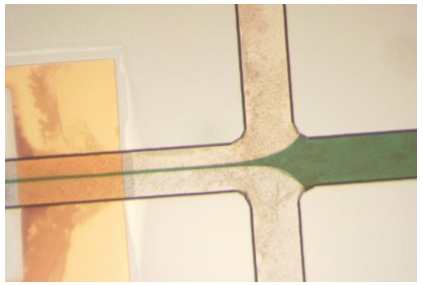


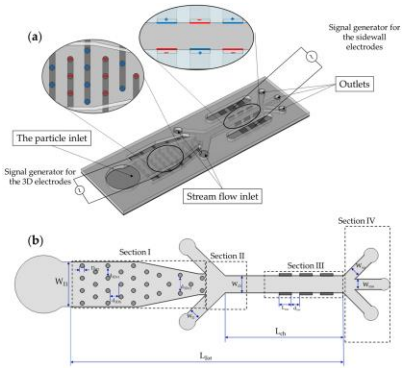
PDMs



Conclusion:

- The device made with PDMs worked for some time but started leaking midway.
- The device fabricated using anodic bonding with glass provided a tight seal and worked perfectly well.
- We successfully utilized flow focusing by using three syringes, each pressed with varying inlet pressures, and observed flow focusing.
- Future work includes the use of aluminum electrodes for further particle and flow focusing. However, this will require attaching electrical connections to the metal pads.
- Further flow focusing can be achieved through techniques such as inertial focusing, Dean flow, and magnetic and acoustic cell focusing. These advancements will require modifications to the device dimensions and the addition of new features for magnetic and acoustic cell sorting applications.





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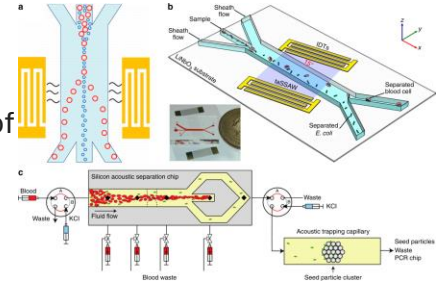

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[1] https://lifesciences.danaher.com/us/en/library/flow-cytometry-guide.html

[2] Brian Kirby - Micro- and Nanoscale Fluid Mechanics Transport in Microfluidic Devices-Cambridge University Press (2010)

[3] Knight, J., Vishwanath, A., Brody, J., & Austin, R. (1998). Hydrodynamic Focusing on a Silicon Chip: Mixing Nanoliters in Microseconds*. Phys. Rev. Lett., 80, 3863–3866.*

[4] Fernandes HP, Cesar CL, Barjas-Castro Mde L. Electrical properties of the red blood cell membrane and immunohematological investigation. Rev Bras Hematol Hemoter. 2011;33(4):297-301.

Thank you