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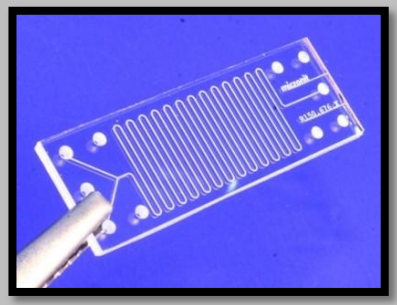
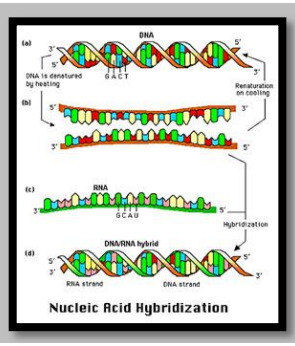
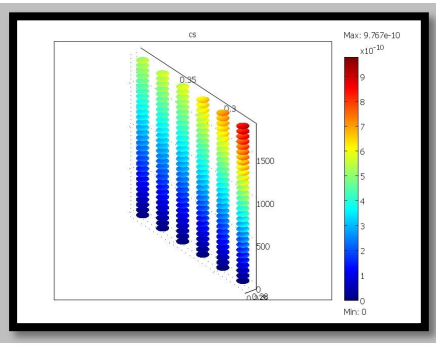
SAND2011-7607C

Sandia National Laboratories

Microfluidics: Kinetics of Hybridized DNA With Fluid Flow Variations







Presented By: Lizzy Schares; Mentor: Ron Manginell

October 19, 2011

MSEC 105, New Mexico Institute of Mining & Technology

Socorro, New Mexico

U.S. DEPARTMENT OF ENERGY

National Nuclear Security Administration

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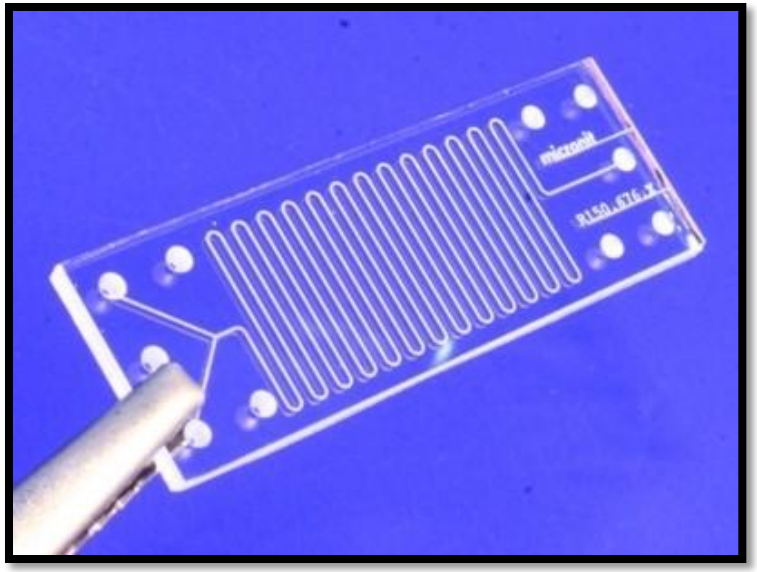
Overview

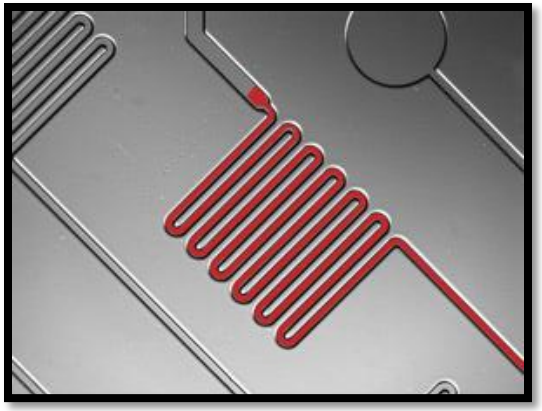
- Background
  - Microfluidics
    - Lab on a Chip
      - DNA Microarrays
  - Fluid Flow in Micro-channels
- Problem Statement
- Parameters
  - Chip Fluid Channel Geometry
  - Flow rates
  - DNA kinetics
- Results
- Conclusion



http://www.webbofscience.com/2009/08/26/melodies-divert-droplets/

Microfluidics

- What is Microfluidics?
  - Relatively new study, beginning in the 1980's
  - The study of fluid flow in geometries with at least 1 dimension less than 1mm in length
  - Fluid flow driven by capillary and interface phenomena
  - Re range from 1-20
  - Uses μL scale volumes which reduce waste, increase reaction time, and increases sensitivity

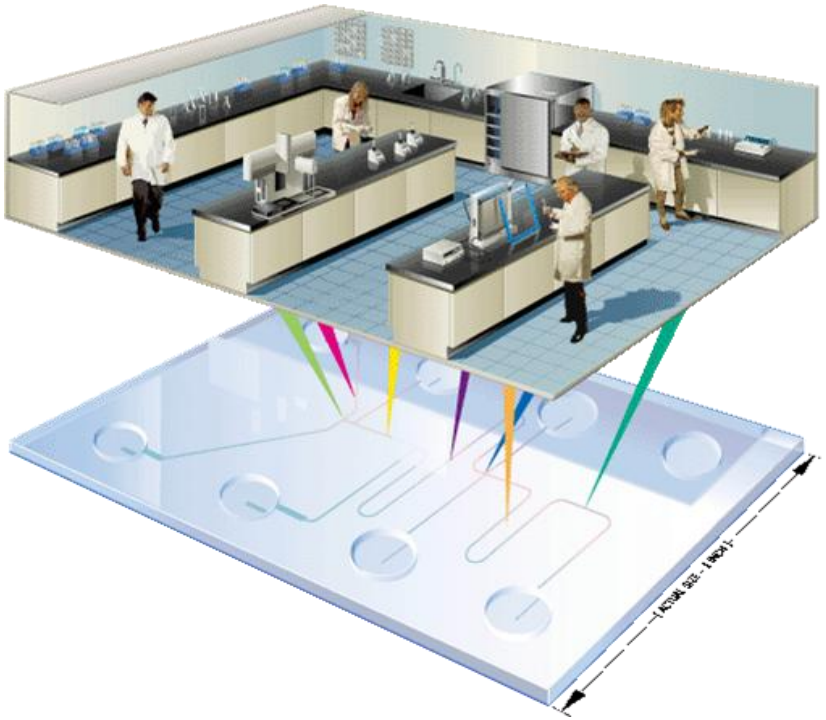


http://intelwars.com/2011/08/01/a-quick-cheap-diagnostic-test-for-hiv-and-other-infections/

Lab on a Chip

- Glass slide size chip or smaller

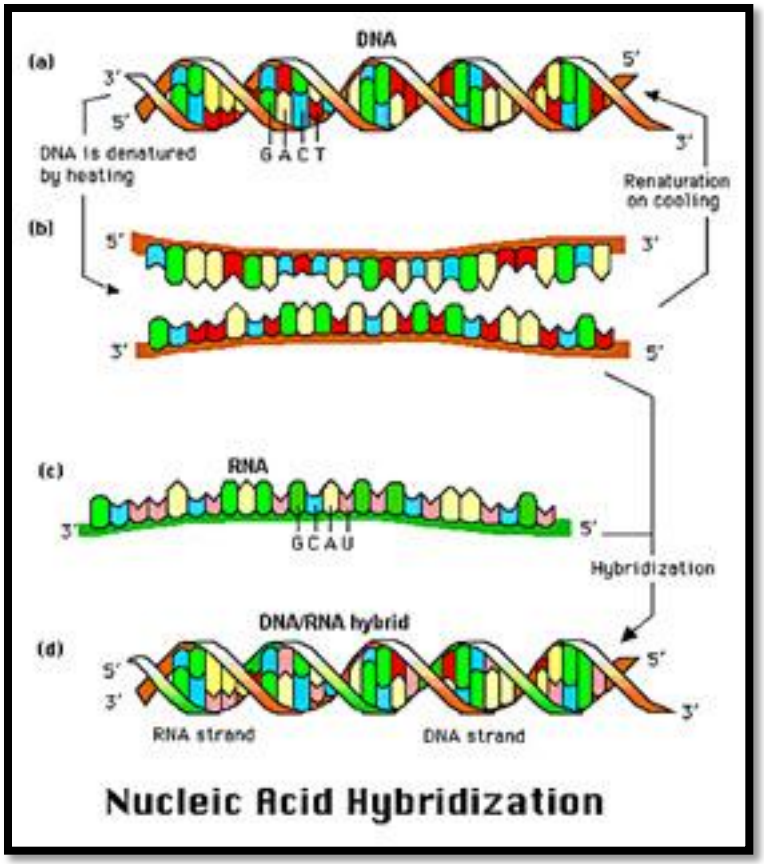
- Used for:
  - Separation Processes
    - Capillary Electrophoresis[1]
  - Biological Organ Functions
    - Lung on a Chip
  - Mass Spectrometry
  - Detection Devices



http://web2.clarkson.edu/projects/nanobird/2.4.php

DNA Microarrays

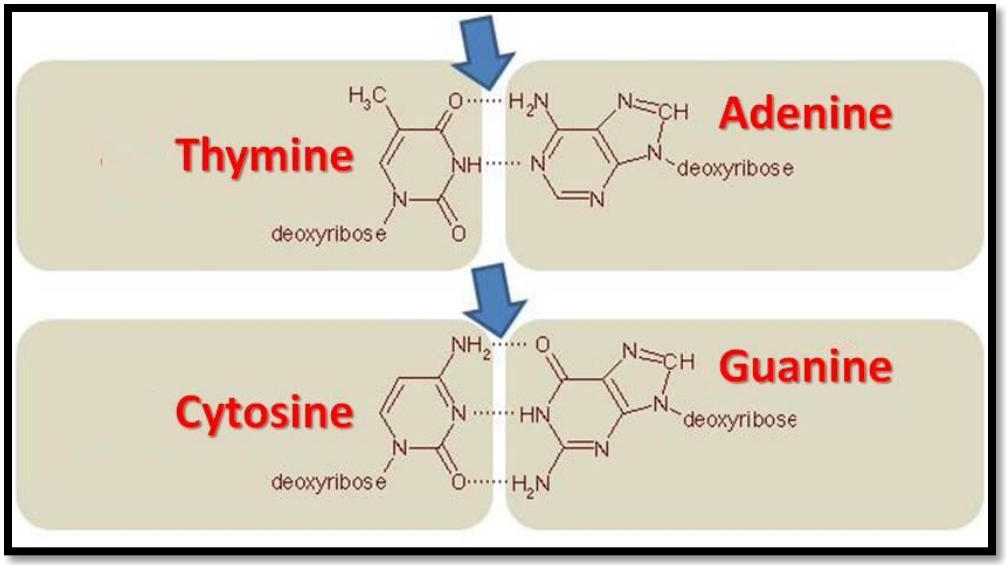
- DNA Detection Method
  - Hybridization
    - Increasing temperature causes denaturing
    - Keeps DNA untangled
    - Fluorescent tag attached
  - Targets on pads oligonucleotides
  - Kinetic parameters based on matching nucleotides
  - Detected by various laser wavelengths
    - Depends on fluorescent tag parameters



http://www.biochem.arizona.edu/classes/bioc462/462a/NOTES/Nucleic_Acids/nucacid_structure.html

DNA Kinetics

- Based on Hydrogen bonds
- Adenine + Thymine
  - 2 Hydrogen bonds
- Cytosine + Guanine
  - 3 Hydrogen bonds



http://en.wikipedia.org/wiki/File:AT-GC.jpg

Problem Statement

- Determine the concentration on the target probes with 16μL/min flow rate, 0.5μM input concentration, and kinetics based on 18 base pair DNA.
- Compare the results with a 30 minute diffusion time based on the same kinetic parameters.

Fluid Flow in Micro-channels

- Re between 1-20
- Flow driven by pressure drop across channel
- Modeled as hyperbolic flow with slowest flow at walls

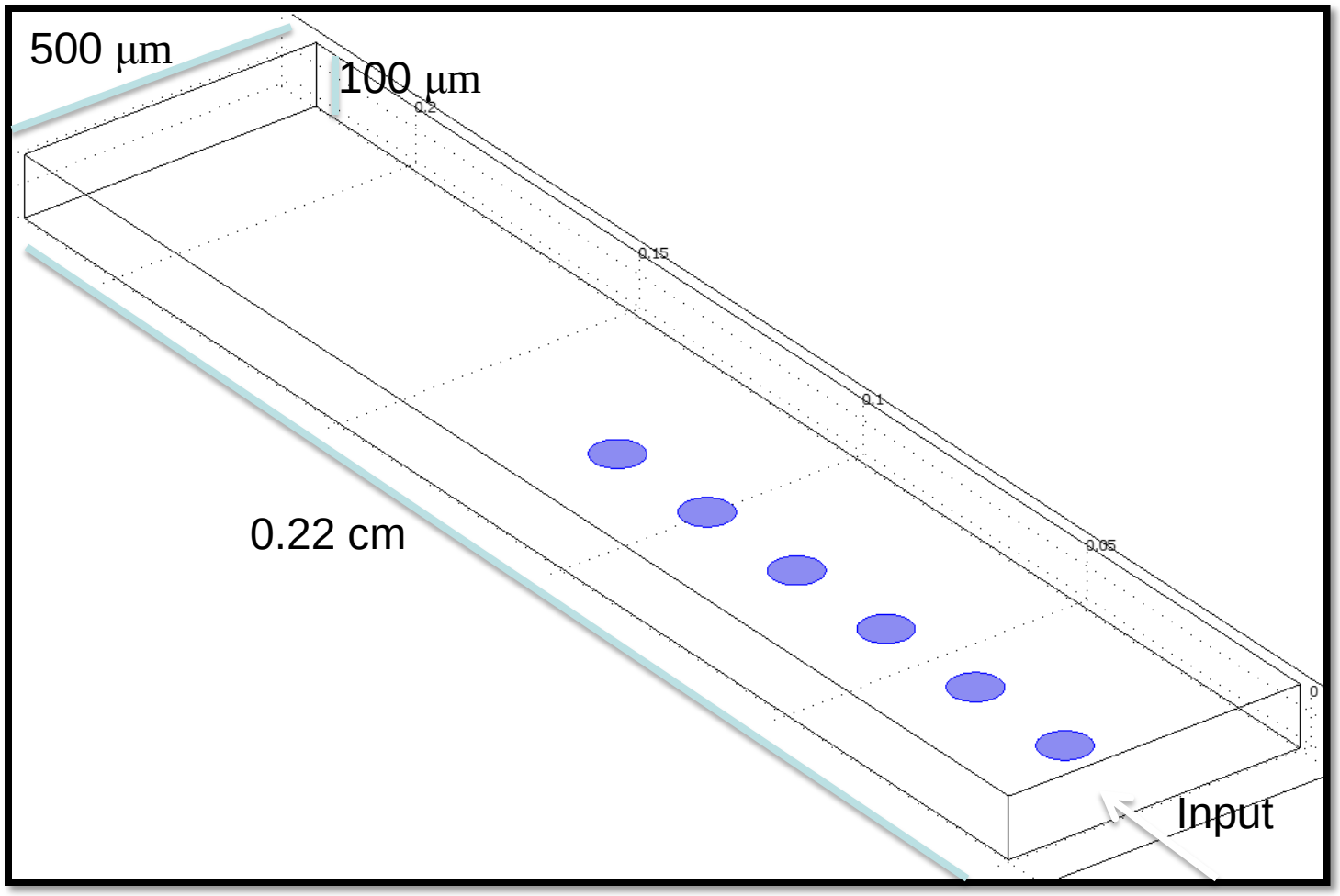
DNA Microarray Probe Specifications

- Kinetic equation from Ho Lee used
  - Ran experiments to determine kinetic parameters
  - Determined parameters:
    - K for perfect match: 3e-4
    - K for 1 base pair mismatch: 3.5e-4
    - K for mismatch: 2e4

Channel Geometry Specifications

- Geometry of Fluid Channel
  - 500μm wide
  - 100 μm height
  - 65mm length
  - Large quantity of target probes on bottom surface
- Geometry of COMSOL Channel
  - 500 μm wide
  - 100 μm height
  - 2.2 mm length
  - 6 target probes located on bottom surface
    - DNA Targets on pads oligonucleotides

Setting up the COMSOL Model: Building the Channel



Setting up the COMSOL Model: Physics Settings

- Transient Analysis:
- Navier-Stokes Flow
  - Fluid flow in the channel based on viscosity, density, linear flow rate and pressure drop
- Convection and Diffusion
  - Mass transfer trend based on Navier-Stokes Flow and diffusion coefficients
- PDE Model Boundary

  Boundary condition to determine pad concentration

DNA Kinetic Parameters

- Treated as a general surface adsorption problem.
  - Governing Equation:

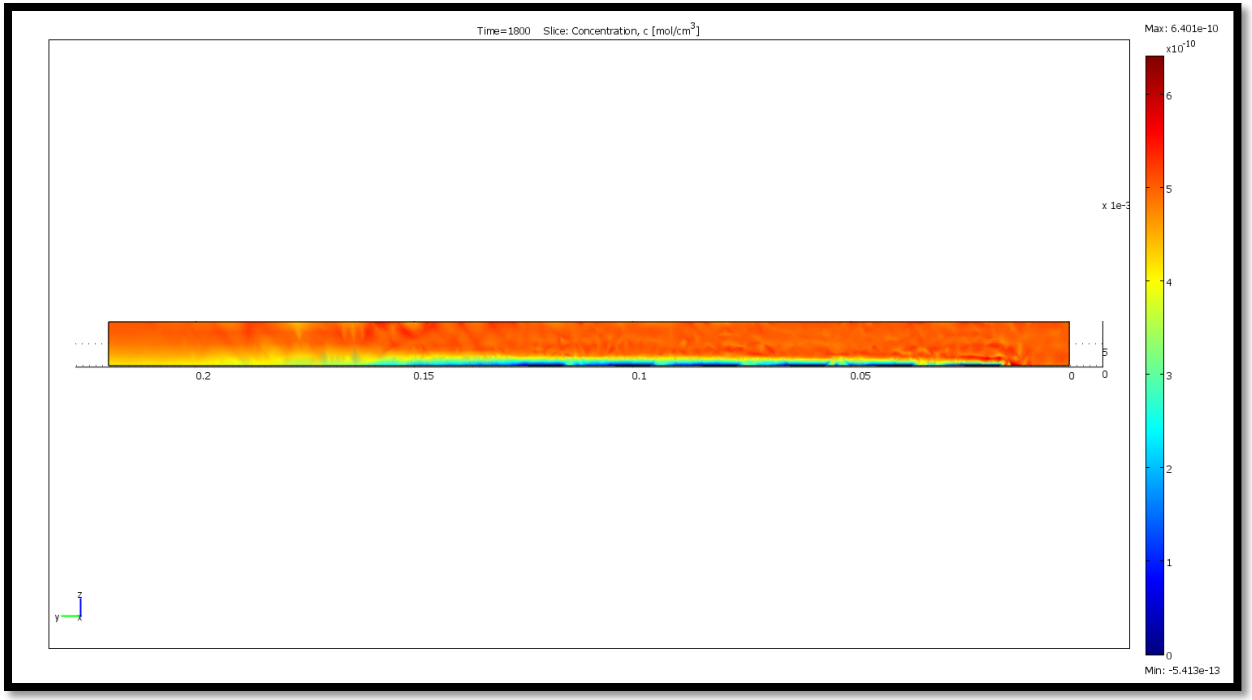
$$C + \Theta \underset{k_{des}}{\overset{k_{ads}}{\rightleftharpoons}} C_s$$

    - C: Bulk concentration
    - Θ: Available number of hybridization or adsorption-active sites
    - C_s: Surface concentration or adsorbed species
    - k_{ads}: Rate constant for hybridization reaction
    - k_{des}: Rate constant for dissociation reaction
  - Rate constants depend on number of base pairs in DNA and Temperature

Recirculating flow accelerates DNA microarray hybridization in a microfluidic device. Hyun Ho Lee

Results: 16 μL/min Flow Rate

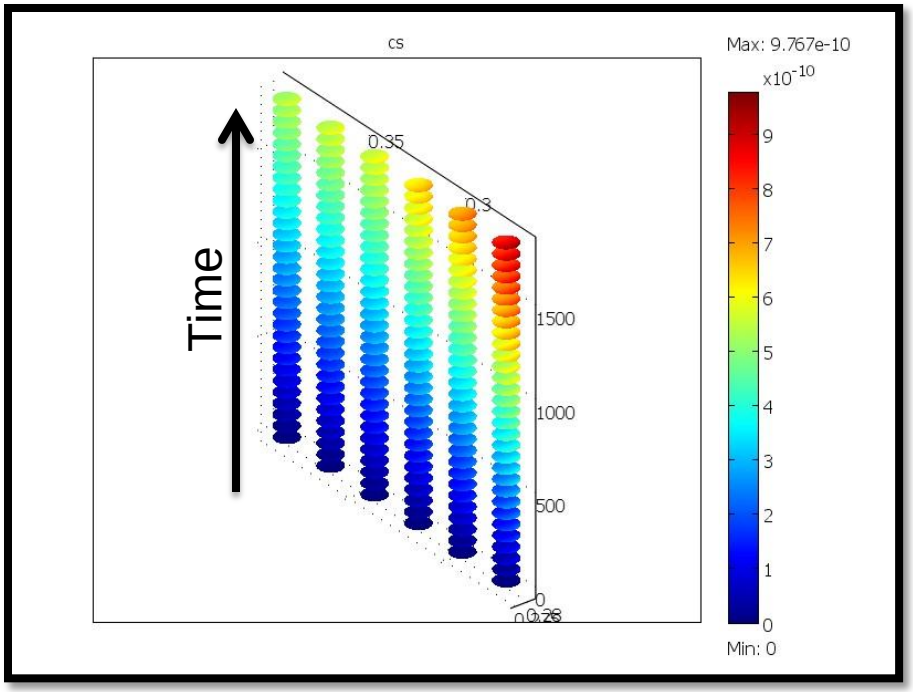
- Input 0.5 μM DNA solution for 30 minutes



Perfect match desorption coefficient

K_{des}: 3e-4

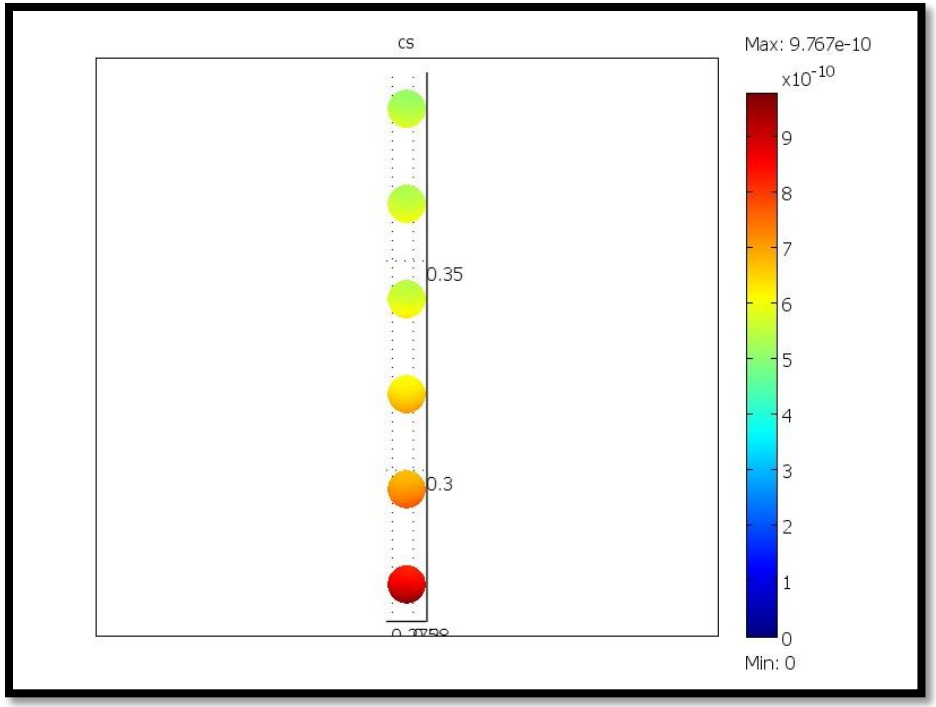
Results: 16 µL/min Flow for 30 Minutes Surface Concentration



3D Concentration from 0-30 min

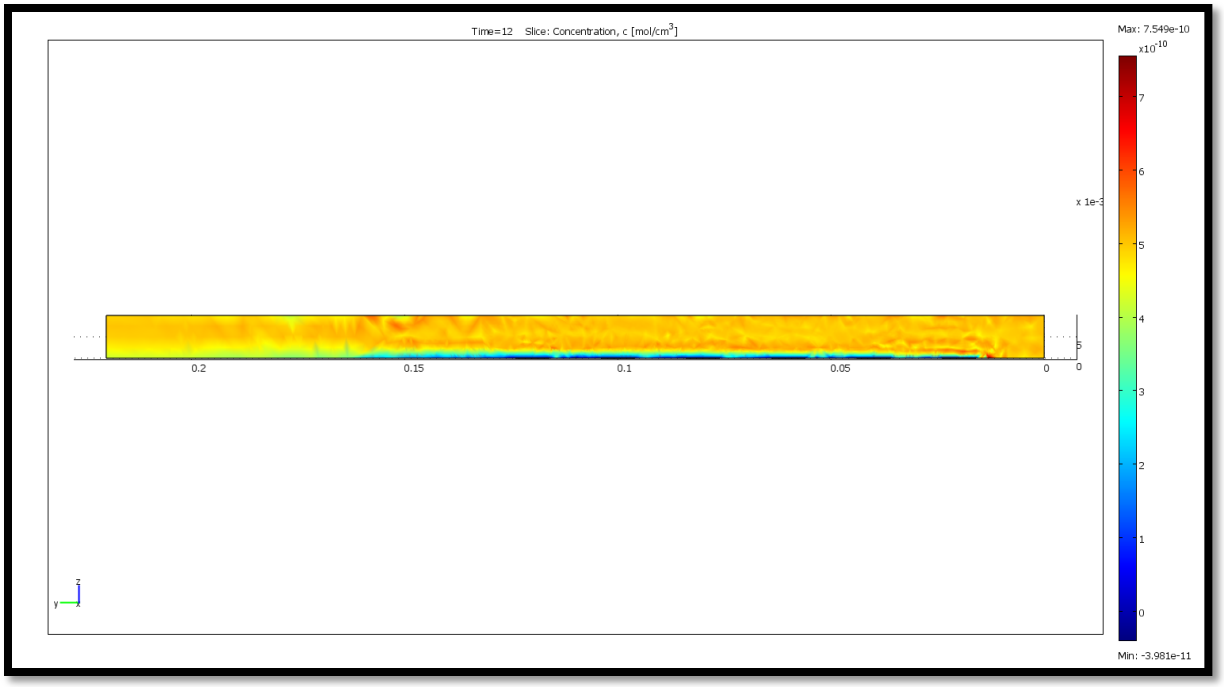
Max concentration 0.0967 µM

Results at 30 minutes



Results: 16 μL/min Flow Rate

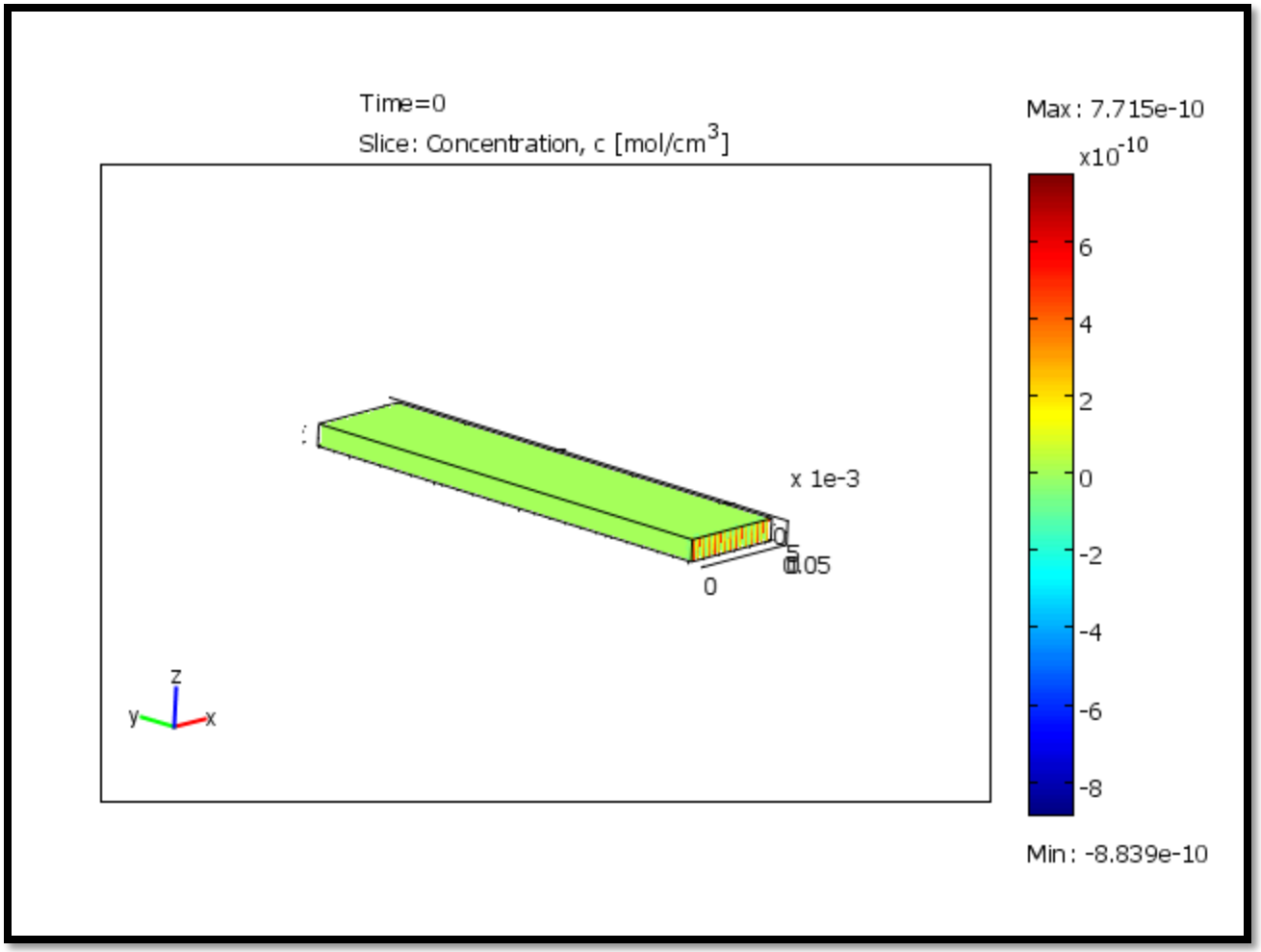
- Input 0.5 μM DNA solution for 12 seconds



Perfect match desorption coefficient

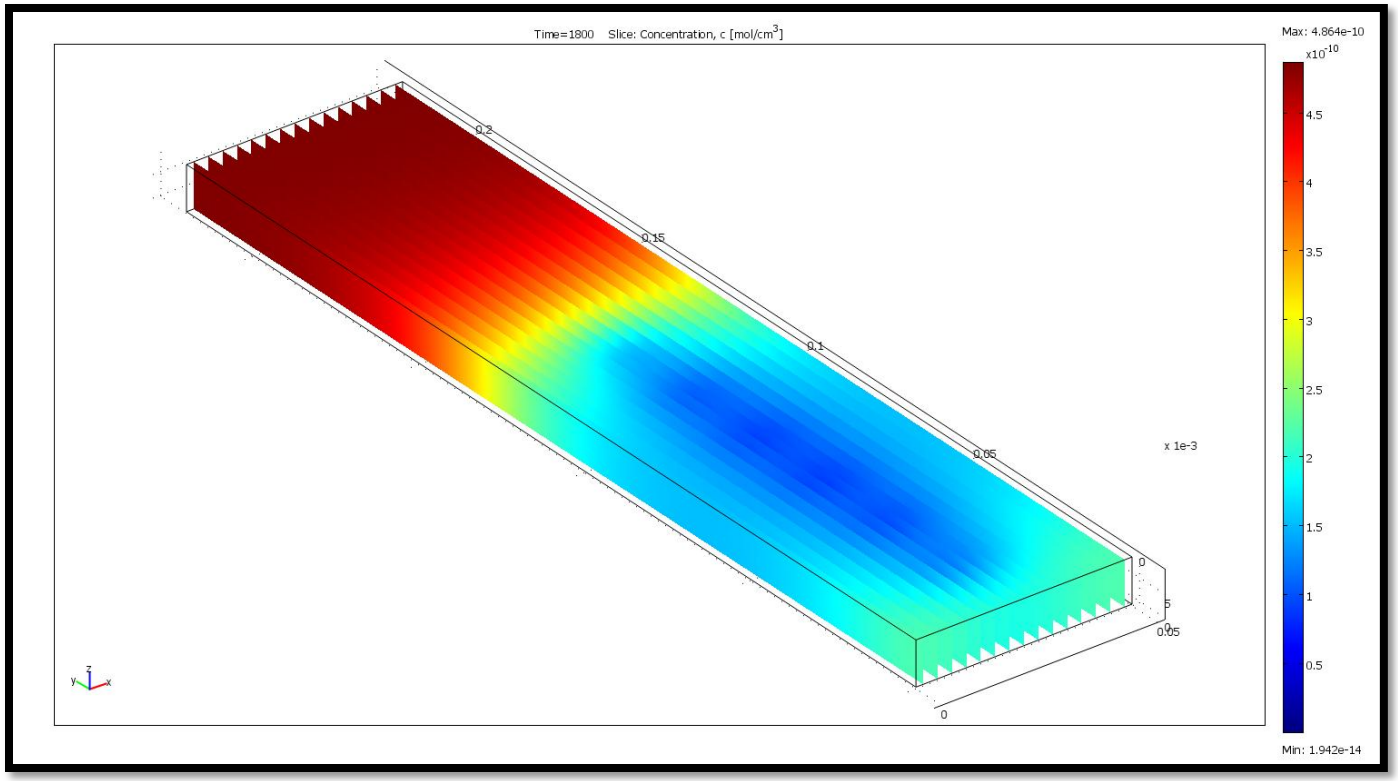
K_{des}: 3e-4

Results: 8 µL/min Flow Rate Video

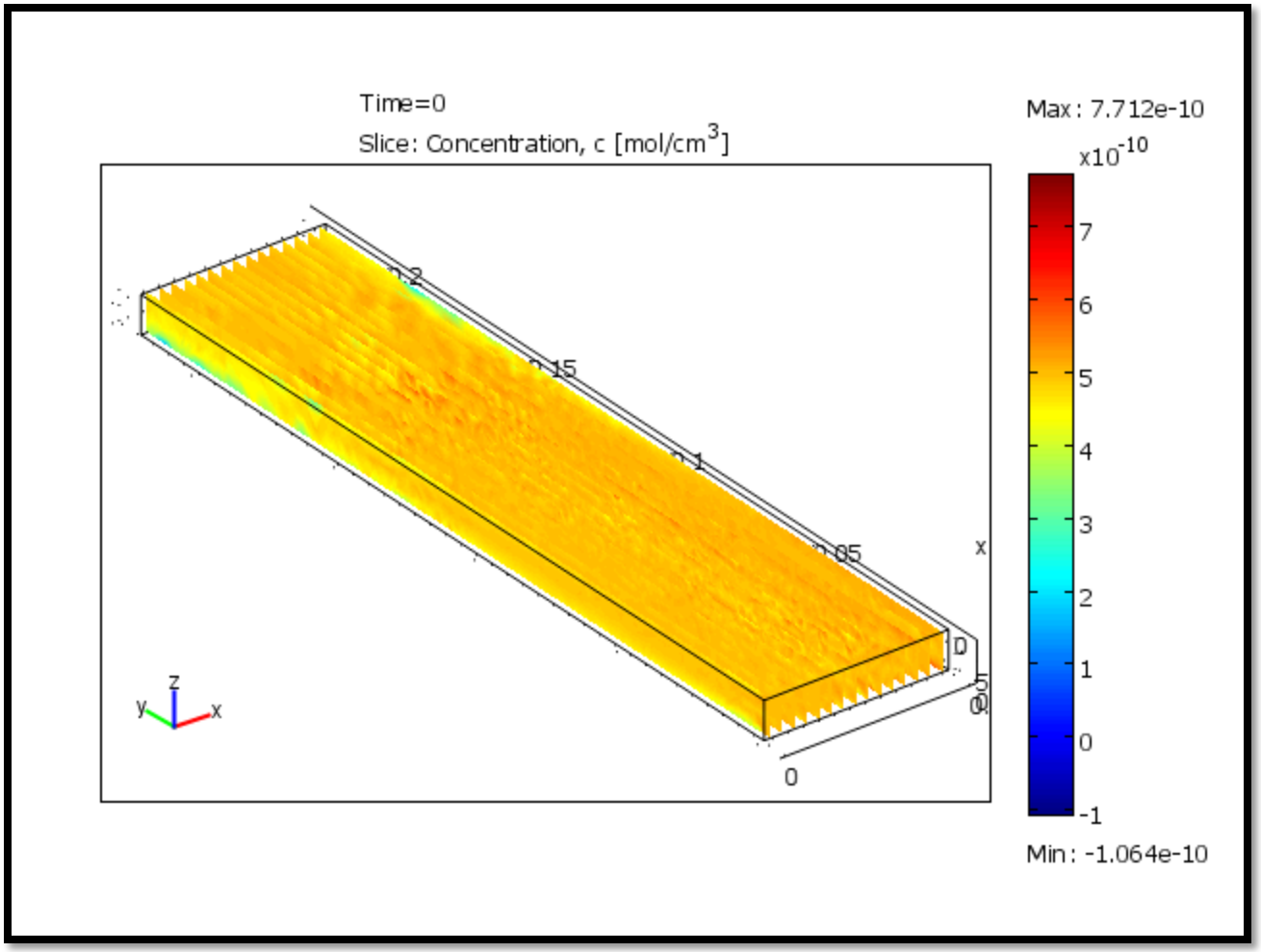


Results: 16 µL/min Flow Rate

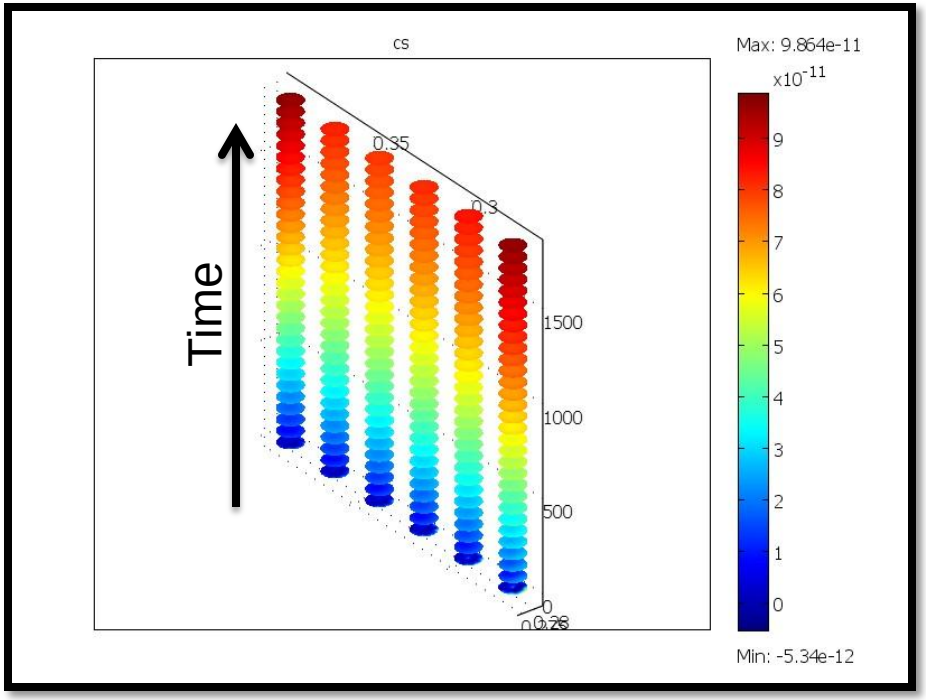
- Concentrated towards front of pads
  - Want a more uniform concentration gradient



Results: 16 µL/min Flow Rate Diffusion Video



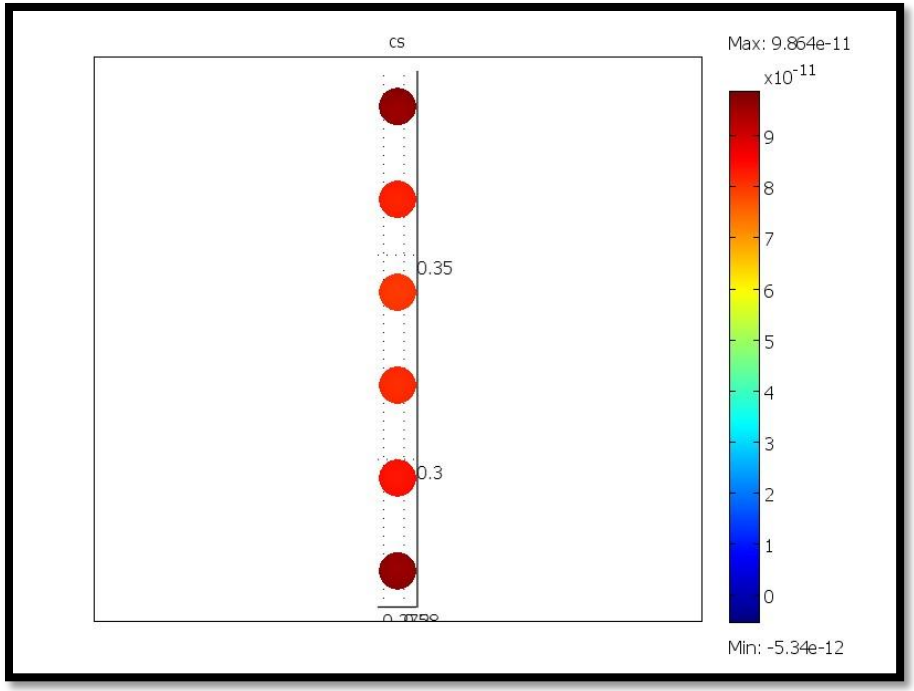
Results: 16 µL/min Diffusion for 30 Minutes Surface Concentration



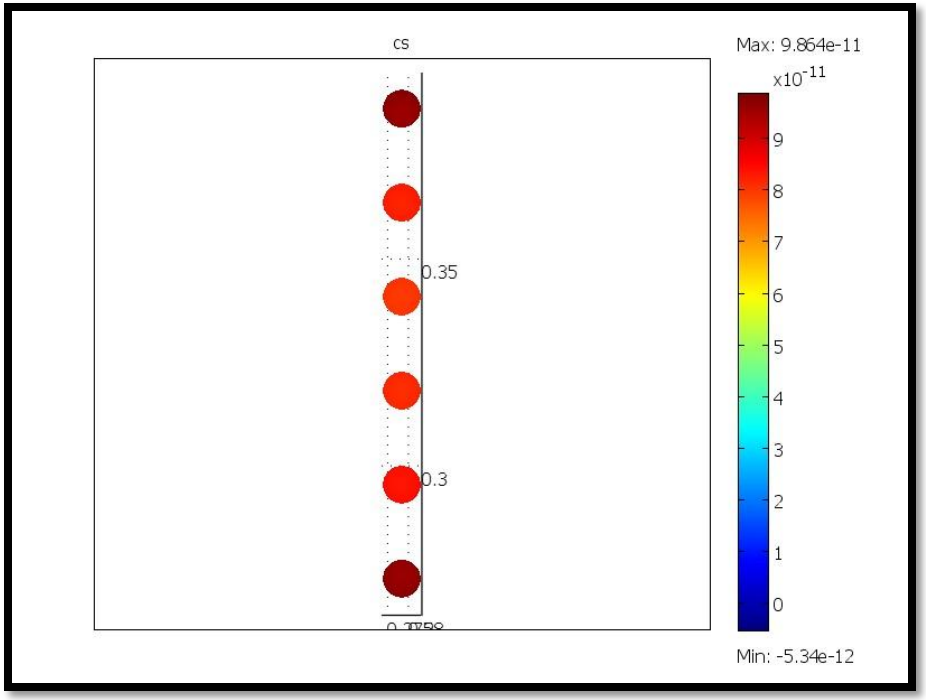
3D Concentration from 0-30 min

Max concentration 0.0986 µM

Results at 30 minutes

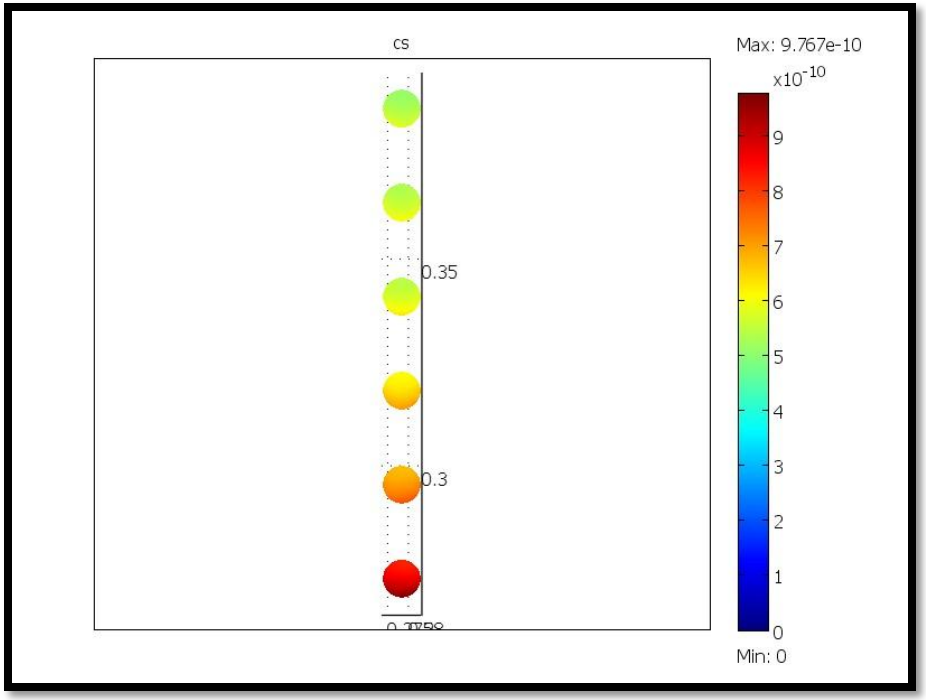


Results: 16 μL/min vs 8 μL/min



30 minute Diffusion

Max concentration: 0.0986 μM



30 minute Input Flow

Max concentration: 0.0976μM

Conclusion

- From the analysis:
  - Short input time + long diffusion time
    - Slightly higher target probe concentrations
    - More uniform target probe concentrations
    - Less waste
- Finite Element Analysis is a very helpful tool.
  - Shows general results without running experiments
- Problems
    - Can only run flow rate to a certain point before model breaks down
      - Past ~25 uL/min model
    - Must have specific data on all parameters

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Further Work

- Changing the number of base pairs
- Increasing system temperature
- Changing dimensions of channel

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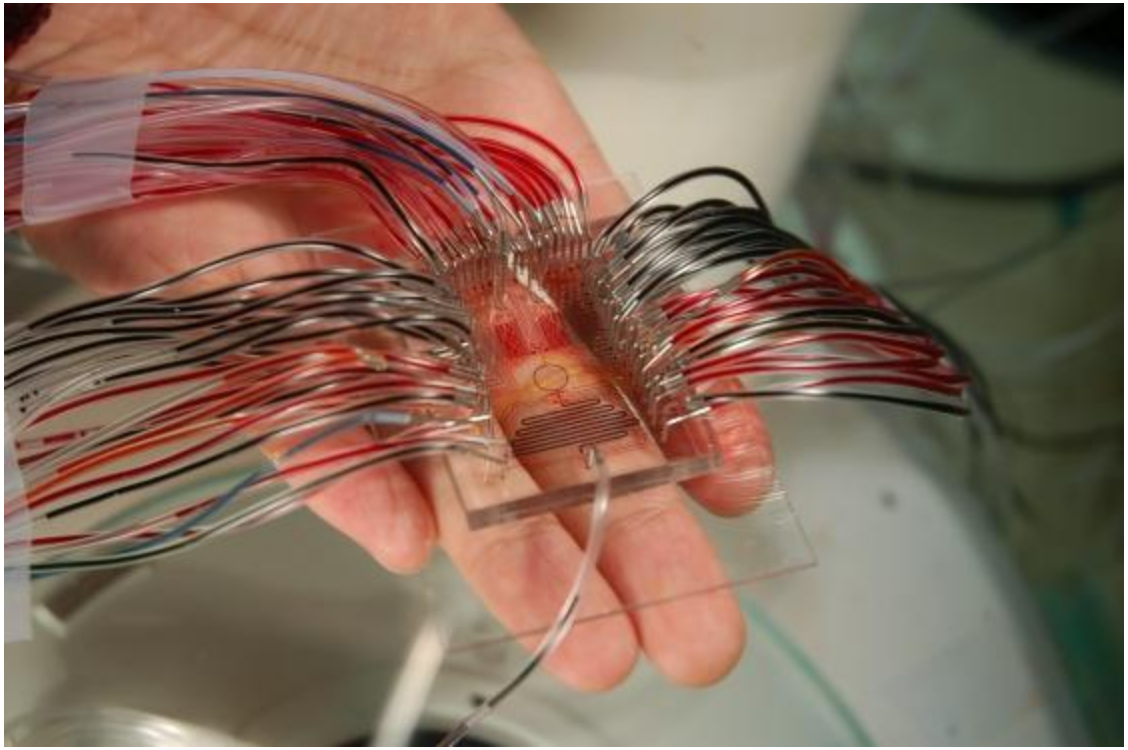
Questions?



http://gizmodo.com/lab_on_a_chip/

References

1. Berthier, J. (2009) *Microfluidics for Biotechnology.*
2. *Lee, H.H. et al, (2006).* Recirculating flow accelerates DNA microarray hybridization in a microfluidic device. Lab on a Chip, 6 (9).
3. Schmidt, L. D. (2005) *The Engineering of Chemical Reactions*.
4. http://nanobio.ftf.lth.se/~tegen/theses/jonsson2004.pdf
5. http://www.mnstate.edu/marasing/CHEM480/Handouts/Chapters/Capillary%20Electrophoresis.pdf