
Reconfigurable Digital Microfluidic Chips for Multiple Chemical Applications

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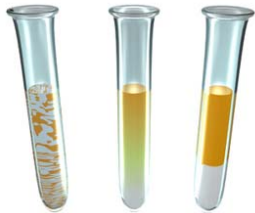


Outline of Presentation

- Background and motivation
 - The single-use-chip paradigm
 - Is there a case for integrated, reconfigurable microfluidic devices?
- Microfluidic reconfigurability issues
 - Architectural choices
 - Digital microfluidics
- The digital microfluidic options and examples
 - Implications of droplet architecture
 - Examples of reconfiguration benefits
 - Cytotoxicity screening
 - DNA sequencing
- Summary and conclusions



Background & Motivation



Test tubes

- ☐ Automation
- ☐ Integration
- ☐ Miniaturization
- ☒ Reconfiguration

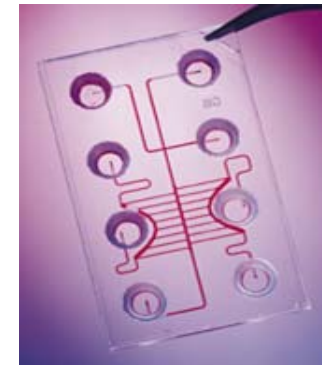


Robotics

- ☒ Automation
- ☒ Integration
- ☐ Miniaturization
- ☒ Reconfiguration

Microfluidics

- ☒ Automation
- ☒ Integration ?
- ☒ Miniaturization
- ☐ Reconfiguration



Start of the Art Commercial Disposable Microfluidics

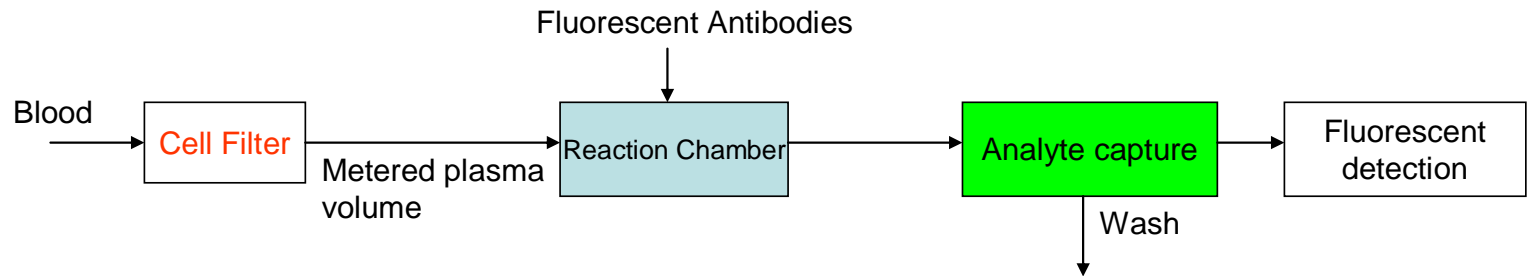
*ANOTHER EXAMPLE
OF A MICROFLUIDIC
SYSTEM*



LAB-ON A CHIP BIOSITE

DIAGNOSES
HEART ATTACK
WITHIN 10 MN

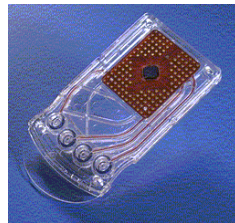
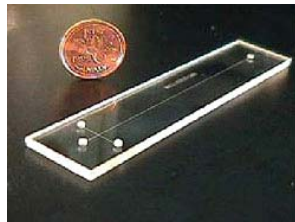
Disposable Chip Paradigm



Concept of Disposable Integration

Application Devices

MICROFLUIDIC

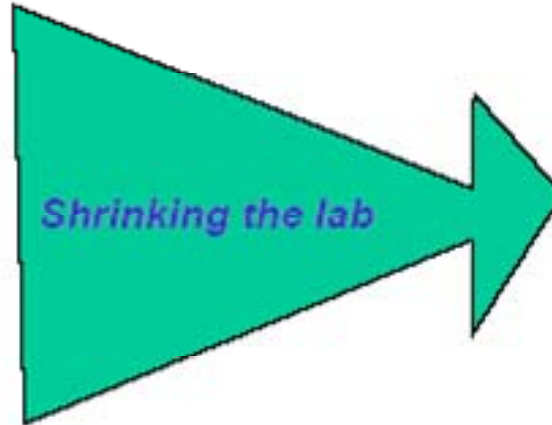


PROCESSING/ANALYSIS



Promise of Biochips

Applications : Biotechnology (eg: high throughput screening , Diagnostics...)



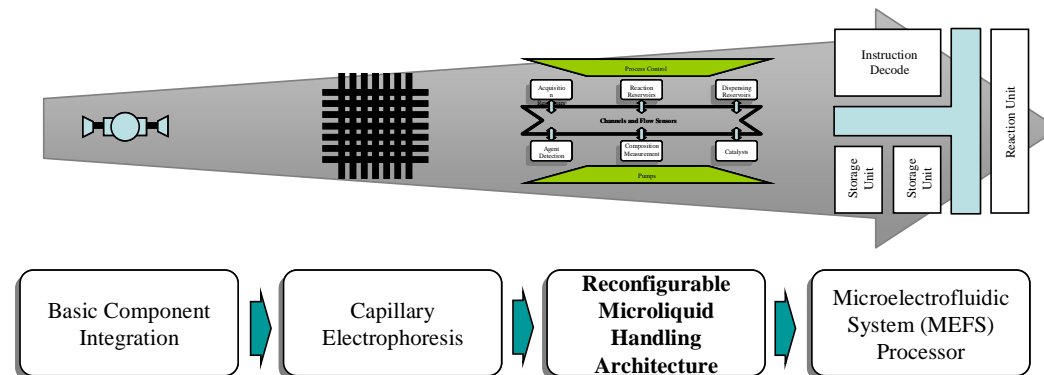
mm, or cm



Can a lab-on-a-chip be as versatile as the macro lab it replaces?

Architectural Choices

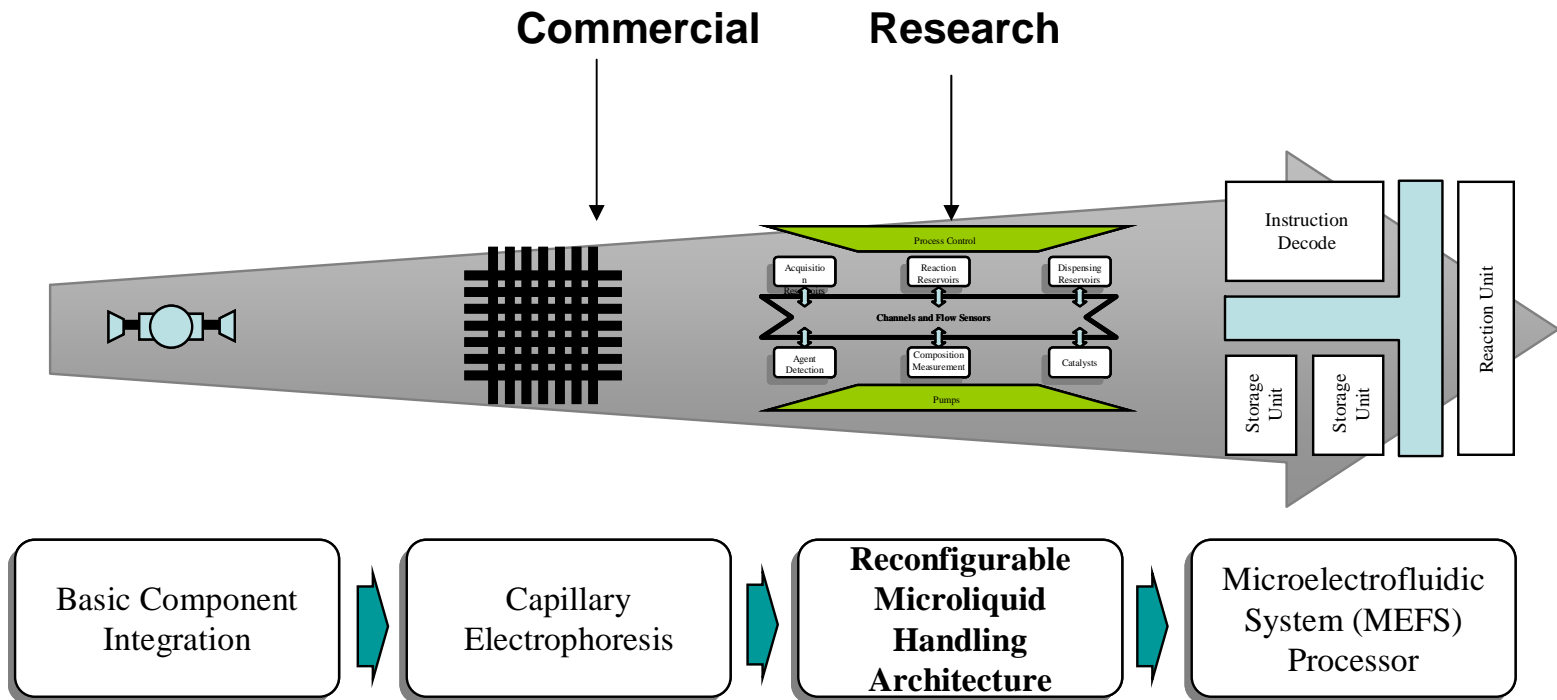
- Fixed data path (application specific)
- Reconfigurable (multiple applications)
 - Shared elemental operations
 - Microfluidic instruction set
 - Programmable
 - Reusable



The Bigger Implications

- Application-specific microfluidic chips
 - Require low-level hardware design
 - Only a relatively few designers available
 - Limited leverage of chip technology
- Reconfigurable microfluidic chips
 - Allow system design at programming level
 - Users with applications >> hardware designers
 - Opens access by users to microfluidic solutions

Where Are We?



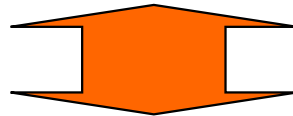
Present Status Summary

- The reality of current lab-on-a-chip technologies...
 - Highly application specific
 - Commercial trend: simple, disposable devices that interface with expensive control boxes
 - Disposable devices may perform limited set of steps
- What is required for a reconfigurable microfluidics?
 - Leverage devices into multiple applications
 - Complexity of diverse applications reduced to a manageable set of fluidic operations
 - Modular architecture gives flexibility of choosing fundamental operations
 - Integrated fluidic I/O
 - Integrated low voltage CMOS control incompatible with current fluidic operating voltages and footprints (optional)
 - Detector integration a priority

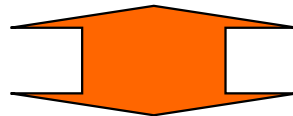


Complexity of Diverse Applications Reduced to a Manageable Set of Fluidic Operations


**Biomedical Fluidic
Functions:** **Func.1, Func.2,.....,Func.n**





**Elemental Set of
Operations:** **Op.1, Op.2,.....,Op.i**



**Elemental Set of
Components** **Comp. 1, Comp. 2,...,Comp. n**

- 
- Agent Detection
 - Precision Dispensing
 - Enzyme Analysis
 - Electrochromatography
 - Capillary Electrophoresis
 - Molecular/Protein Analysis
 - Isotachophoretic Separation

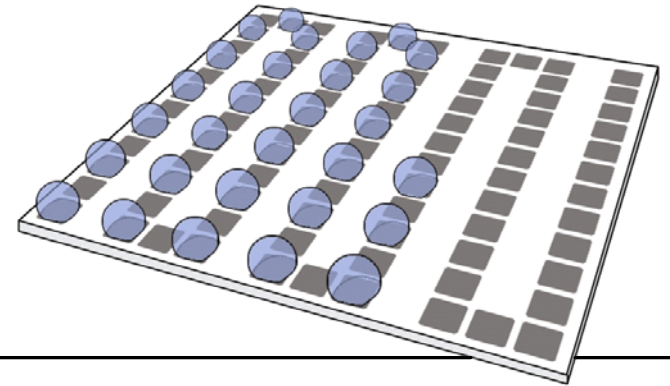
- 
- Transport
 - Mixing
 - Flushing
 - Filtering
 - Analysis
 - Detection
 - Monitoring

- 
- Buffers
 - Channels
 - Valves
 - Mixers

Reconfigurable Digital Microfluidics

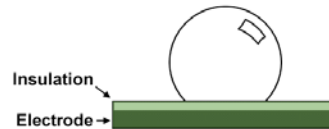
Features

- Droplet-based microfluidic devices
 - Droplets are moved in “virtual channels” defined by electrodes
 - Programmable electrodes in an array directly control discrete droplet operations – dispense, transport, mix, split, incubate – to perform any liquid-based test

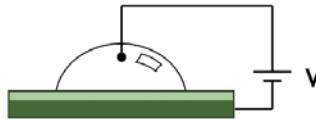


How It Works

Voltage Off:



Voltage On:



- Electrowetting
 - Modulation of solid-liquid interfacial tension by the application of an electric field
- Works with or without a top plate
 - Newly developed coplanar electrowetting method

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Technology Advantages

Digital Microfluidics

- Very accurate droplet volumes
- Droplet-based digital microfluidics is functionally more similar to bench protocols
 - Assays more easily adapted
- Programmable, software-driven electronic control
 - No moving parts, tubes, pumps or valves
- More efficient use of samples and reagents
 - No liquid is wasted priming channels
- Extremely energy efficient
 - Suitable for low power and portable applications
- Development cycles are short, and assays can be tuned with software changes
- Low cost, production-ready lab-on-a-chip on printed circuit board substrate

Other Microfluidic Technologies

- Pump fluids through channels
- Must adapt assays to channel-based format
- Complex or multiplexed assays become a plumber's nightmare
- Off-chip pumps and valves mean large, expensive equipment and low reliability
- Expensive, time consuming, up-front investments required for most chip developments
- Designs are fixed in the development process



Digital Microfluidic Toolkit

Implementing numerous applications on a elemental set of components:

Reservoirs ➡ droplets

Dispensers ➡ electrode sets

Pumps ➡ electrode sets

Valves ➡ electrode sets

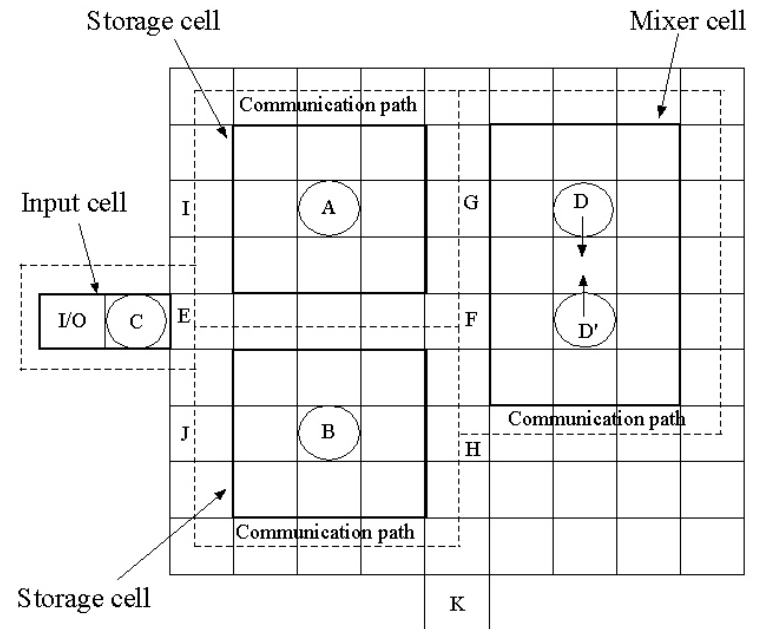
Reaction vessels ➡ droplets

Mixers ➡ electrode sets

Collection ➡ scanning droplet

Implications of Droplet Architecture

- Droplets allow microfluidic functions to be reduced to a set of basic operations
- Numerous elemental fluidic operations can be accomplished with a common set of elemental components
- Array can be partitioned into “cells” that perform fluidic functions
- Functional cells dynamically reconfigured at least once per clock cycle

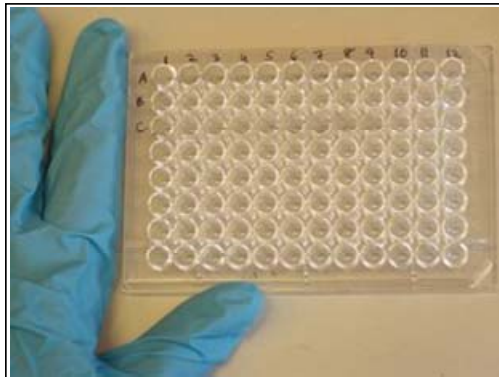


Reconfigurable Lab-on-a-Chip Status

- Digital microfluidic toolkit demonstrated
 - All fluidic functions demonstrated
 - Lacking molecular separation method
- Commercial prototypes available (ALL)
- Example from ECE299 (Duke Univ. Fall 2007)
 - Cytotoxicity screening
- Example from current research
 - DNA sequencing by synthesis



On-chip Dilution Tree for Cytotoxicity Screening (Y. Zhao, A. Wang, Y. Yamanaka)



Grow cells in 96 well plate



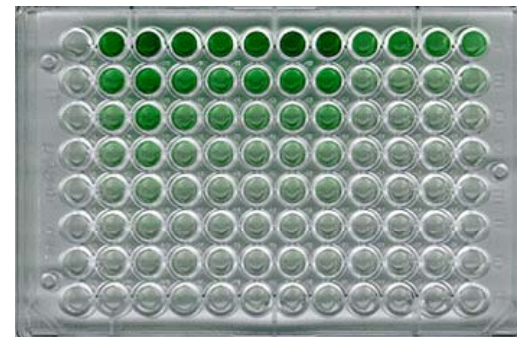
Add various concentrations of compound to be tested to cells



Wait specified length of time

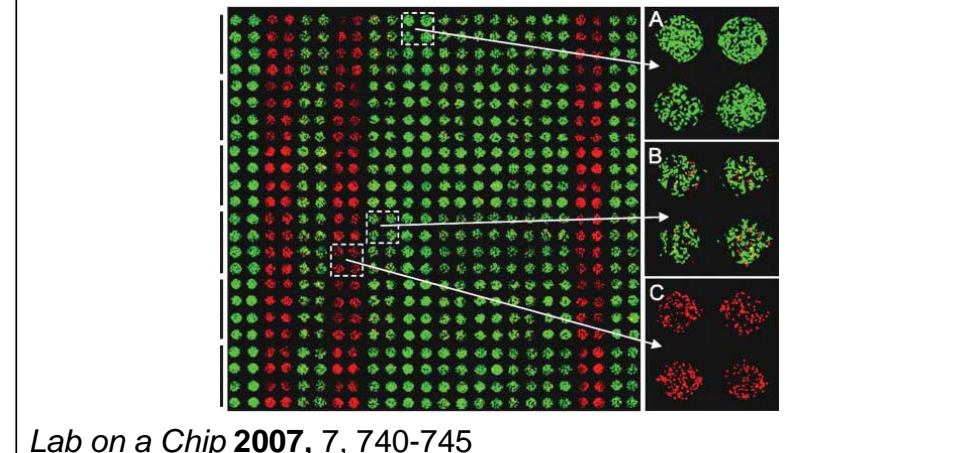
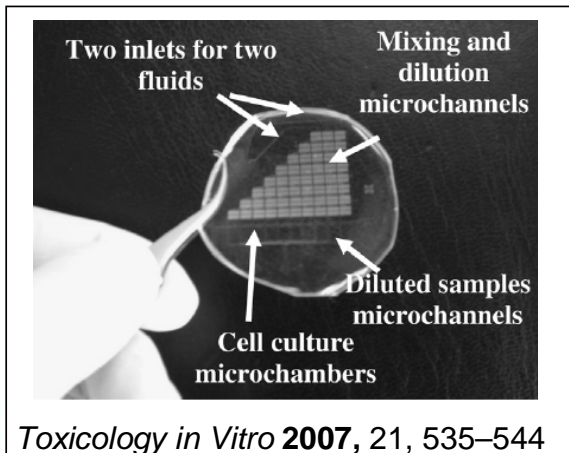
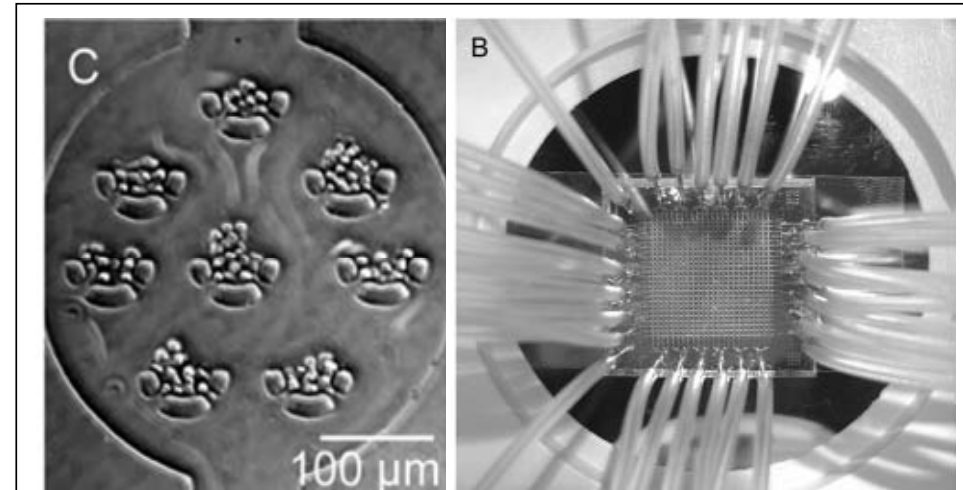
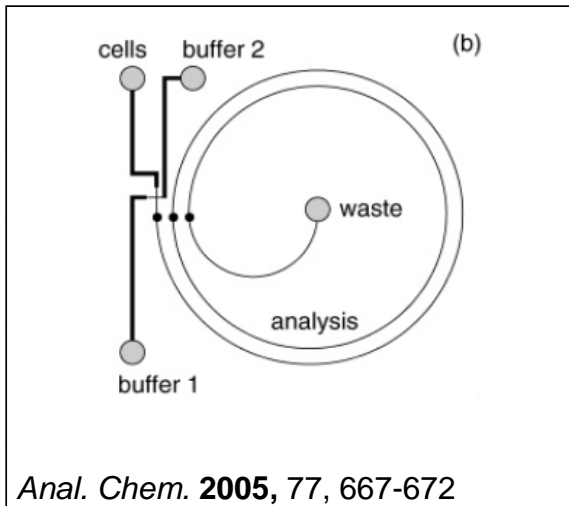


Add Cytotoxicity Assay reagent 1, incubate, add reagent 2

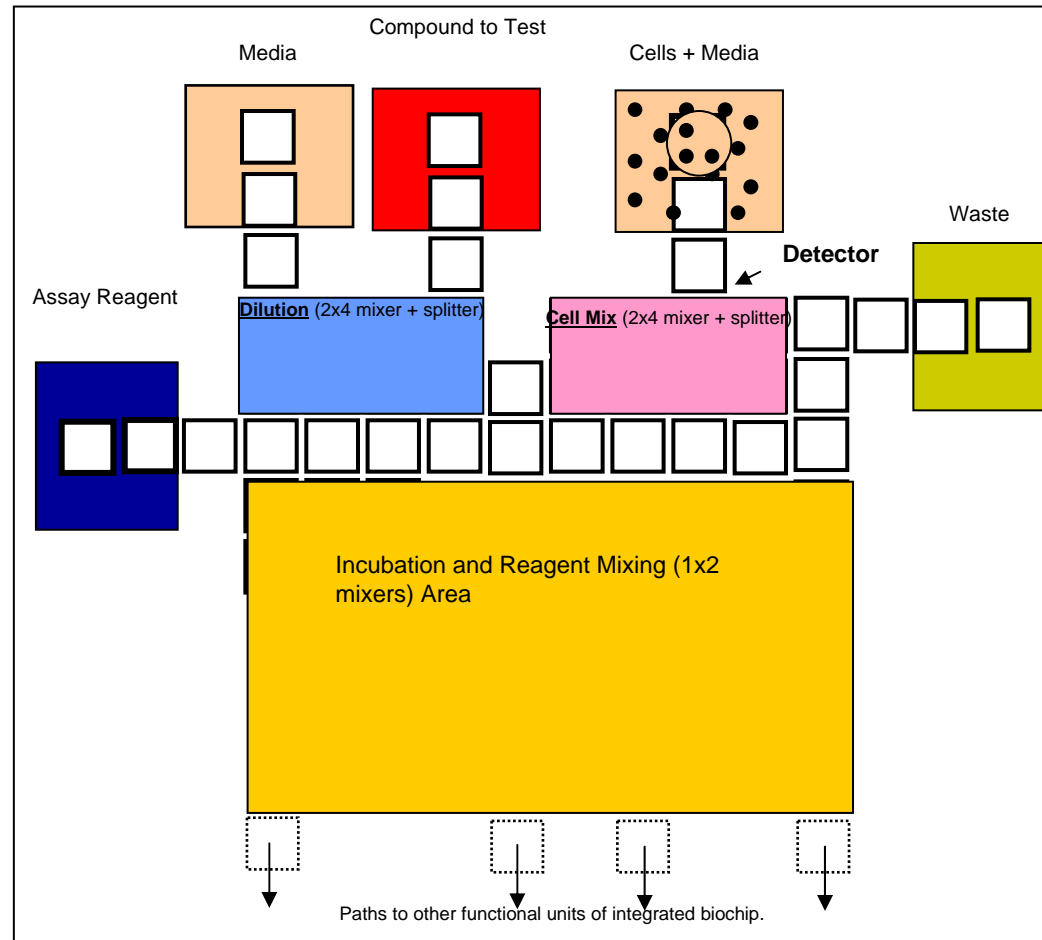


Use plate reader to measure color intensity (proportional to survival)

Previous Work



Digital Microfluidic Screener

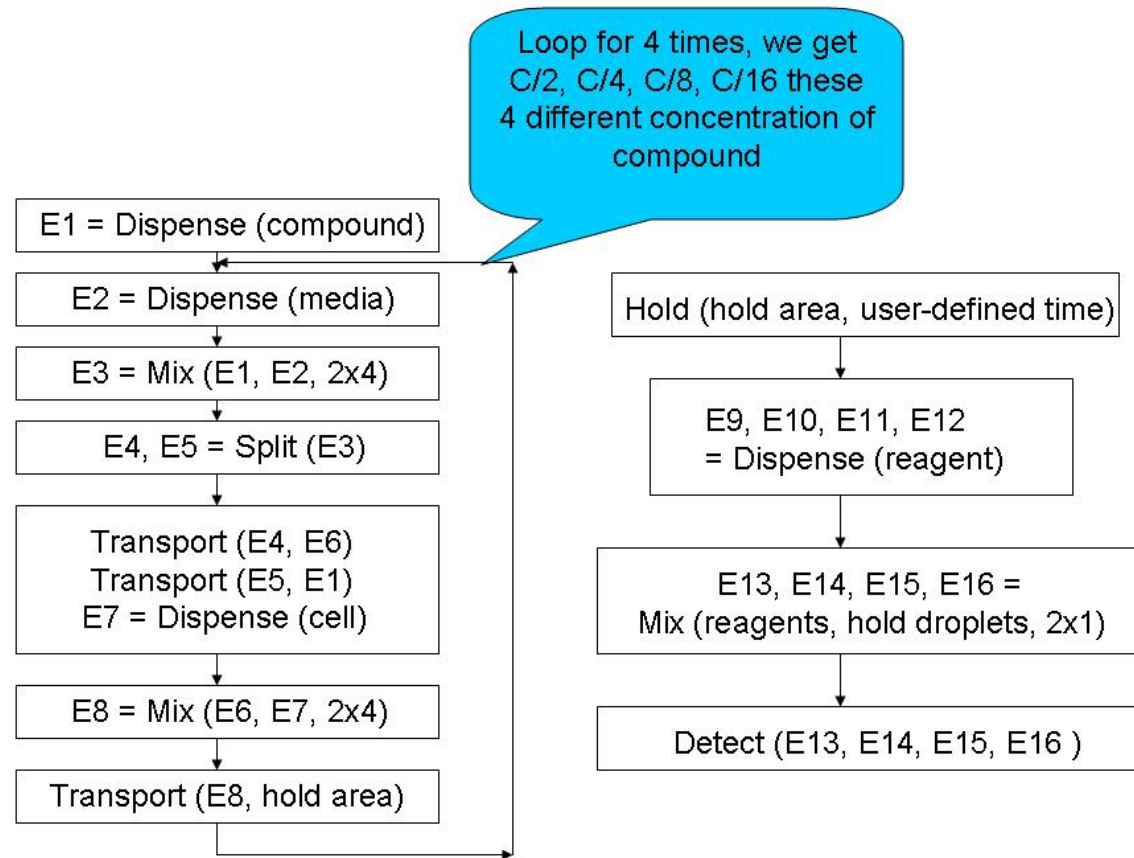


Algorithms and Programming

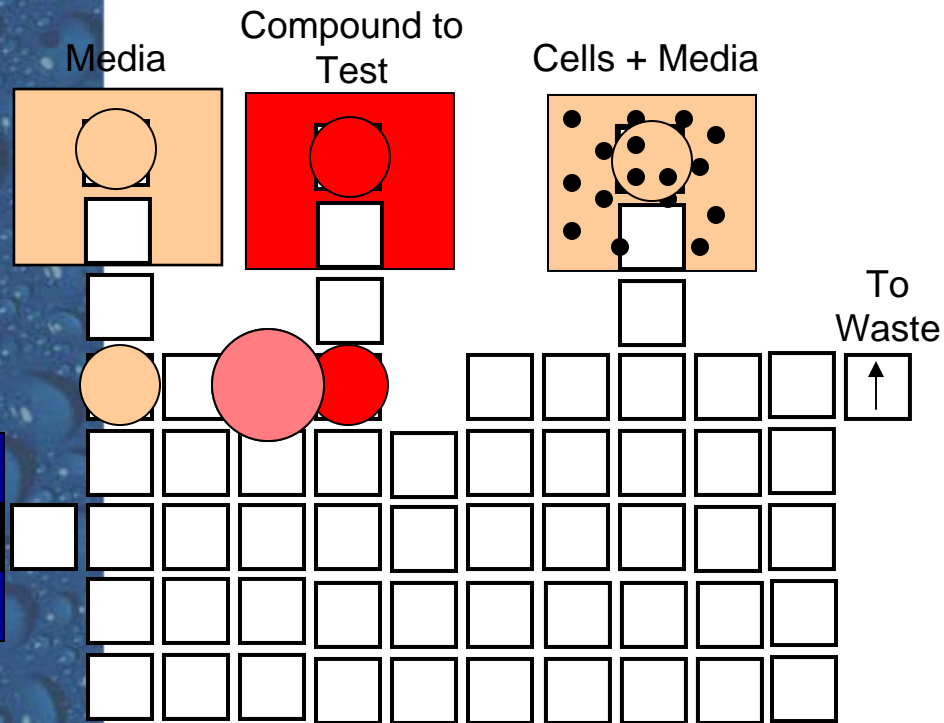
- Cytotoxicity screening implemented on platform using basic microfluidic operations
 - Transporting, merging, mixing, and splitting
 - Requires on-chip binary dilution
- Functional control requires abstraction layer between protocols and microfluidic operations
- Abstraction layer translate protocols into programming control statements
 - Dispense, transport (a,b), mix (a,b, type), split (a), detect (a)



Programming flow for dilution and cell injection

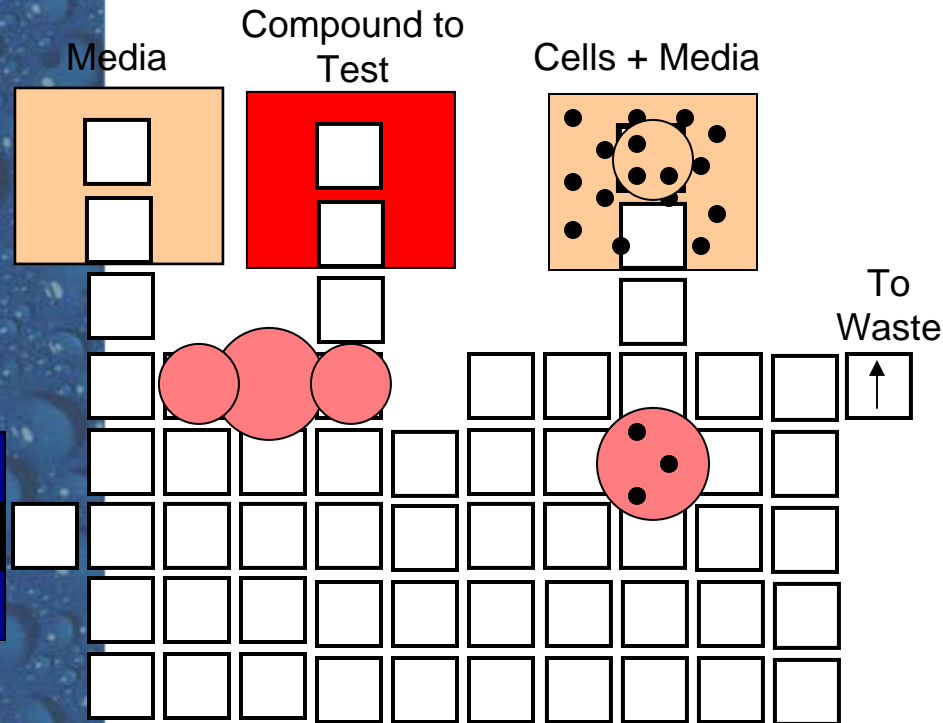


Architecture



1. Dispense buffer and compound droplets, mix.

Architecture

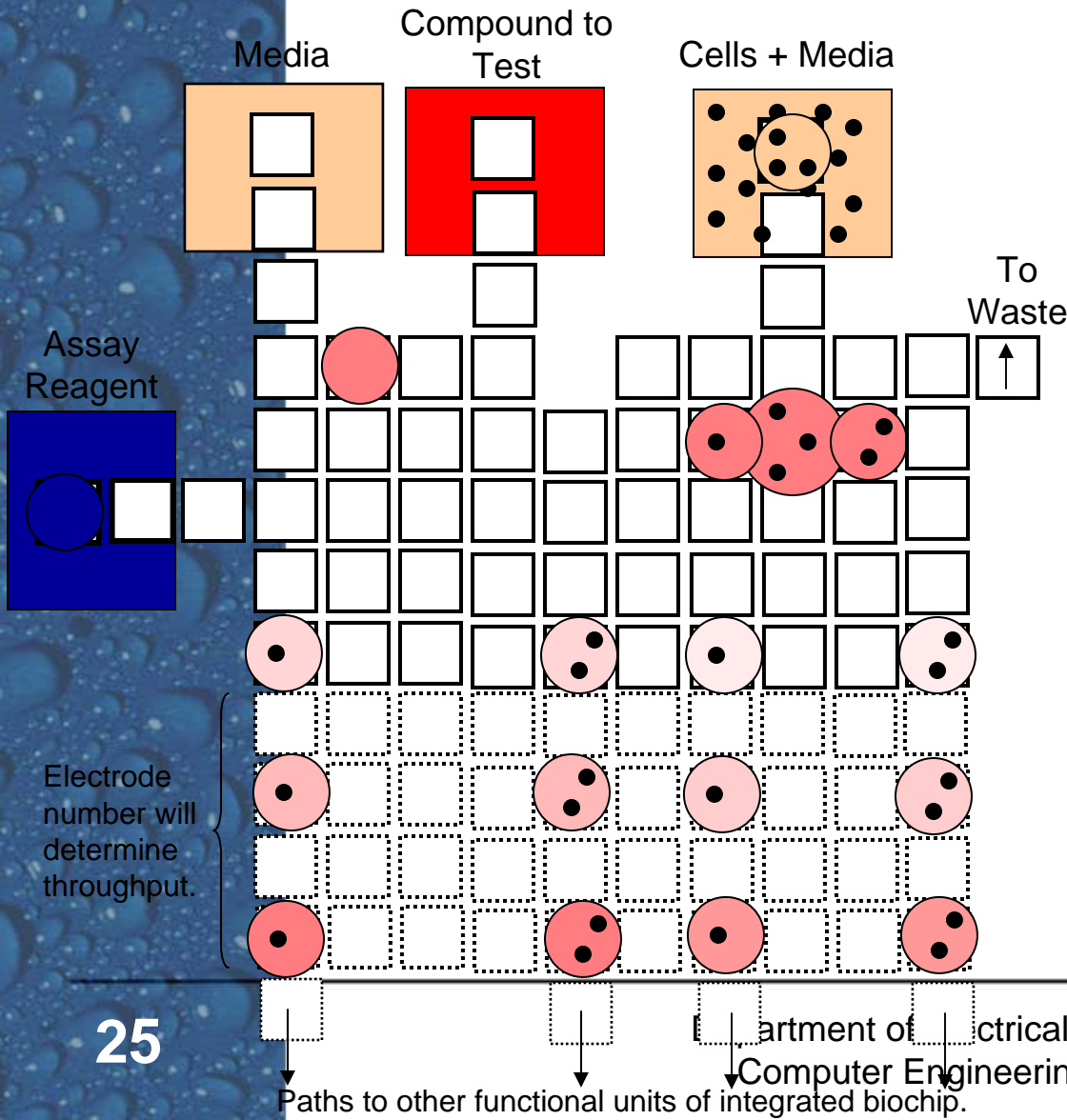


1. Dispense buffer and compound droplets, mix.

2. Split. One droplet stays for further dilution, one droplet gets mixed with cells.

3. Dispense cell solution. Optical absorbance check of concentration (optional). Mix with diluted compound droplet.

Architecture



REPEAT to test multiple dilutions

1. Dispense buffer ~~and compound droplets~~, mix. with previous dilution drop.
2. Split. One droplet stays for further dilution, one droplet gets mixed with cells.
3. Dispense cell solution. Optical absorbance check of concentration (optional). Mix with diluted compound droplet.
4. Split. Both droplets go to holding.
5. Incubate desired length of time.
6. Transport droplets to integrated on-chip functions (lysis, PCR, etc)



Inputs, Outputs, and On-Chip Function

- **Inputs**

- (1) Cell suspension, (2) Cell media for dilutions, (3) Solution of compound to be tested for cytotoxicity, (4) Reagents for the cytotoxicity assay
- If portable: include Lithium ion battery

- **On-chip functions**

- Create droplets of input liquids, split and mix droplets, incubate droplets for programmed length of time, detect intensity of droplet color or presence of stained cells.

- **Outputs**

- Color intensity of droplets or presence of stained cells.



Nucleotide

Pi

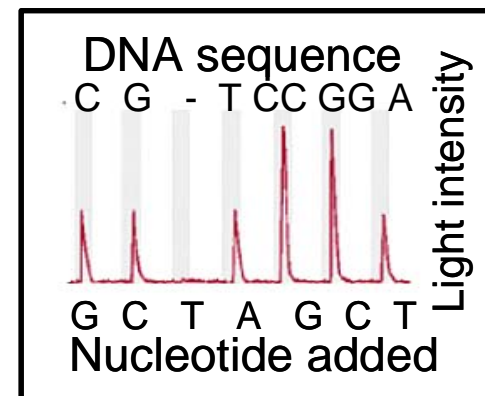
ATP-Sulfurylase

Pi

ATP

Luciferase

562nm



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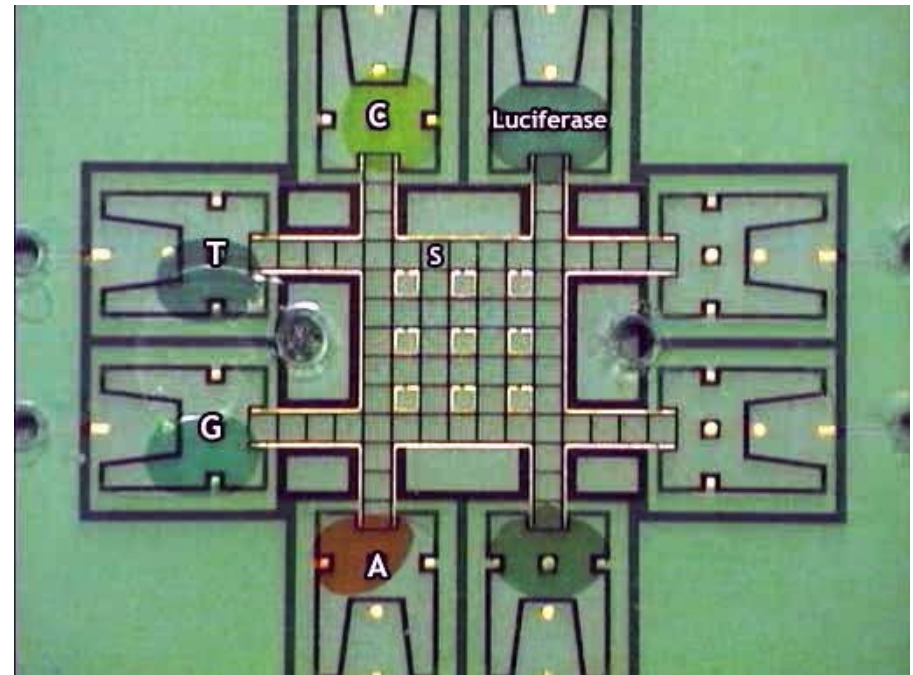


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Example Fluidic Protocol for DNA Sequencing

- Dispense droplets of each dNTP
- Transport droplets to synthesis reaction site and allow to react
- Transport droplets to storage area
- Mix each dNTP droplet with light producing droplet
- Transport combined droplets to detector site

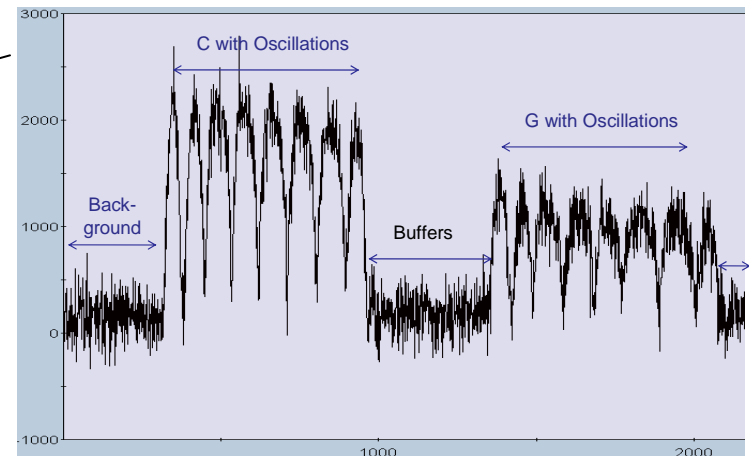
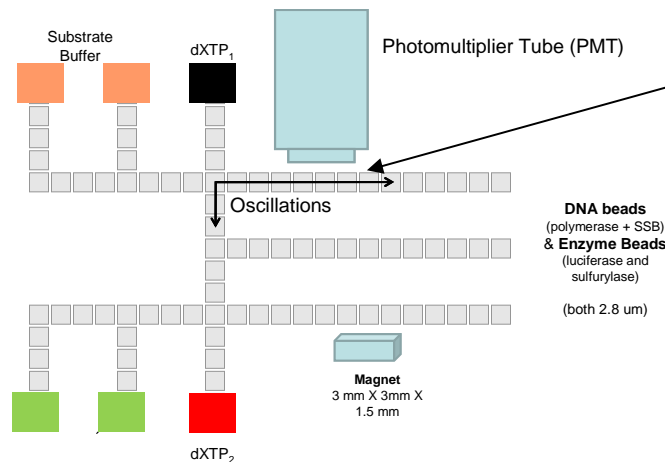


Dyed liquids represent pyrosequencing reagents, droplet volumes are 50 – 100 nL.



On-Chip Pyrosequencing

- Pyrosequencing protocols run at Stanford on ALL platform having six reservoirs and three transport lanes.
- Magnet under bottom lane immobilizes DNA and enzyme beads
- After incorporation, reaction products transported to PMT



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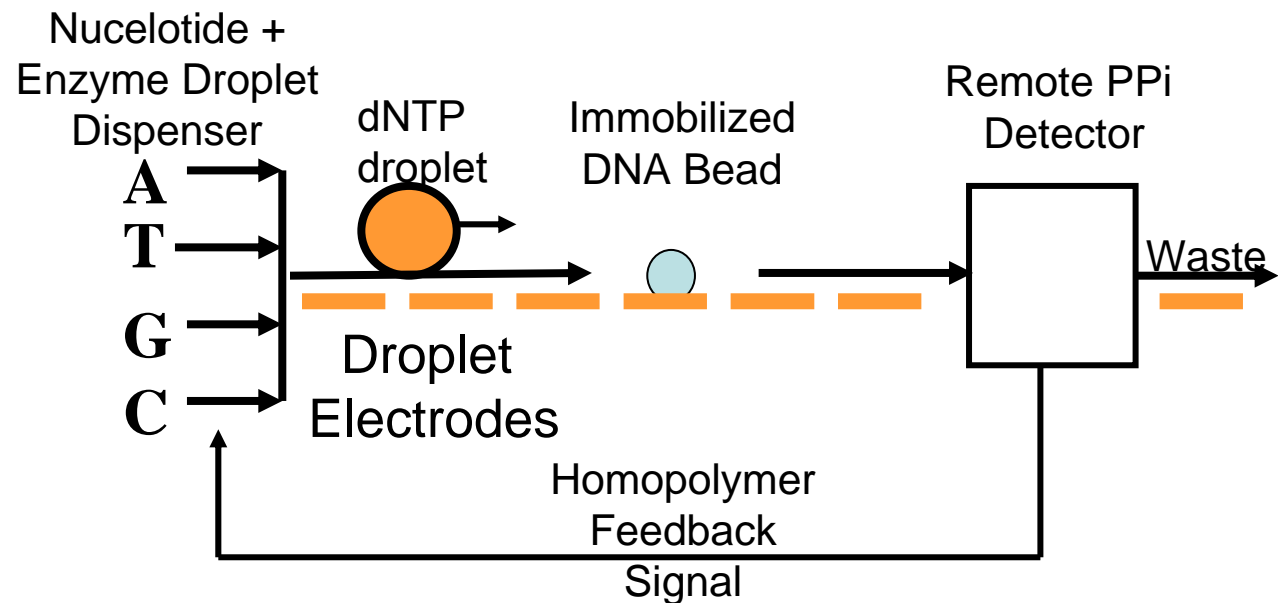


Benefits of Reconfiguration in Pyrosequencing

- Enzyme contamination of chip surfaces may occur
 - Route wash droplets over transport buses for reuse
- Feedback-controlled nucleotide addition for sequencing through homopolymer regions of DNA
- Look-ahead sequencing and voting schemes possible for reliable and high throughput sequencing
- Other benefits of digital microfluidic platform:
 - Continuous droplet dispensing
 - Scalable

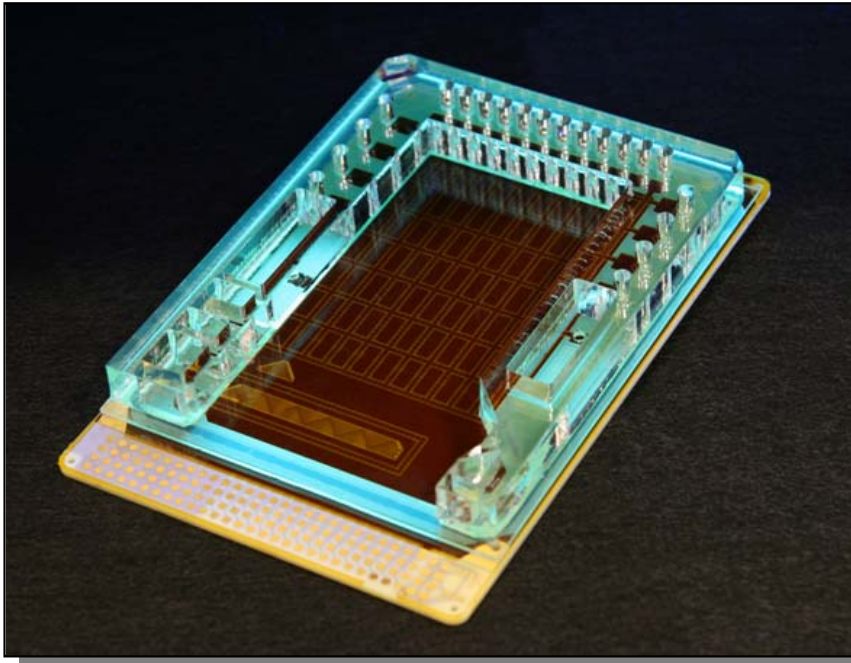


Feedback Controlled Nucleotide Addition For High Fidelity Homopolymer Sequencing



- Deliver dNTP droplet
- If excess light is detected (homopolymer), add more of same dNTP
- Continue adding same dNTP until full incorporation detected
- Else, deliver next nucleotide

World to Chip Interface



- Well-plate interface
 - Easy and familiar loading
 - 384-well spacing
 - Inputs from microliters to milliliters
- Wash/waste reservoirs support 48+ tests
 - Load and go

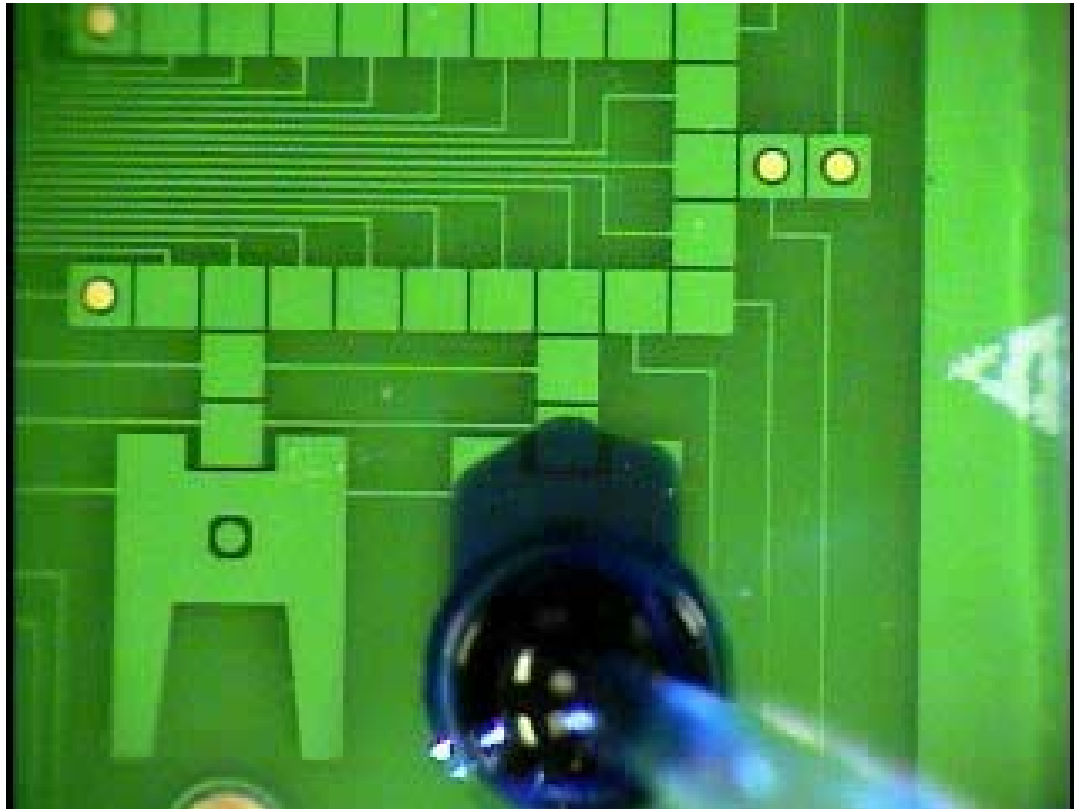
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High Speed Continuous Droplet Dispensing



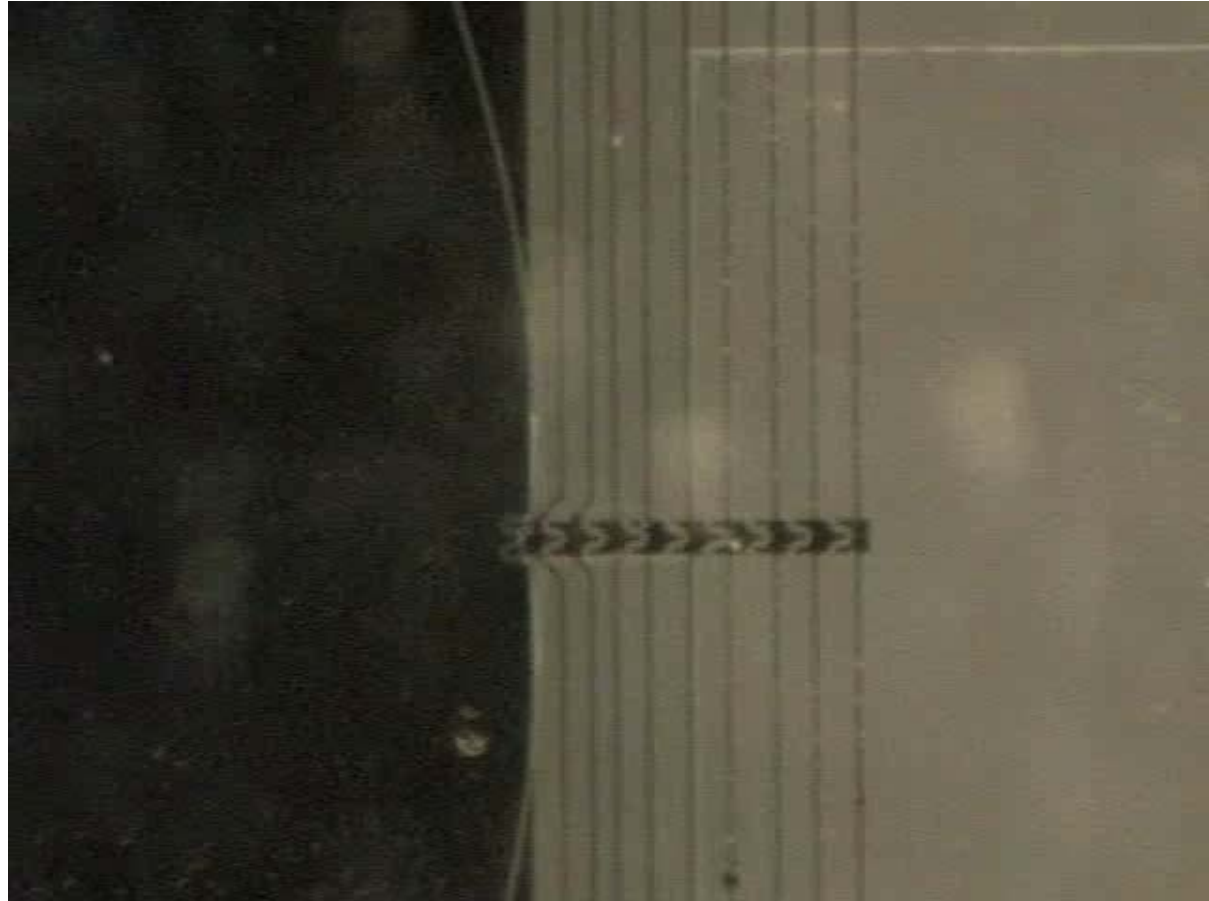
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35 Picoliter Droplet Dispensing



Remarks on Applications

- Extensive biomedical application base can leverage microfluidic operations in an electrowetting system.
- Based on:
 - Shared elemental fluidic operations
 - Reconfigurability
 - No cross-contamination/wash droplets
 - Multitasking by components
 - Few bottlenecks.
- Wide diversity of applications can be parsed into manageable components and assembled into a programmable, reconfigurable and reusable architecture.



Summary and Conclusions

- Reconfigurable microfluidic chips require integration of elemental components to support multiple fluidic functions
 - Requires programmable level for customization
 - Shared fluidic buses (contamination free)
 - I/O compatible with external world
- Electrowetting-based digital microfluidics is good candidate for multifunctional microfluidics
 - Programmability
 - Reconfigurability
 - Multifunctional
- Open issues:
 - On-chip sample preparation
 - Lack of a molecular separation method
 - Capillary electrophoresis
 - Accurate on-chip dilution an open issue
 - Scalable, compatible detector technologies needed



Acknowledgements

- NSF
- NIH
- ECE299 students

