Microfluidics for bio-sample pretreatment
用於生物樣本前處理之微流道系統

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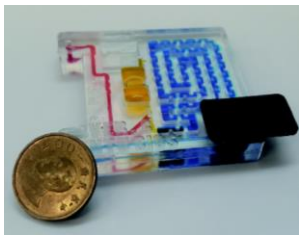
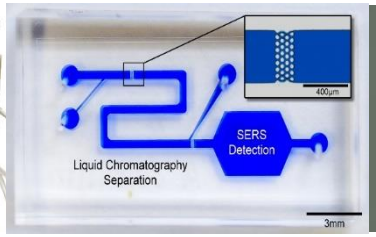
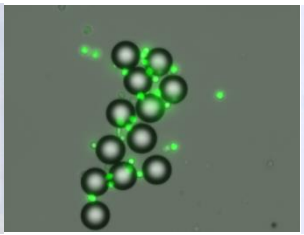
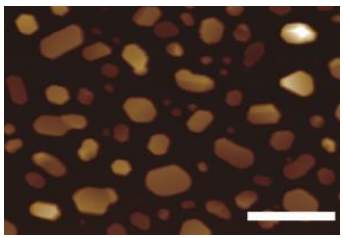
仿生與實驗室晶片導論

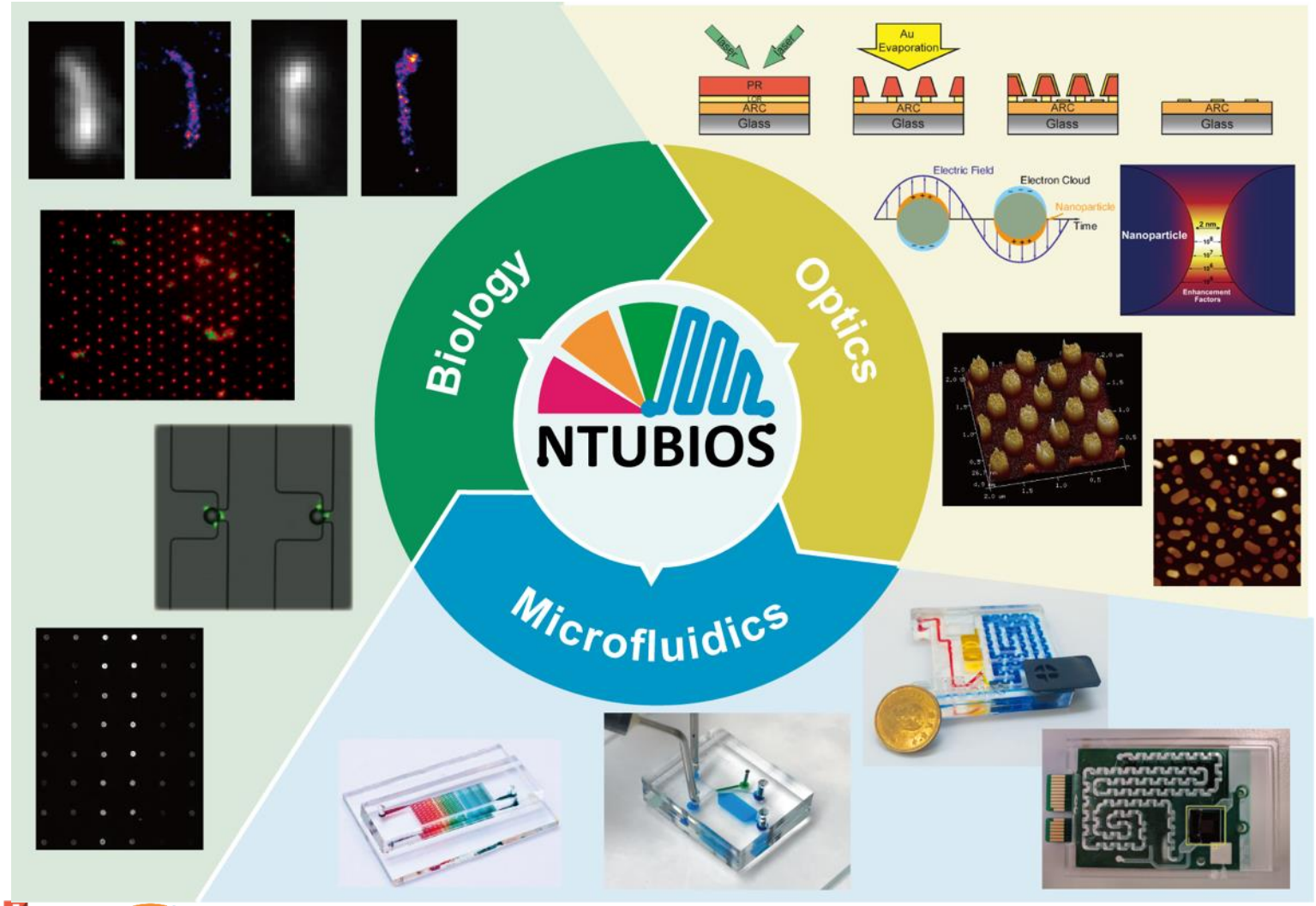






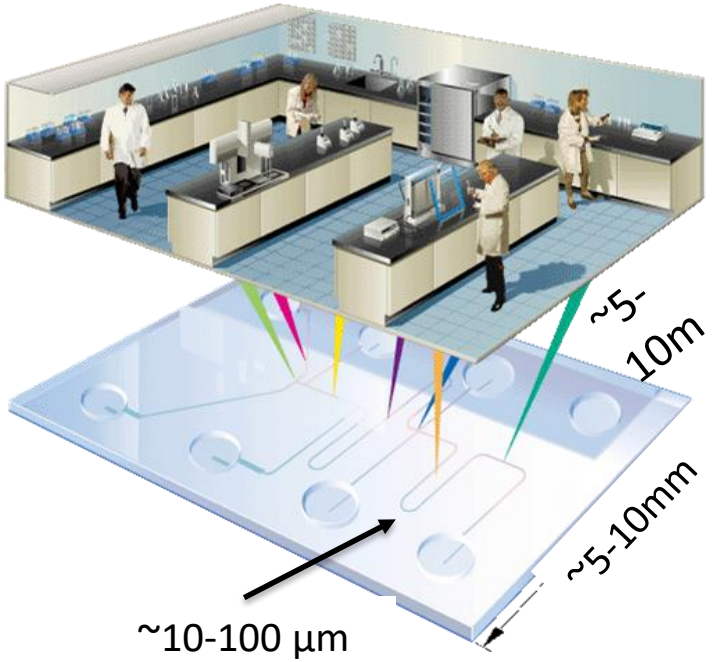


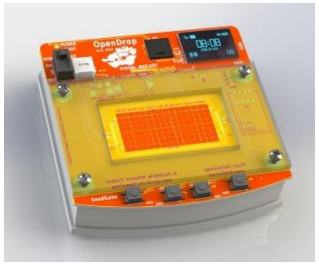
Bio-Optofluidic System (BIOS) Lab



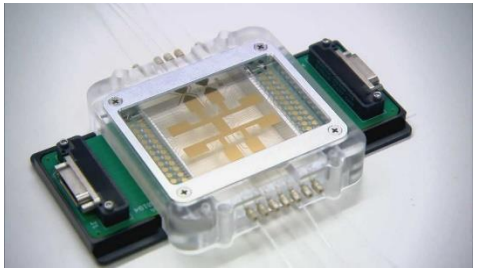
Lab-on-Chip

- Miniaturization and integration of laboratory biochemical processes
  - Microfluidics, micro-sensors and micro- actuators
  - Reduce cost and waste of bio-diagnostics
- **Problems to address**
  - Heterogeneous materials bonding
  - Standard fabrication protocols
  - Optical alignment
  - Buffer condition
  - Leakage
  - Packaging

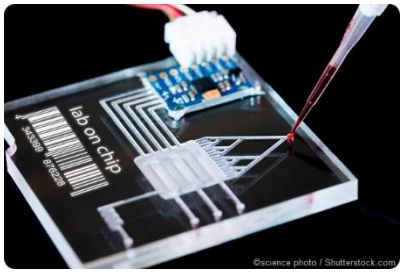




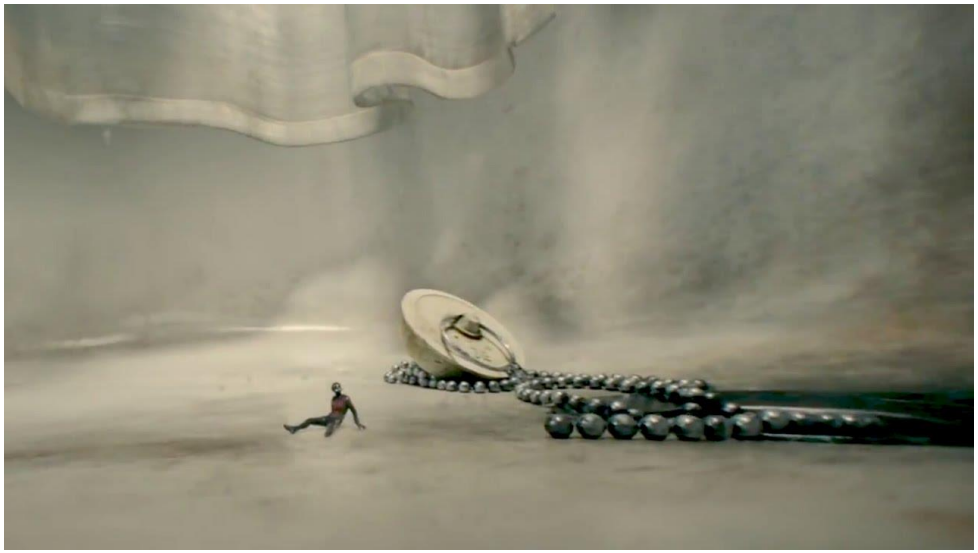
(Ref: OpenDrop)



(Ref: Sandia lab)

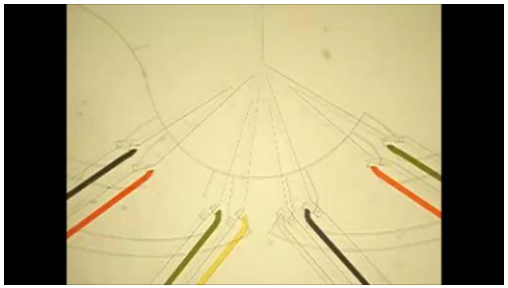


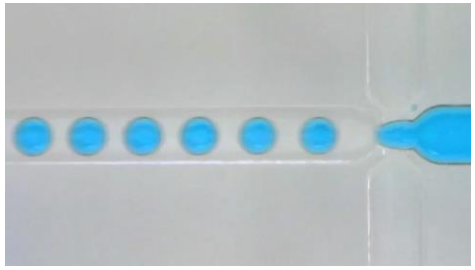
Examples of miniaturization

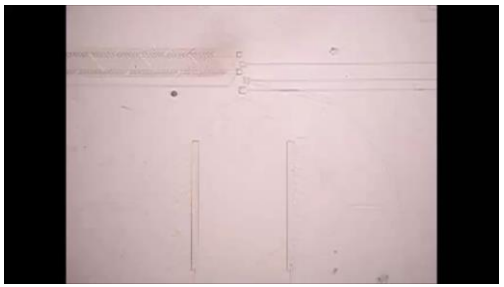


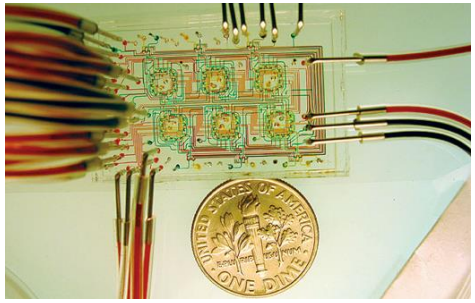
Microfluidics

- Technology of manipulating and controlling fluids, between 10^{-6} to 10^{-12} L in ~10-100 μm microchannel
- It is a multidisciplinary field from the development of analytical chemistry and microelectronic fabrication technologies



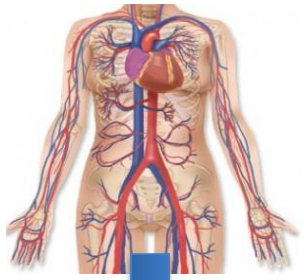


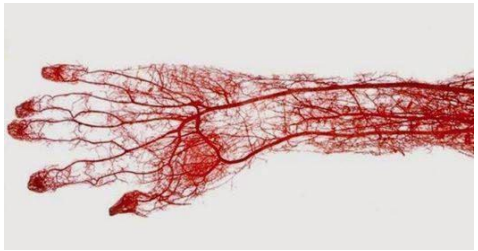


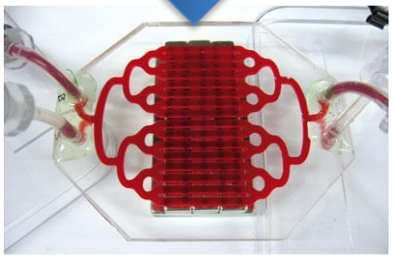


(Science, 309, 137-140, 2005)

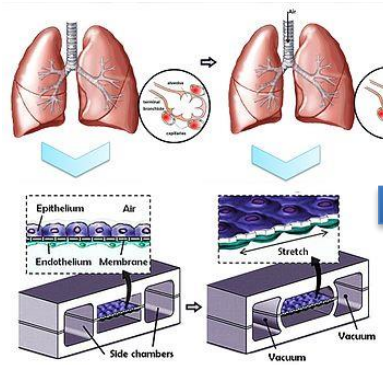
- Microfluidics in the human body?
  - Blood vessel
  - Organs on Chip

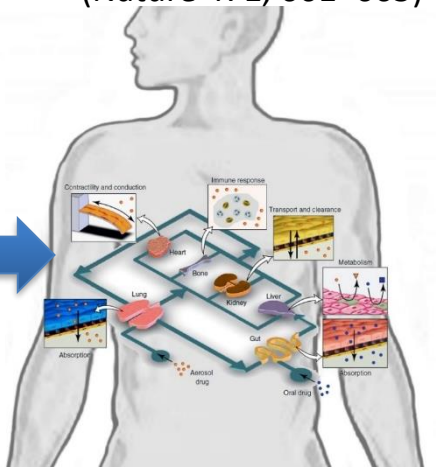






(Nature 471, 661–665)

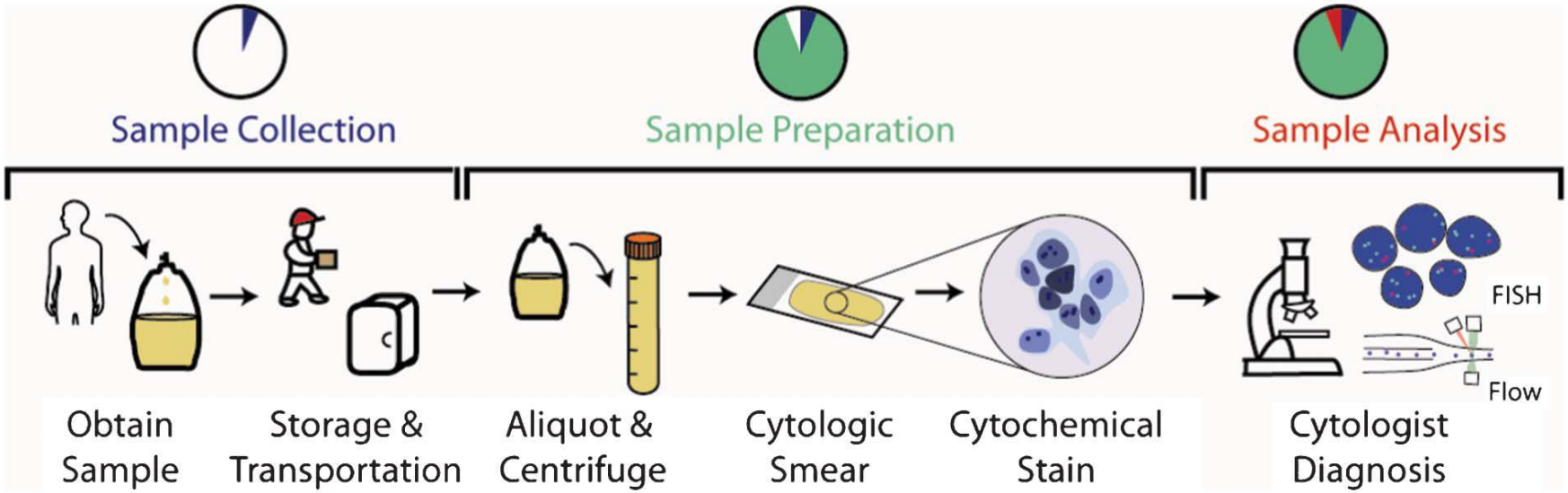




Microfluidics for whole blood process and detection

Current Bio-Sample Process Problem

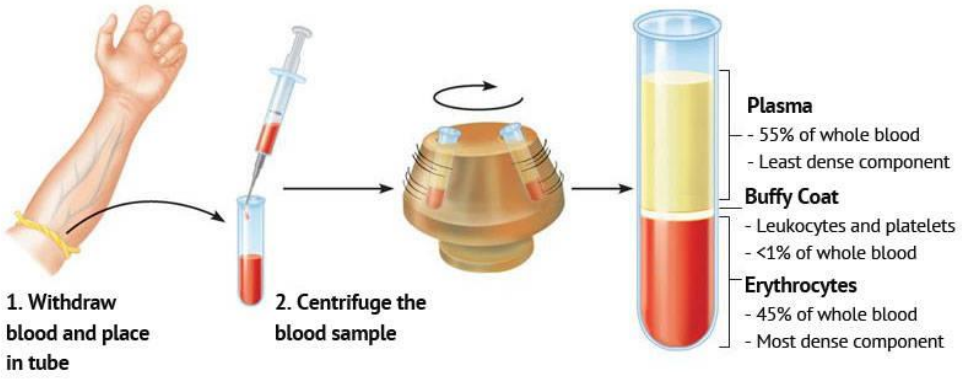
- Sample preparation: centrifugation, cell fixing, washing and cytochemical staining
- The quality of biomarker detection will be affected by sample preparation



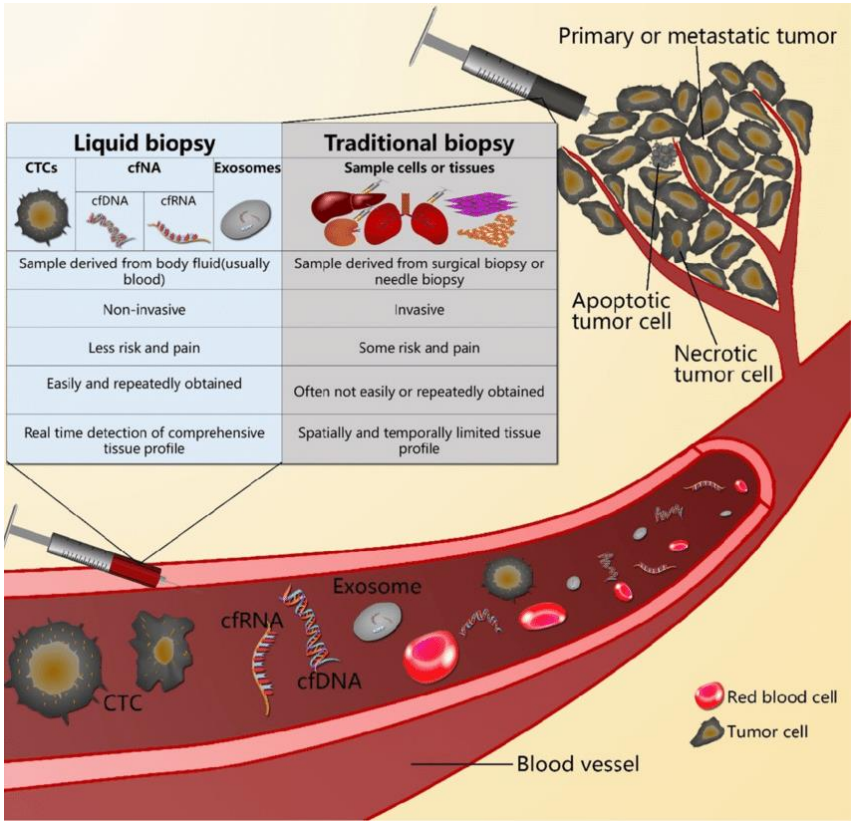
A platform to efficiently perform sample preparation and in-situ analyte detection!!!

Diseases diagnosis using whole blood

- Whole blood consists of 54.3% plasma, 45% RBCs, 0.7% WBCs and platelets

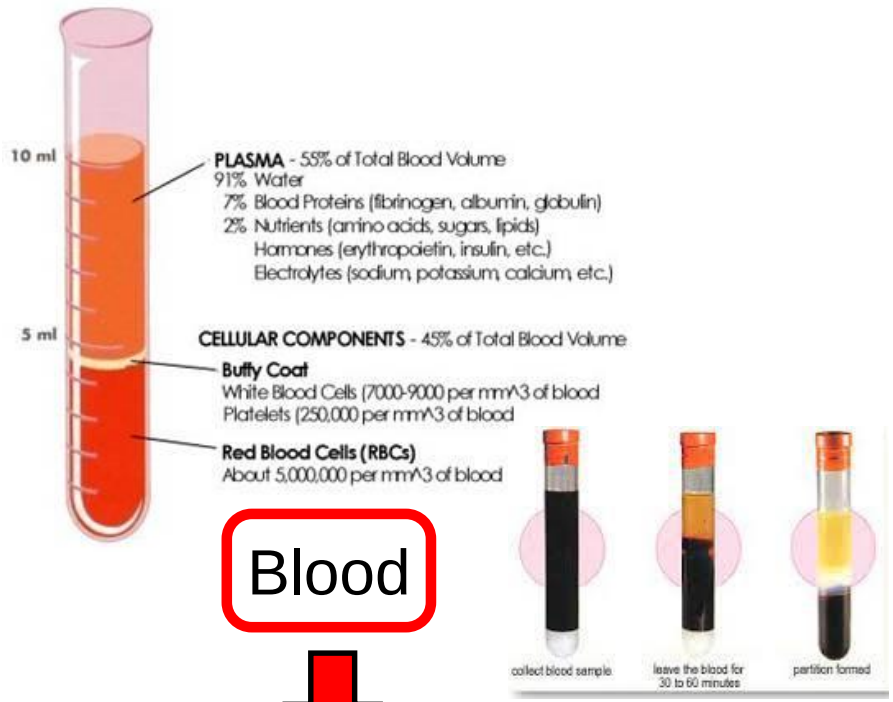


- **Biomarkers in whole blood**
  - Protein, DNA and metabolites=> diabetes, mutation disease
  - Immune cells or tumor cells=> inflammation, cancer
  - Bacteria => sepsis, infection



(Journal of Cancer 9(18):3417-3426)

Blood Separation Methods



Blood

- Plasma Separation
- Cell Separation

Label-free

No external field

- Micro-scale filters
- Hydrodynamic filtration
- Deterministic lateral displacement
- Inertial
- Gravity and sedimentation
- Aqueous two-phase systems

With external field

- Acoustophoresis
- Magnetophoresis
- Optical
- Dielectrophoresis

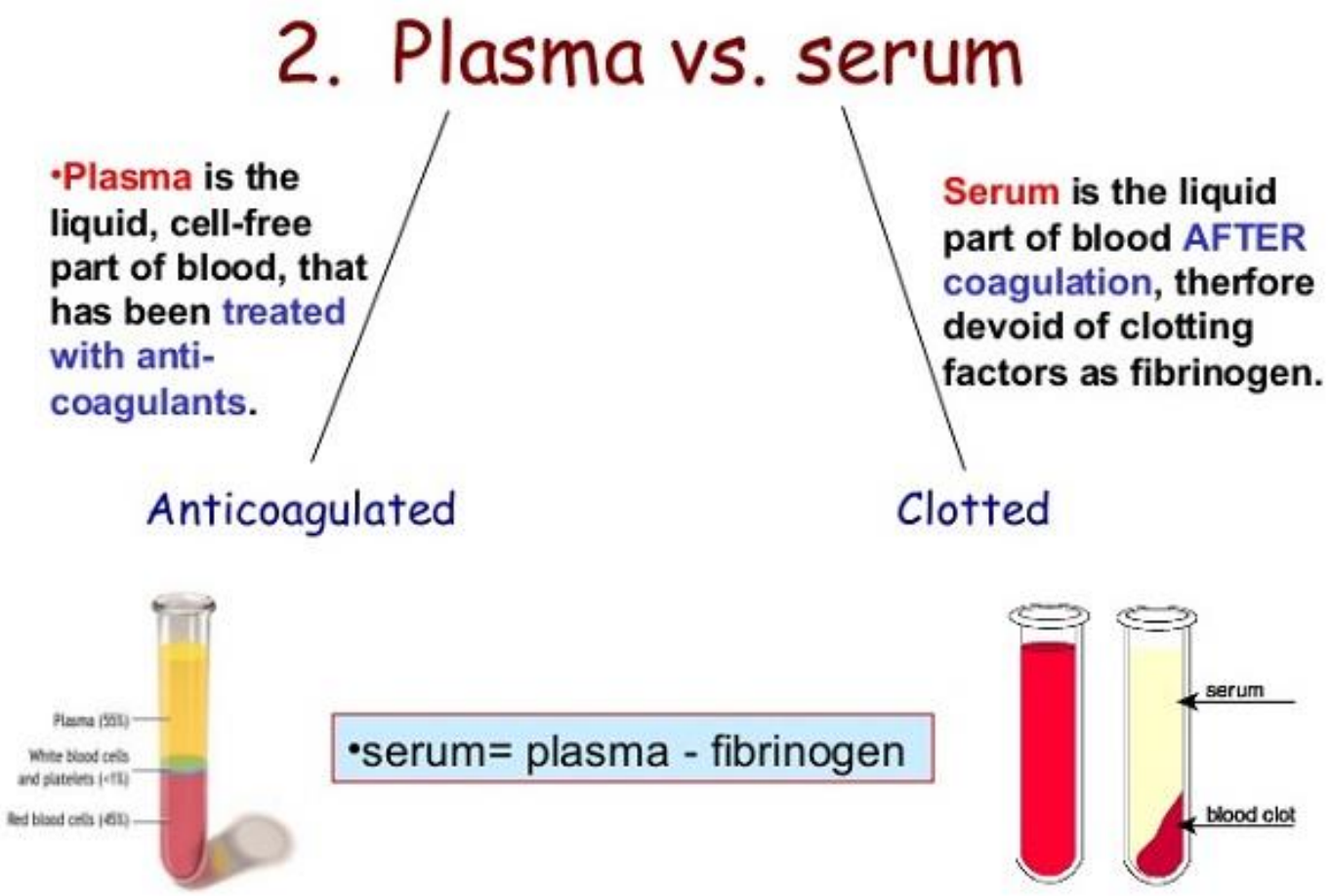
Other format

- CD
- Paper

Labeled

- Fluorescence-activated cell sorting
- Magnetic-activated cell sorting

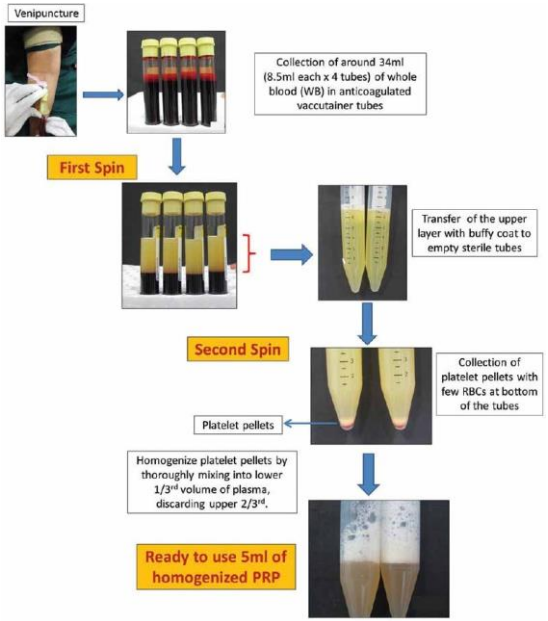
Difference between Plasma and Serum



http://www.microbiologynotes.com/differences-between-serum-and-plasma/

Blood Separation Method

- Centrifuge



- Platelet-Rich Plasma (PRP) treatment

PRP刺激細胞修復 Kobe 伍茲都說讚

2016年05月12日 傳送 讚 99 G+

Kobe曾接受PRP治療膝傷。資料照片

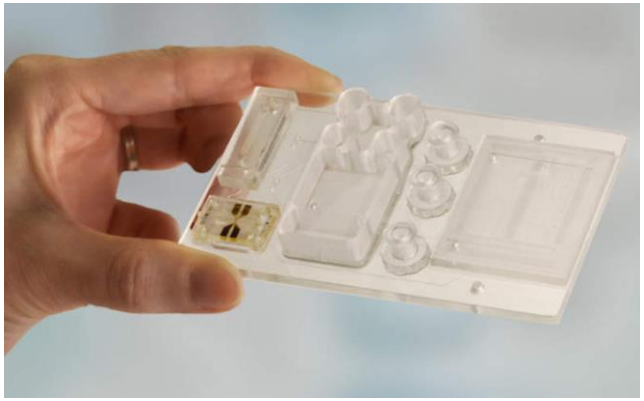
【王翊亘／綜合報導】PRP治療全名為platelet-rich-plasma，高濃度血小板血漿治療，許多國外運動名將都曾使用過此方式治療患處，包括高爾夫前球王老虎伍茲(Tiger Woods)、男網球星納達爾(Rafael Nadal)以及今年剛從NBA退休的Kobe(Kobe Bryant)。

PRP治療是透過使用患者自身的血液，用離心機把血小板分離出來，再把血小板注射到受傷部位，刺激細胞修復，手術過程1小時完成。技術原理為將血漿濃縮，讓血液中的血小板破裂，釋出生長因子，促進軟組織修復和再生。

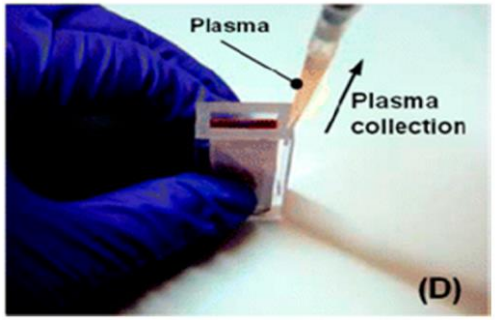
這種治療方式一開始是使用在醫學美容，後來改應用在關節、韌帶的治療修復，在歐美已存在許久，Kobe在2011年時，就曾到德國進行PRP治療。在台灣若進行PRP治療，價格約在1萬至2萬元間，配合手術使用效果最佳，林智勝2013年進行左膝手術時，也曾在縫合的韌帶上施打PRP，幫助加速癒合。

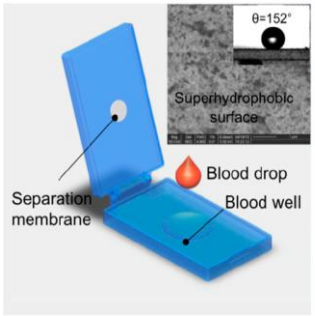
Microfluidics for whole blood processing

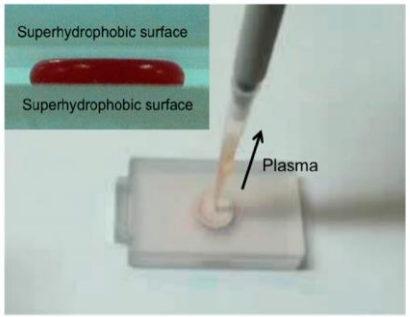
- PDMS/PMMA-based, CD-based, paper-based microfluidics
  - Low sample requirement & shorter sample process time
  - Low power consumption or power-free
  - Multiple bio-components separation
  - Integrate optical or electronic sensors for in-situ biomolecule detection



Microfluidic cartridge
(NOWDiagnostics)



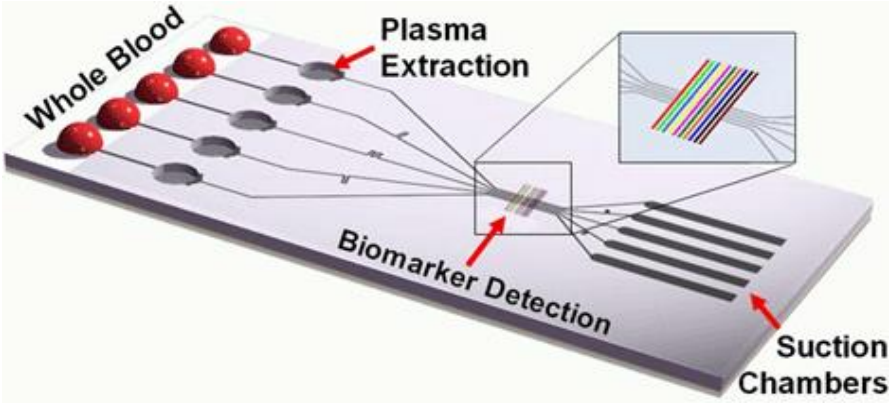




(Liu et. al. Anal. Chem. **2013**, 85, 10463–10470)
(Liu et. al; Lab Chip **2016**, 16, 553)

Microfluidics for Whole Blood Process

- Why microfluidics for whole blood process?
  - Cost-effective, portable, disposable
  - Low sample volume
  - Fast response
  - Multi-functional
- Four important parameters:
  - Dilution ratio
  - Throughput
  - Purity
  - Yield

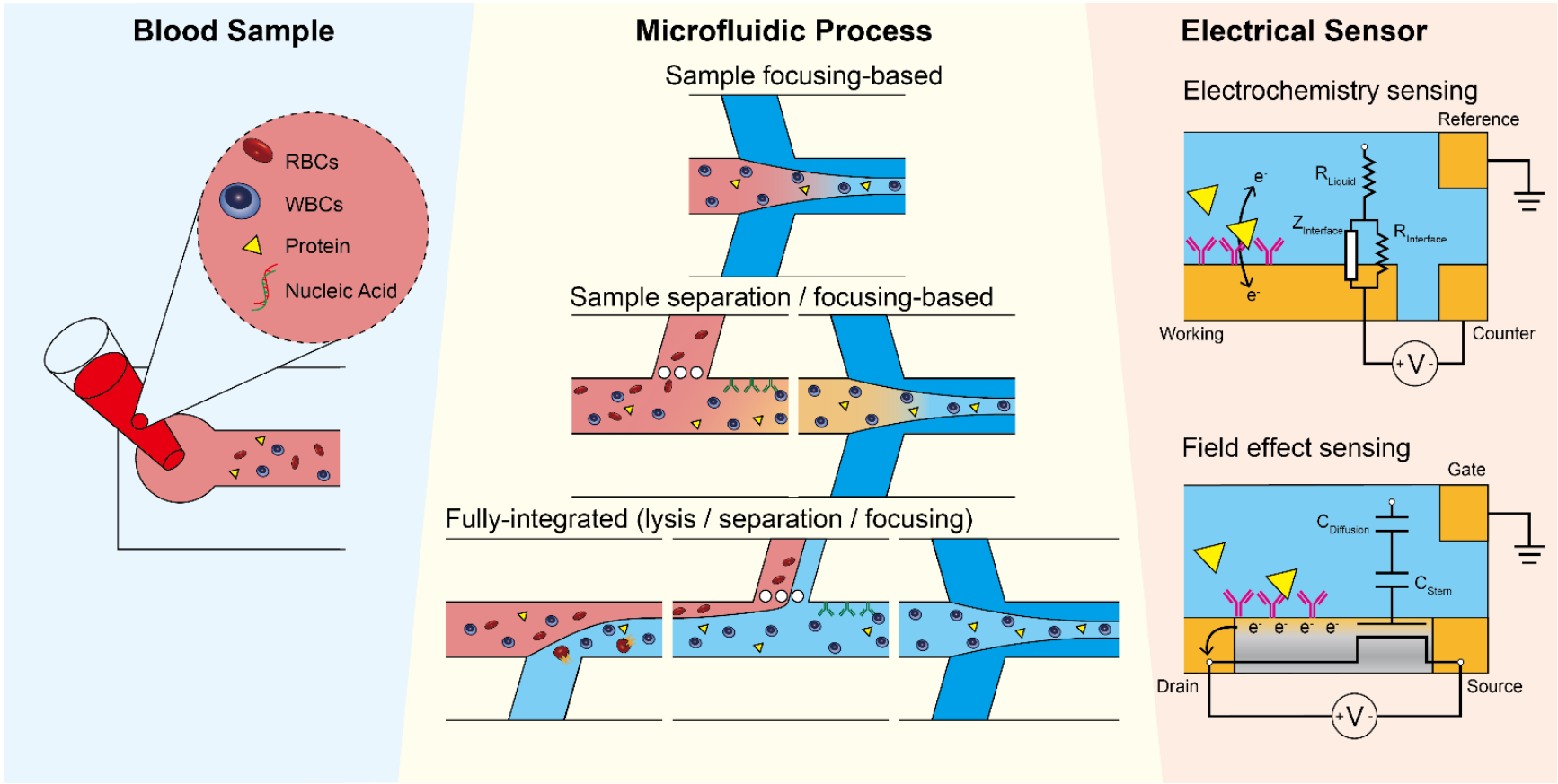


L. Lee Group at UC Berkeley

Integration of microfluidics with electrical sensors for whole blood analysis

- Why integrating microfluidics with electrical sensors?

(1) on-chip sample process; (2) multiplicity; (3) low sample volume;
(4) fully automated system with embedded signal processing



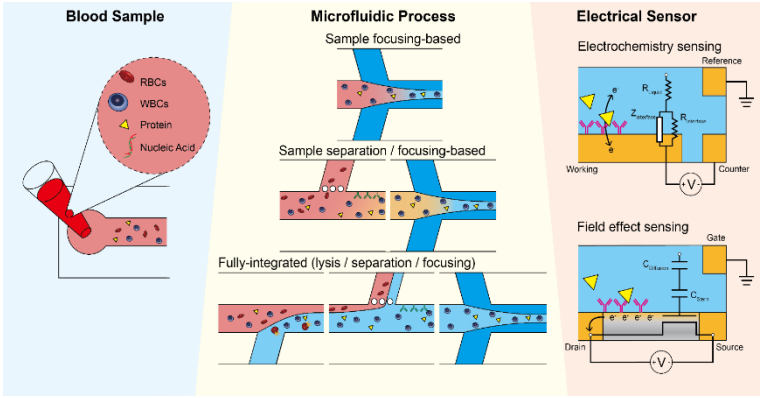
(Kuan et. al., Analytical Methods, 2020)

Microfluidics for blood cell lysis and focusing

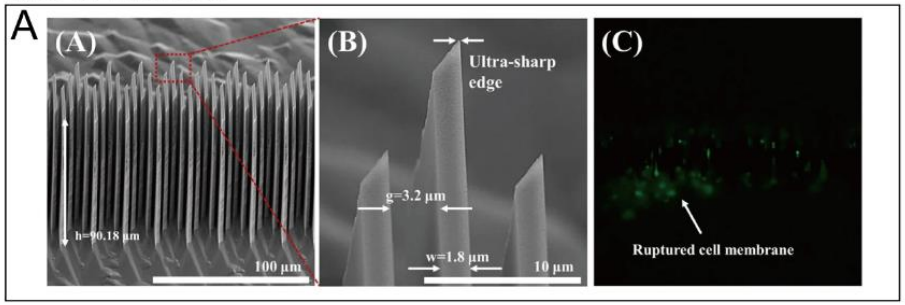
- Mechanical lysis: nanoblades, microbubble
- Chemical lysis:

(1) hypotonic solution (DI water) for selective lysis of RBCs and WBCs

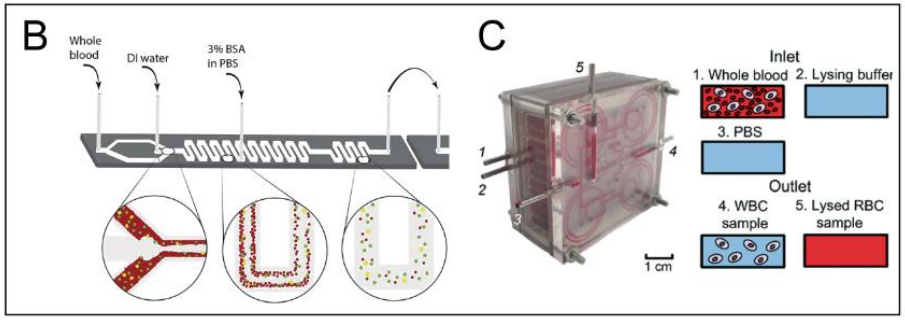
(2) lysing buffer to eliminate red blood cells



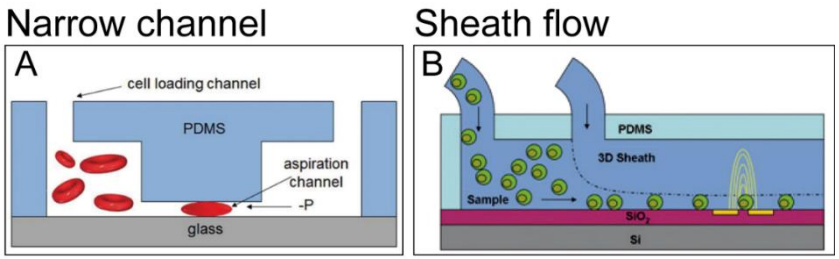
Mechanical



Chemical

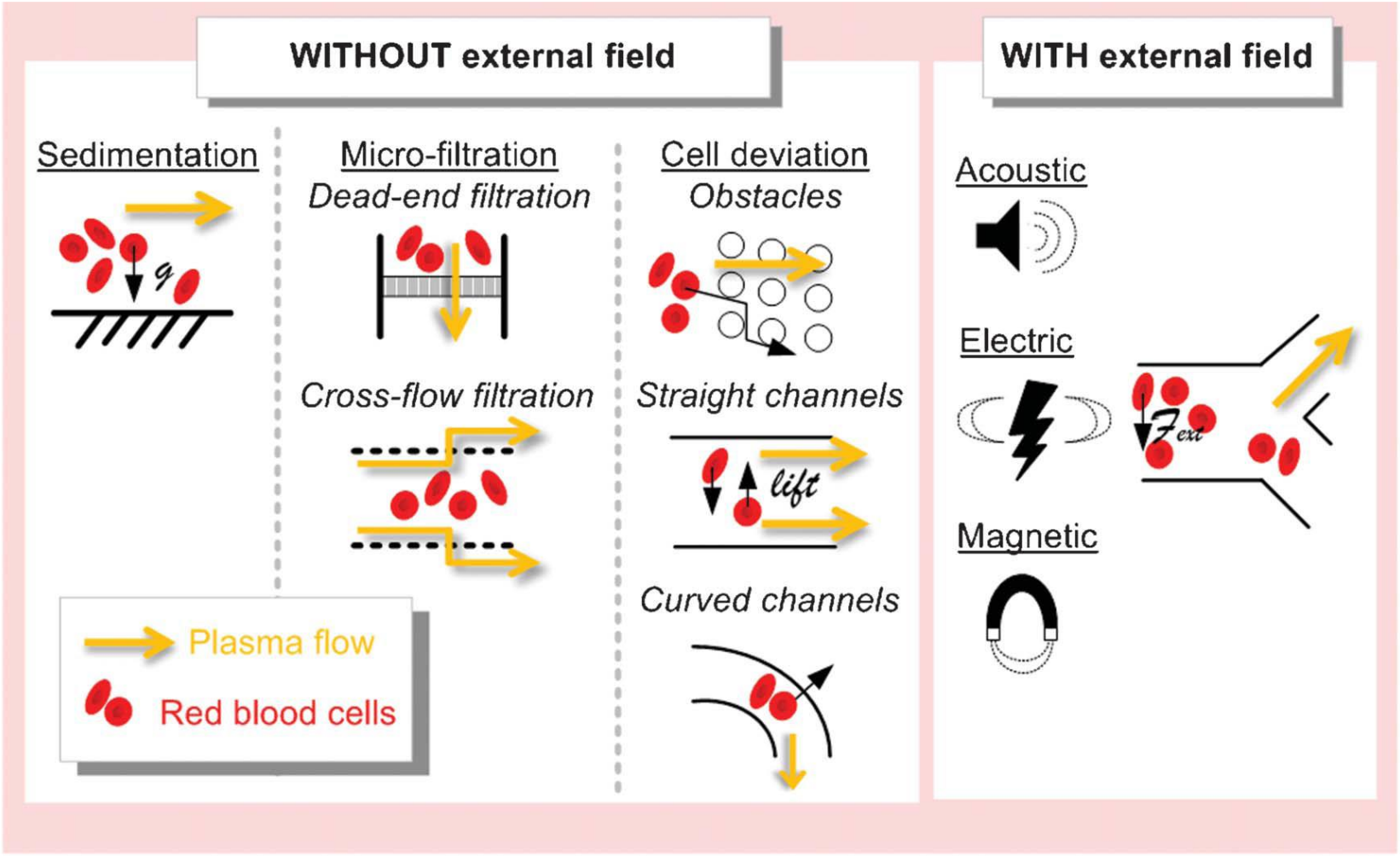


- Narrow channel focusing
- Sheath flow focusing



(Kuan et. al., Analytical Methods, 2020)

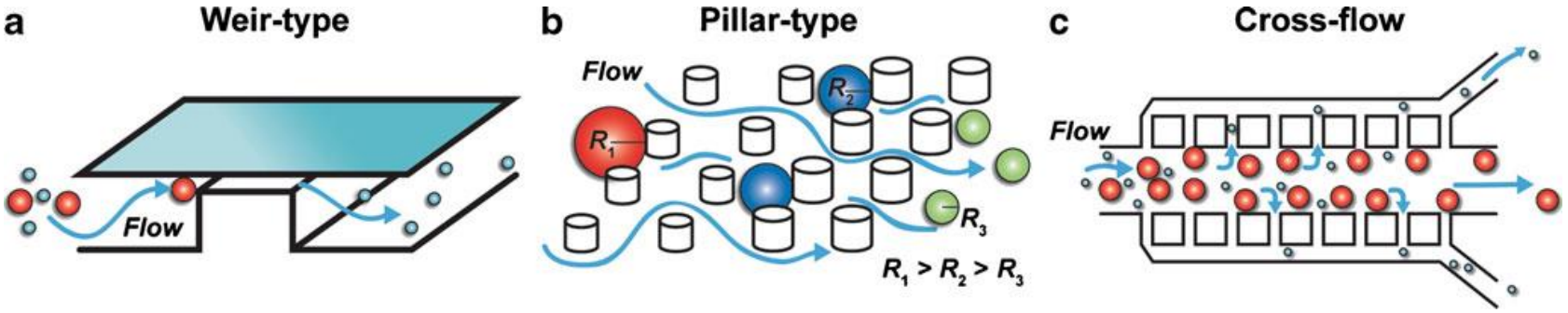
Microfluidics for blood cell separation



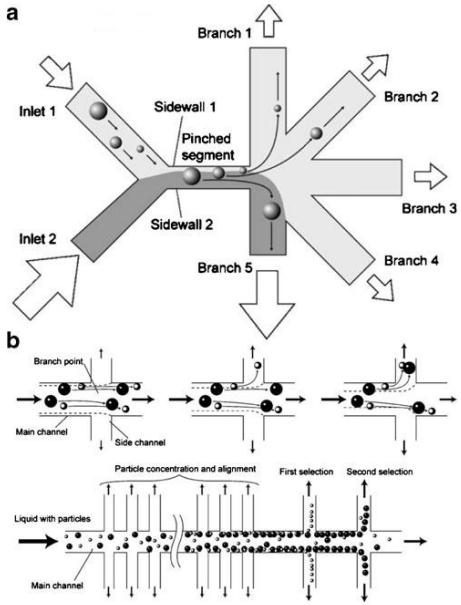
Do we want blood plasma or blood cells?

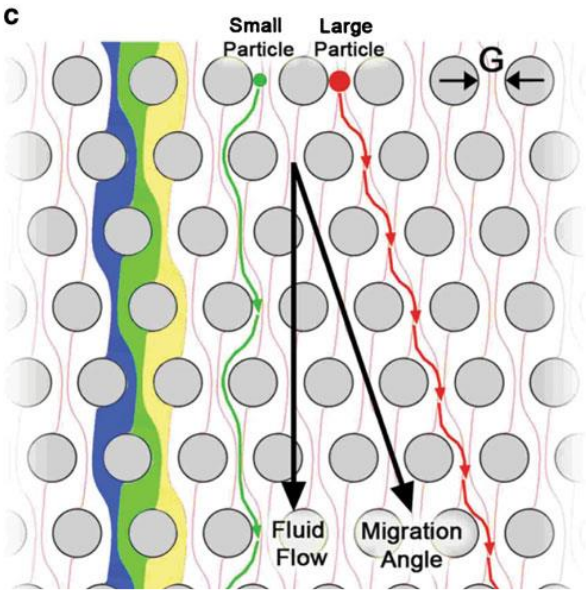
Microfluidics for blood cell separation

- Micro-scale filters (Size, deformability)

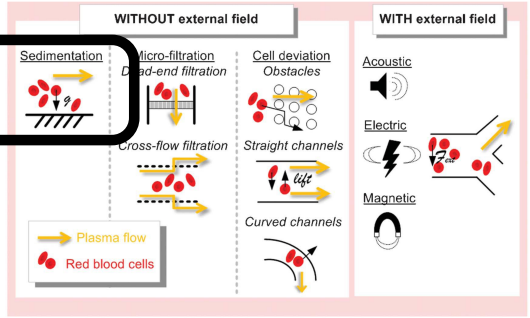


- Hydrodynamic filtration (size, shape)
- Deterministic lateral displacement (DLD) (size)

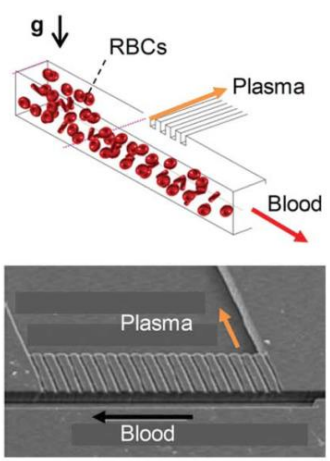
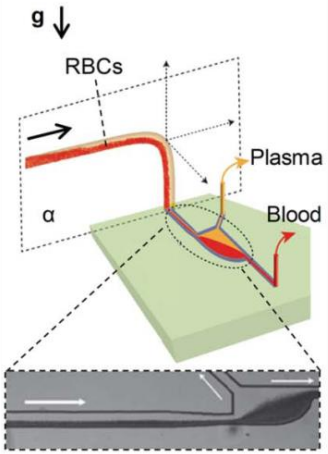
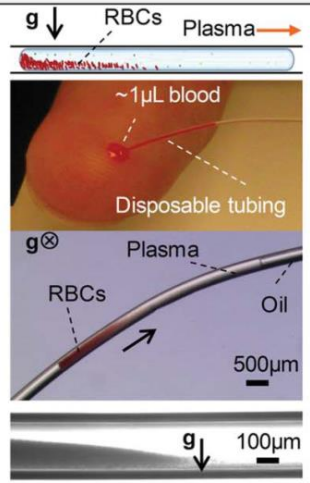
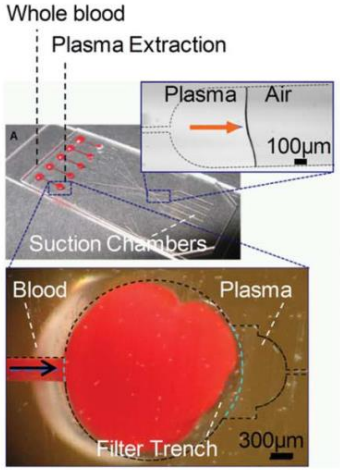




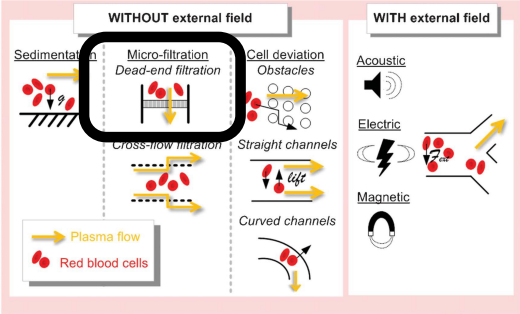
Microfluidics for blood cell separation - Sedimentation



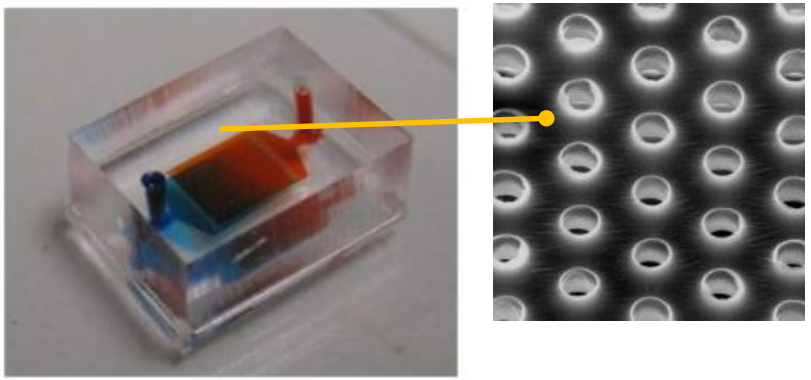
- Based on gravity
- Blood cell separation
  - May not be suitable
- Blood plasma separation
  - The easiest method

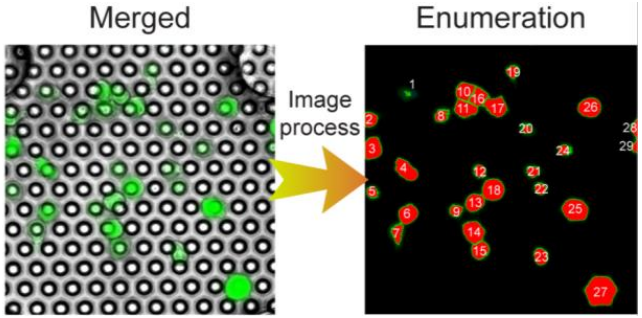
A - Sedimentation with Cross-Flow Filtration	B - Sedimentation with Back-Facing Step	C - Sedimentation in a Plug	D- Sedimentation in Trenches
			
Dilution ratio: None Flow rate: 0.1 μL/min Purity: N/A Yield: 20%	Dilution ratio: 1:5 Flow rate: 15 μL/min Purity: 99% (Hematocytometer) Yield: 66%	Dilution ratio: 1:5 Flow rate: 0.5 μL/min Purity: 100% (Image Analysis) Yield: 60%	Dilution ratio: None Flow rate: 0.83 μL/min Purity: 100% (Image Analysis) Yield: N/A

Microfluidics for blood cell separation - Micro-Filtration



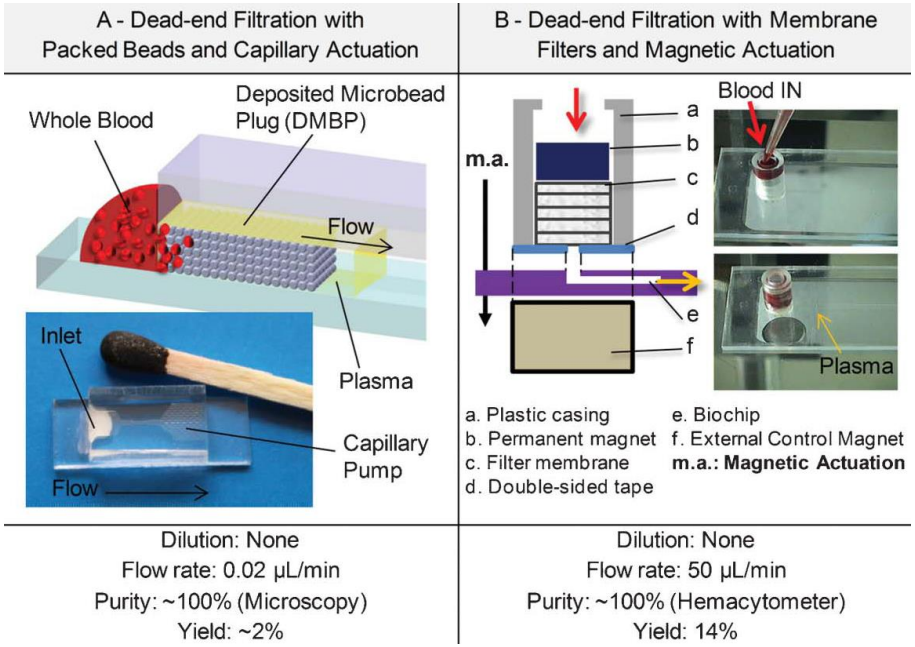
- Based on cell size
- White blood cell separation



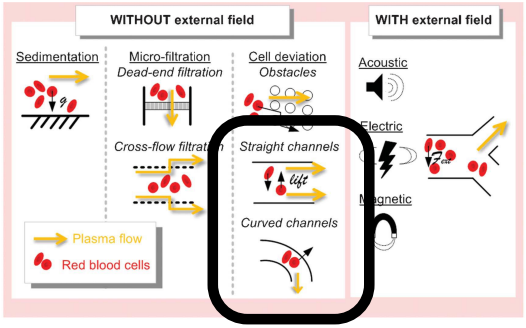


K. Kurabayashi Group, U Michigan

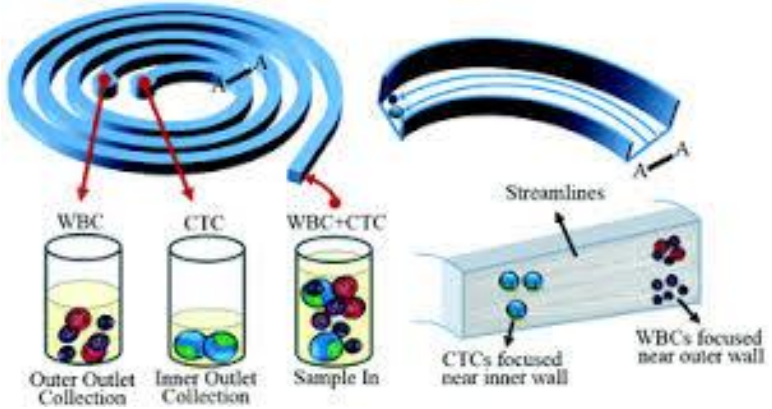
- Blood plasma separation

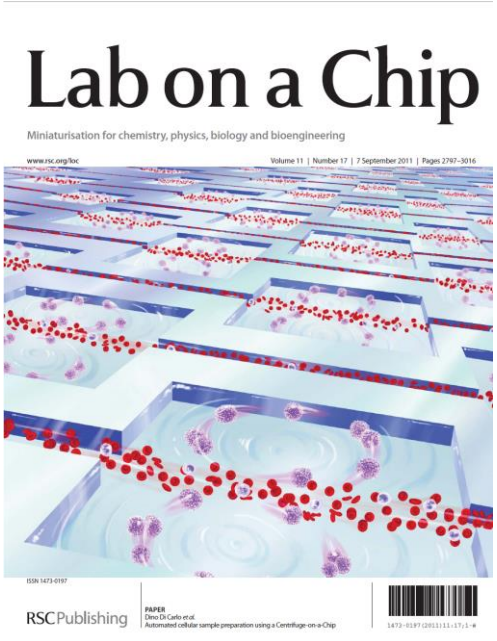


Microfluidics for blood cell separation - Cell Deviation

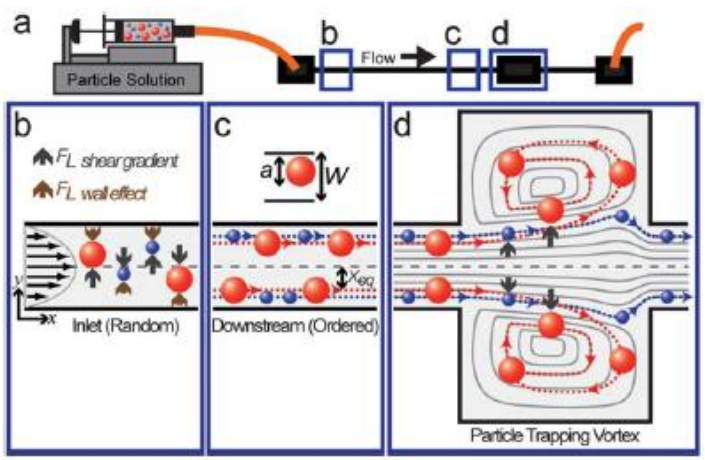


- Blood Cell Separation



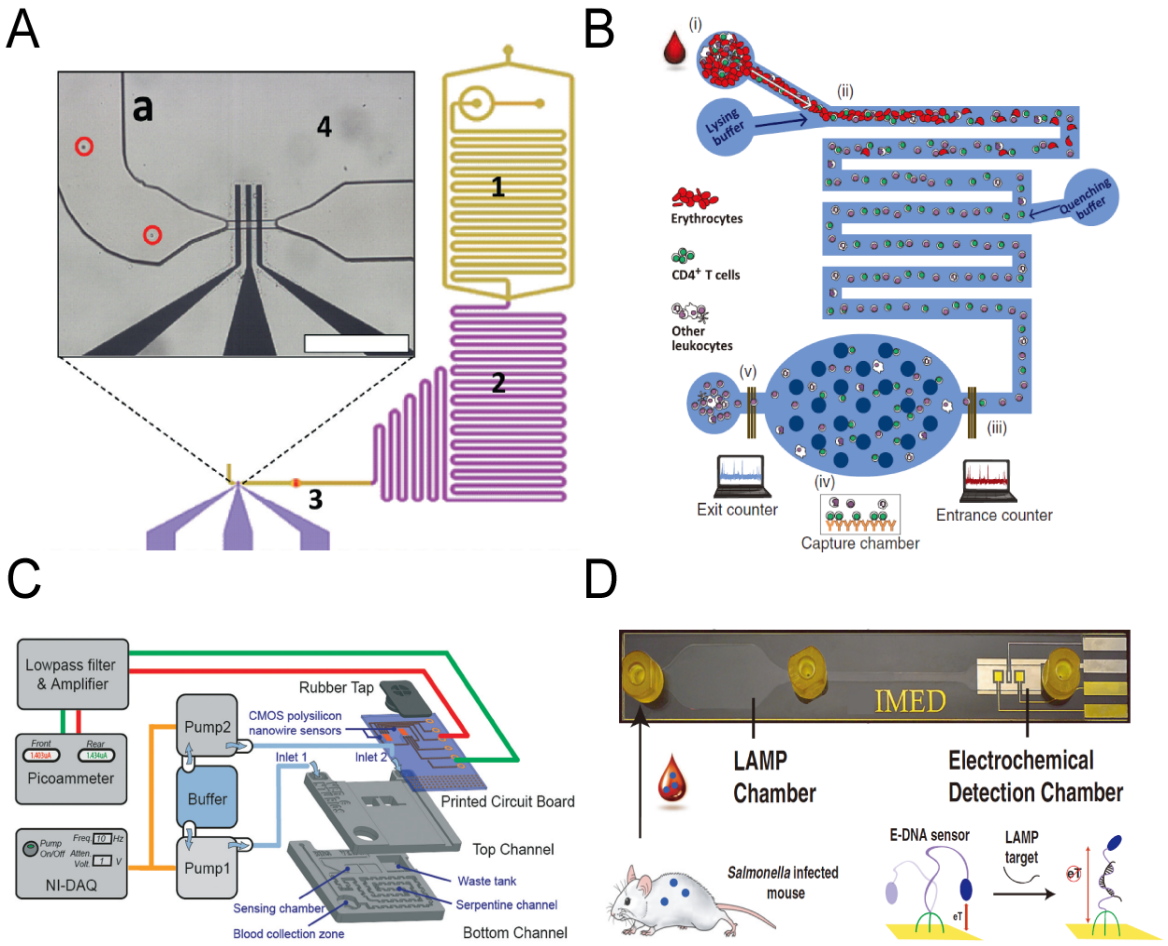


- The balance of shear gradient and wall effect life force



The fully integrated microfluidics with electrical sensors for whole blood analysis

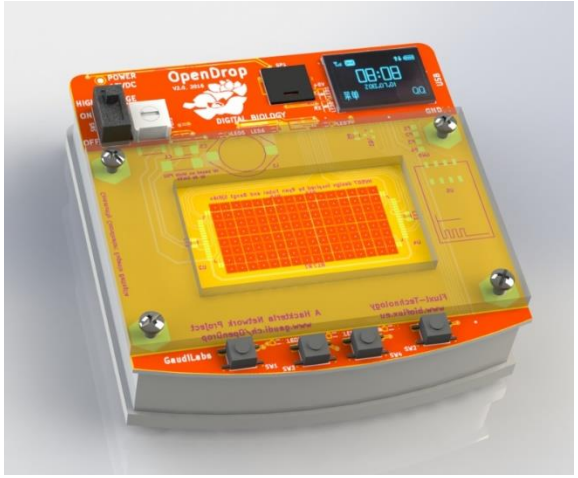
- Microfluidics
  - Cell focusing
  - Cell lysis
  - Cell guiding
  - Cell separation
- Electrical sensors (Electrode, ISFET, EIS)
  - Cell counting
  - DNA detection
  - metabolite detection



(Kuan et. al., Analytical Methods, 2020)

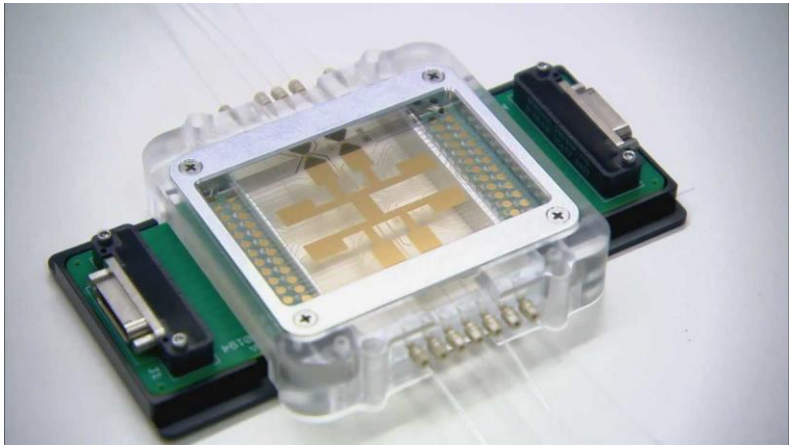
When Microfluidics meet Electronics or Optics…

- Problems need to solve…
  - Bonding of different materials
  - Buffer conditions
  - Leakages
  - Packaging
  - Optical alignment
  - Standard fabrication protocols

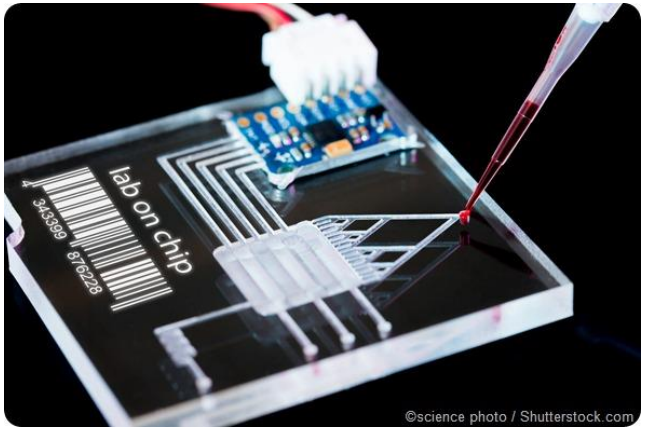


(Ref: OpenDrop)

https://www.youtube.com/watch?v=o9n0tfutOp4

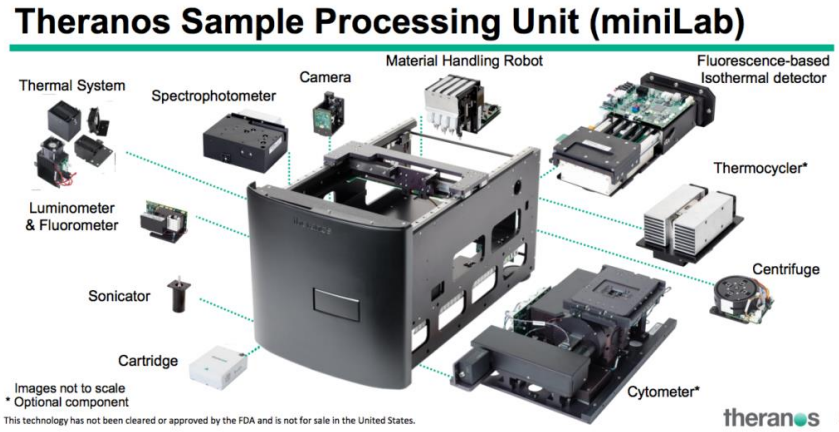


(Ref: Sandia lab)



Case Discussion: Bad Blood

- Elizabeth Holmes, the CEO of Theranos
- Theranos was a health-care company, but subsequently infamous for its false claims to have devised blood tests that only needed very small amounts of blood



https://www.youtube.com/watch?v=wtDaP18OGfw

- Q1.Issues when minimizing the sample volume?
- Q2.Issues when integrating multiple modules into one system?

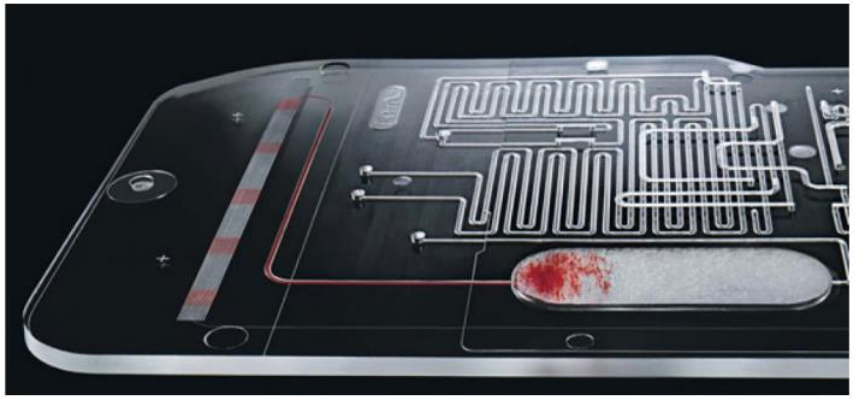
mChip from Harvard for hepatitis C and HIV

Innovation: A Blood Test on a Chip

Claros Diagnostics has created the mChip, which can produce accurate test res
10 minutes.

By Christine Lagorio-Chafkin *Senior writer, Inc.* @Lagorio

WRITE A COMMENT



Complete blood counting (CBC) for malaria detection (e.g. Sight Diagnostics' OLO analyzer)

microRNA detection for cancer diagnosis (e.g. Toshiba)

東芝、血液1滴から2時間で"がん13種を99%検出"できる検出技術

佐藤 岳大　2019年11月25日 14:42

リスト　B!

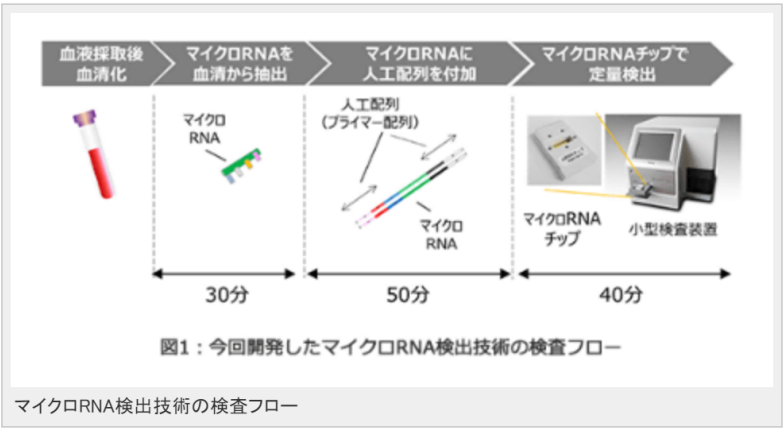
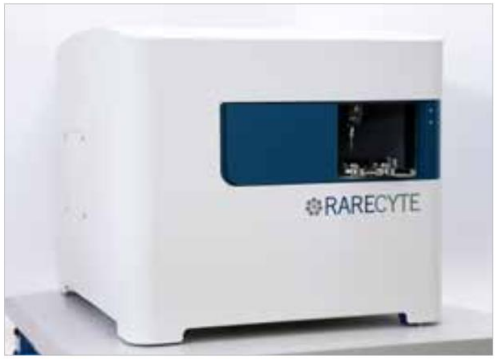


図1：今回開発したマイクロRNA検出技術の検査フロー

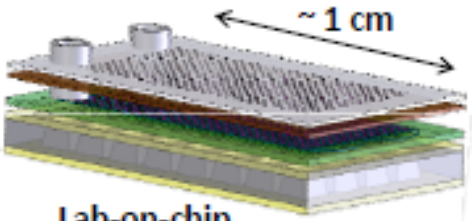
マイクロRNA検出技術の検査フロー

Circulating tumor cell (CTC) detection (e.g. CellSearch, Leica)



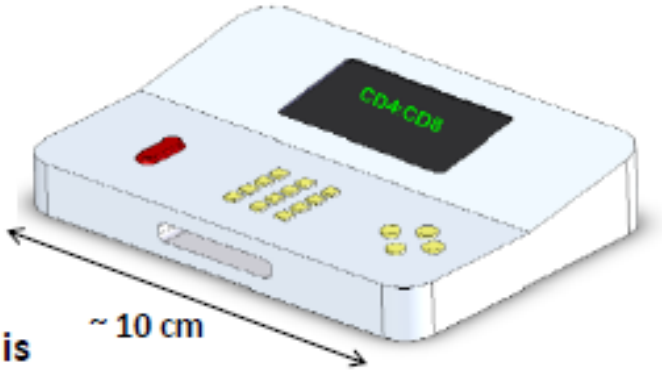
Point-of-care (POC) devices

1. Blood is obtained from the patient's finger



2. The sample is being processed by the biochip

3. The biochip is placed inside the data analysis device for blood analysis.

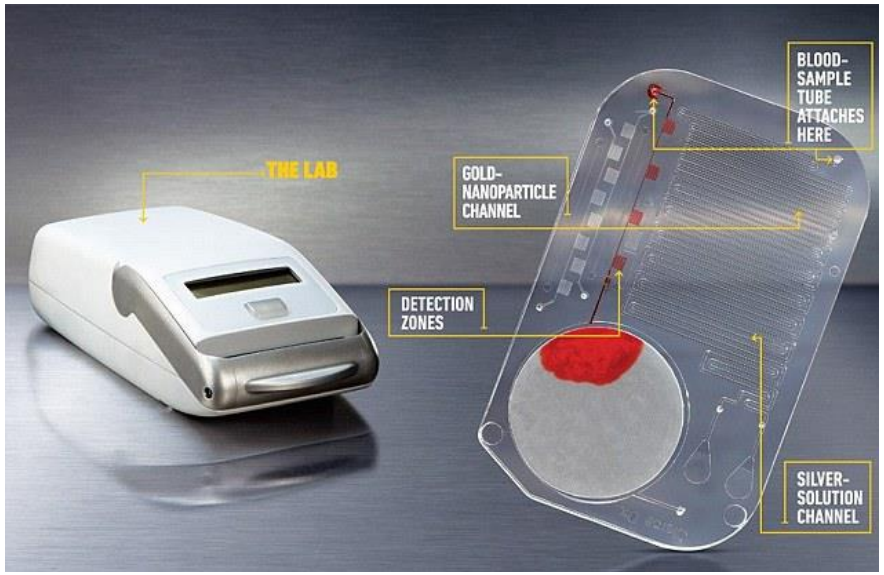


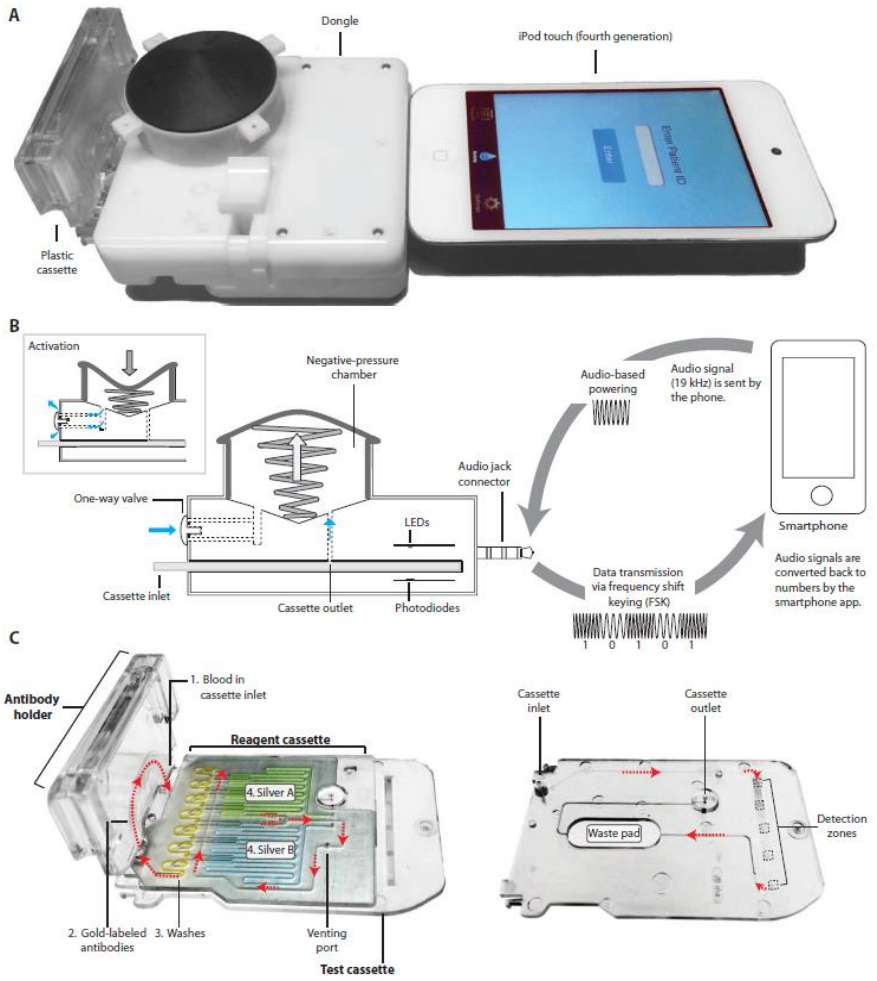
What are the challenges of POC analysis?

1.Simple: power-free, or automatic fluidic control
2.Sample efficient: no dead volume, high selectivity and purity
3.Sensitive: detection spot has to be highly specific

M-Chip for HIV Test

- Serial sample loading by preloaded droplet
- Each detection zone represent to one biomarker
- The total analysis time is ~min



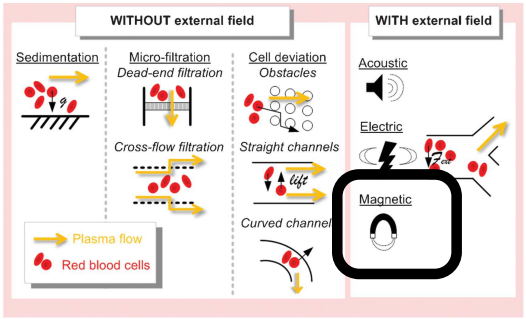


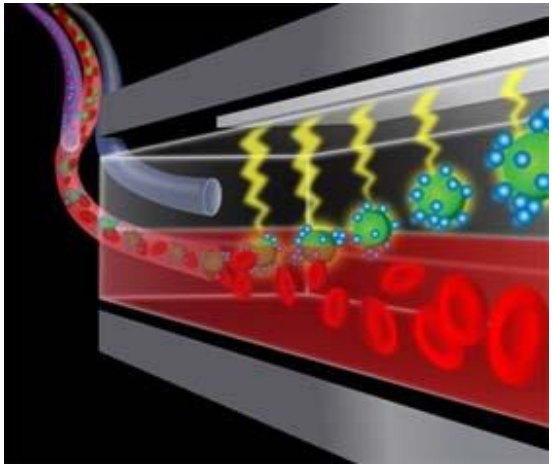
S. Sia Group at U columbia

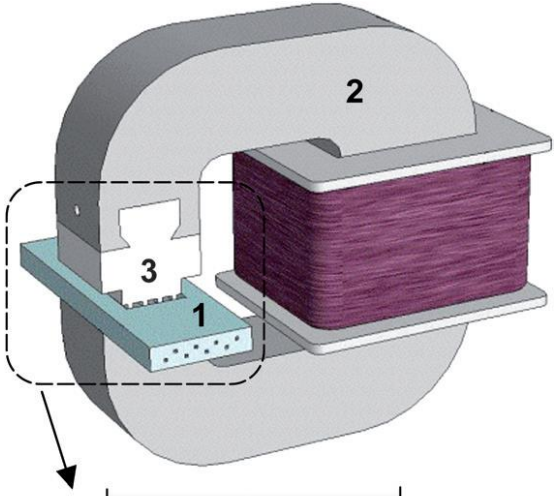
http://www.youtube.com/watch?feature=player_embedded&v=vpxnJM2jSVg

Magnetic Field Separation

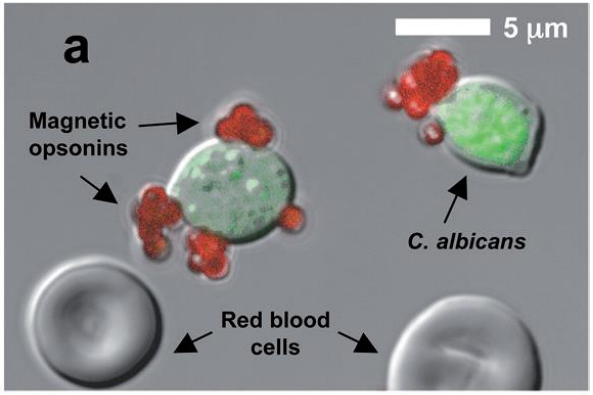
- Blood Cells Separation

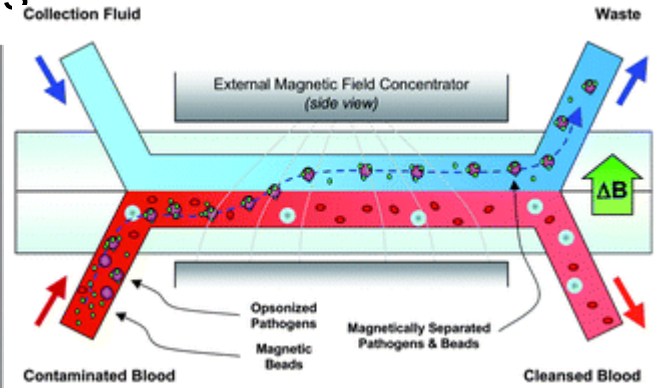


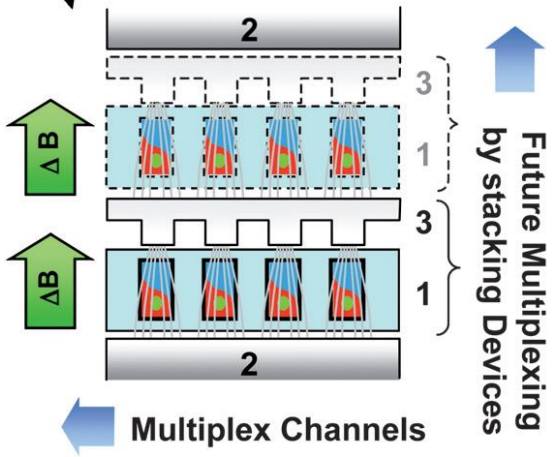




- C.albicans fungi:
  - a leading cause of sepsis-related deaths





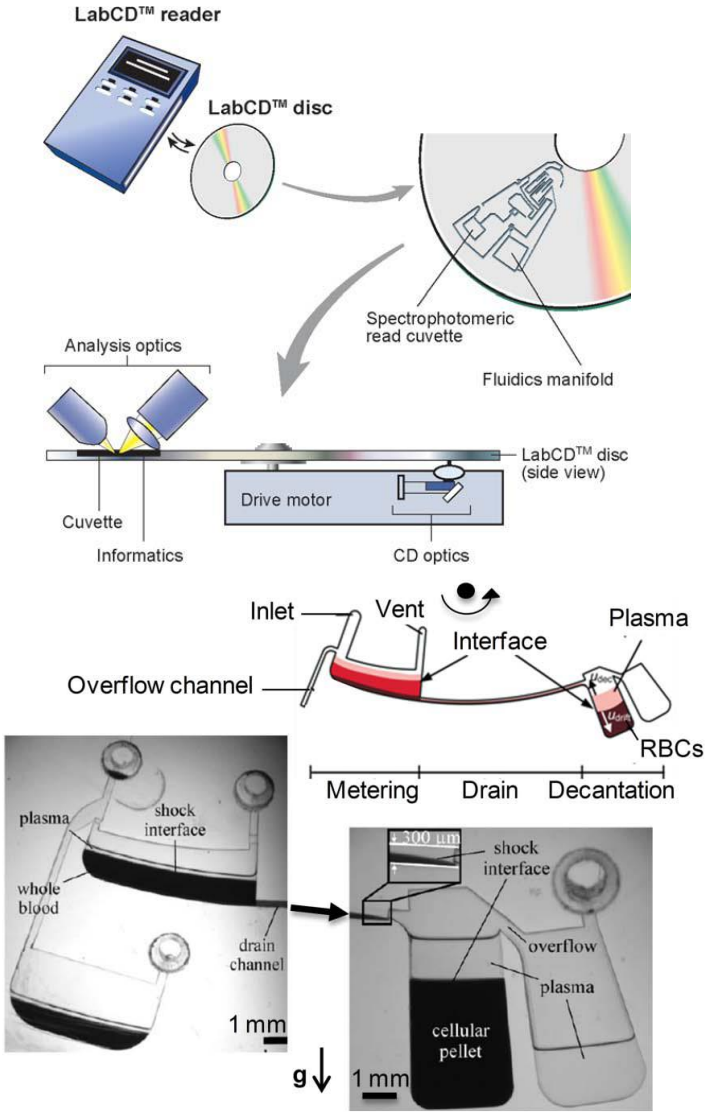


Micromagnetic–microfluidic blood cleansing device ***Lab Chip***, 2009,**9**, 1171-1177

Blood Plasma Separation by CD

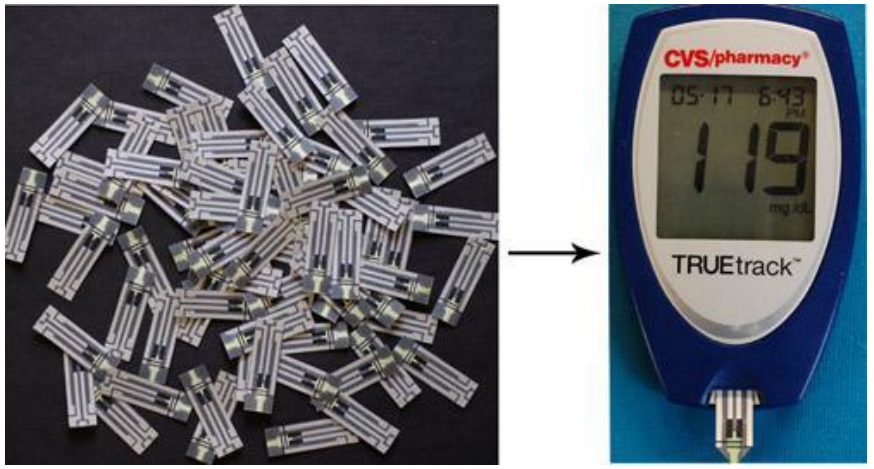
- Use centrifugal force to guide blood sample
- Advantages:
  - Cost-effective
  - High throughput
  - Fast response
- Problems:
  - Not easy to adjust flow rate
  - Require valves
  - Tubing is difficult

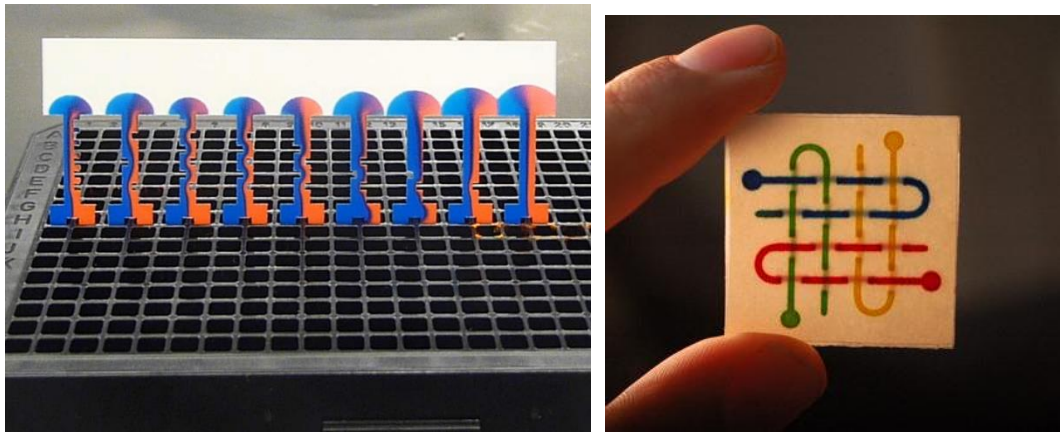
https://www.youtube.com/watch?v=iXUtVtpP6Q8



Paper-based Microfluidics

- Use capillary force to guide blood sample
- Advantages:
  - Low cost
  - Easy of fabrication
  - Long term storage
- Problems:
  - Flow rate is not consistent
  - Difficult to perform fluidic valving
  - Difficult to integrate sensors

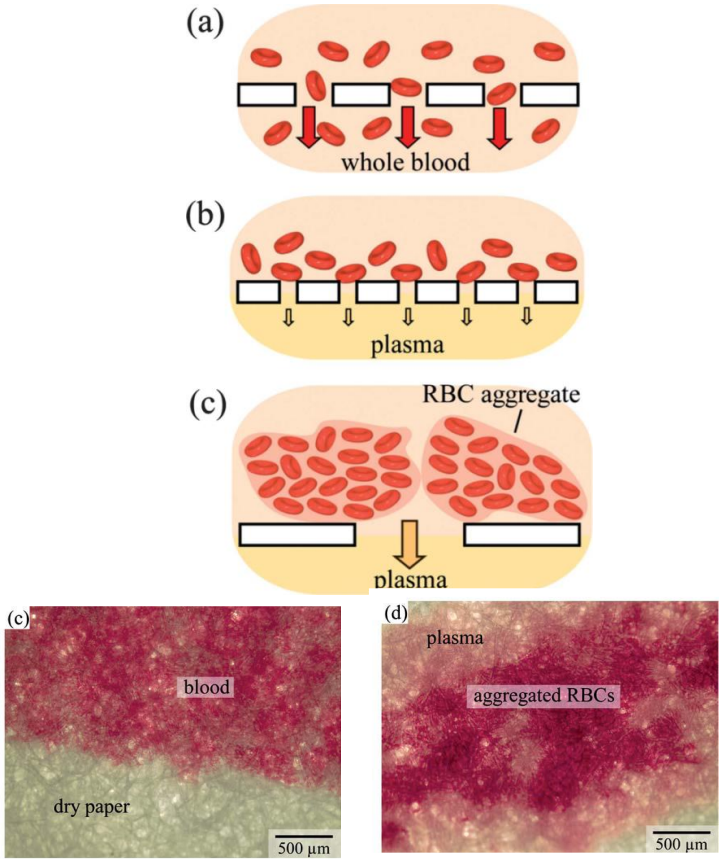


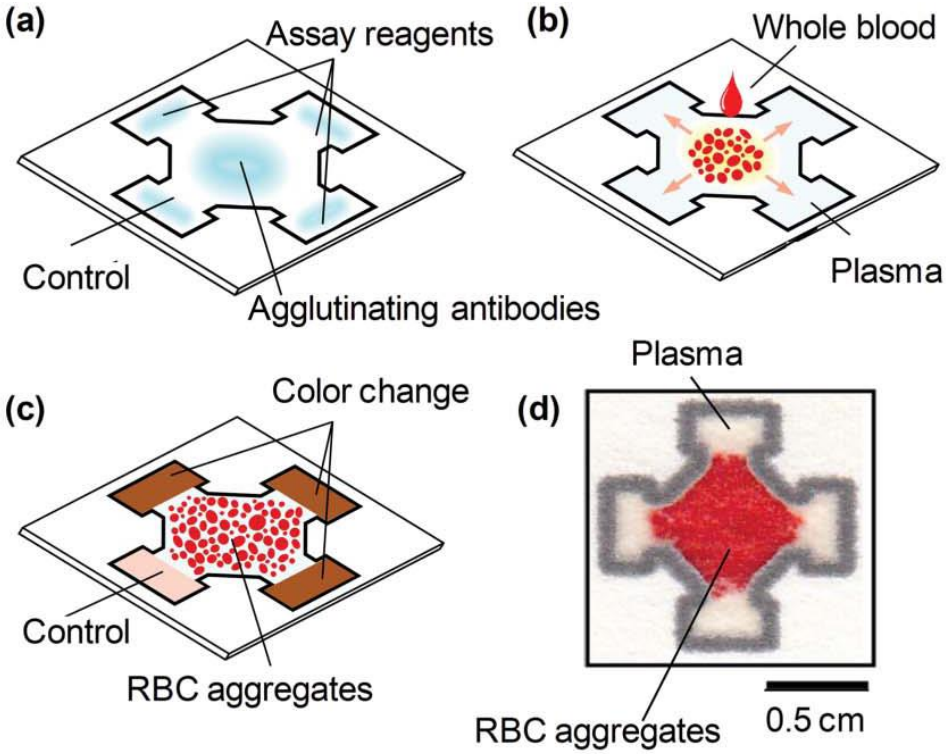


P. Yager Group, U Washington G. Whiteside Group, Harvard

Blood Plasma Separation by Paper

- Use capillary force to draw liquid
  - RBC aggregation helps plasma separation

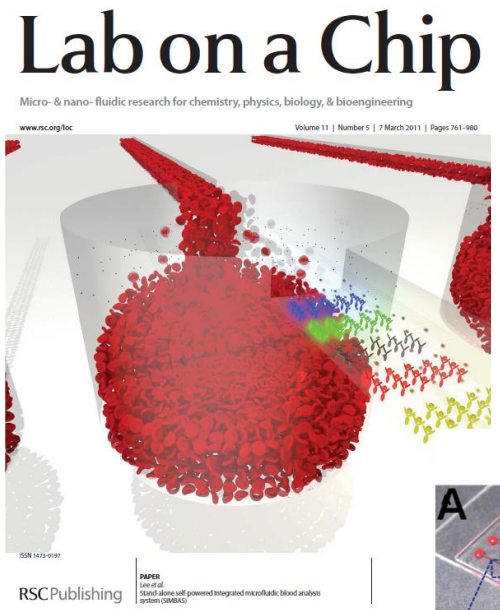


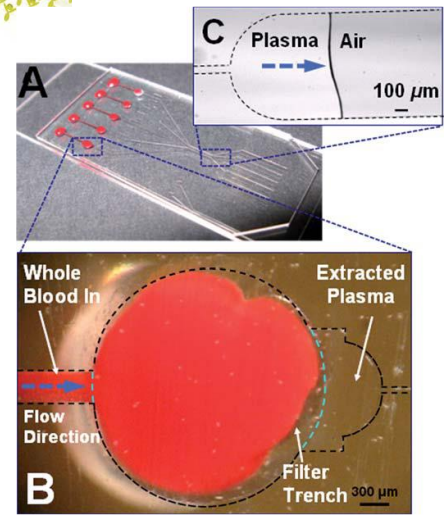


X. Yang, O. Forouzan, T. P. Brown and S. S. Shevkoplyas, Lab Chip, 2012, 12, 274–280

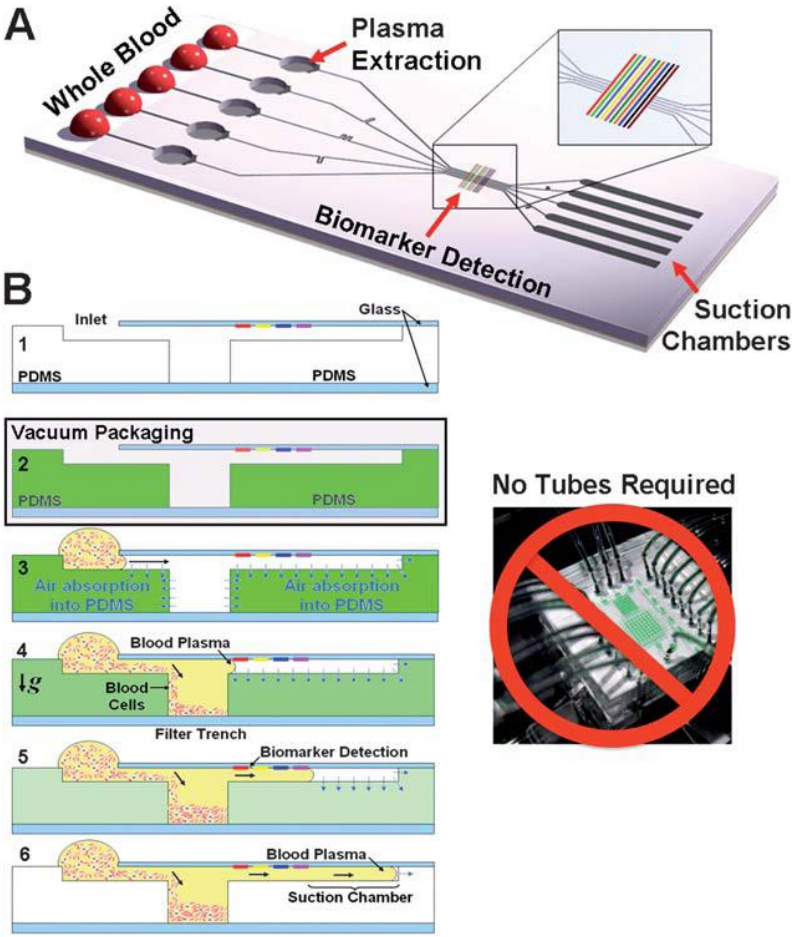
Power-free Blood Separation Microfluidics

- Fluid is driven by vacuumed PDMS





L. Lee Group at UC Berkeley



Challenges of Whole Blood Process

- High cellularity of samples
- Cell components aggregation
  - EDTA: prevent platelet activation
  - Dilution factors
  - Red blood cell lysis: remove 99% of cellular contents
- Large blood volume process
- Long sample culture time

- Requirements of whole blood process device:
- Easy-of-use: automating multi-step sample preparation
- Yield: Preparing samples with high cellularity
- Purity: achieving high purity cell populations
- Throughput: concentrating rare cells from large volumes
- Multiplexity: preparing small volume sample for multiple assays

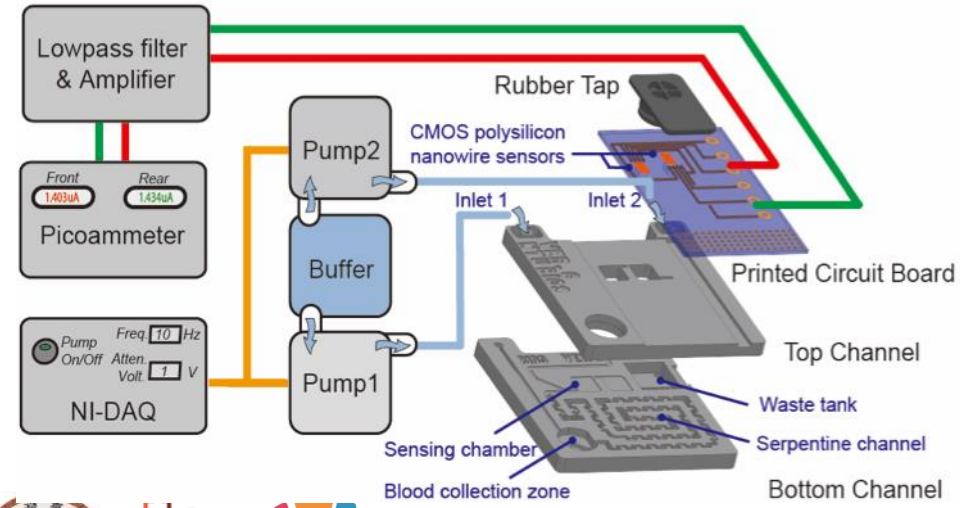
Diabetes Diagnosis Methods

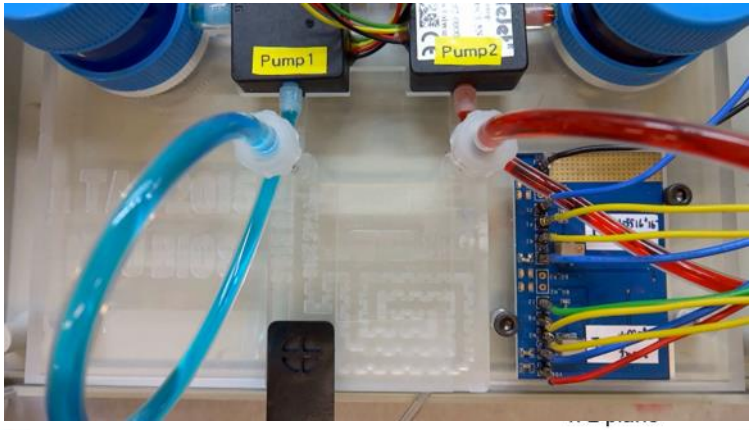
- Diabetes diagnosis methods in the hospital
  - Fasting plasma glucose (FPG) level
  - 2-h value in the oral glucose tolerance test (OGTT)
- POC glucose meters at home
  - Day-to-day glucose level varies by diet, stress levels and illness.
  - Different hematocrit levels in patients
- **Hemoglobin-A1c test (HbA1c / Hb)**
  - Risk for diabetes: HbA1c ratio = 5.7 to 6.4%
  - Require whole blood processing: cell lysis, plasma purification => laborious, time-consuming and require 1~1.5 mL blood

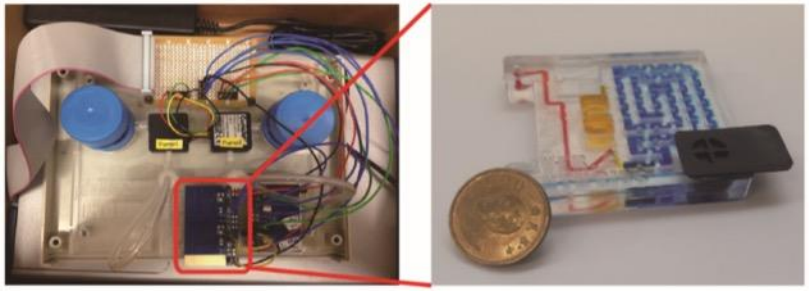
POC Glucose Meter

Microfluidics Integrating Nanowire Sensors

- Microfluidics + nanowires: on-chip whole blood processing and analytes detection
- Three-dimensional microchannel: blood cells trapping and plasma dilution
- Programmable piezoelectric pumps: automatic fluidic control
- CMOS nanowire sensors: label-free and dynamic detection of analytes
- Total assay time: <30 minutes
- Required blood volume: 5 μL

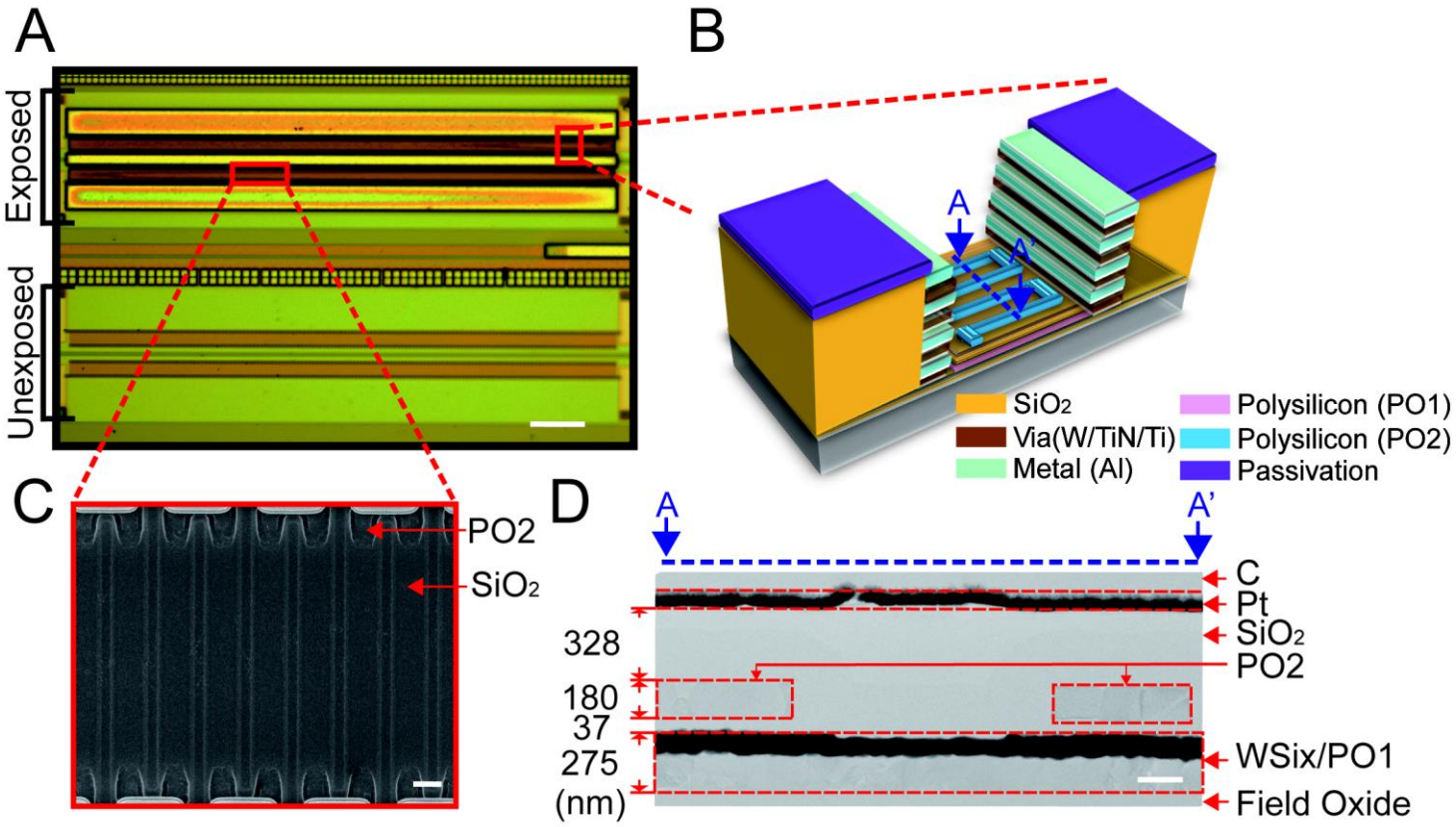






Schematic of CMOS Nanowire Sensors

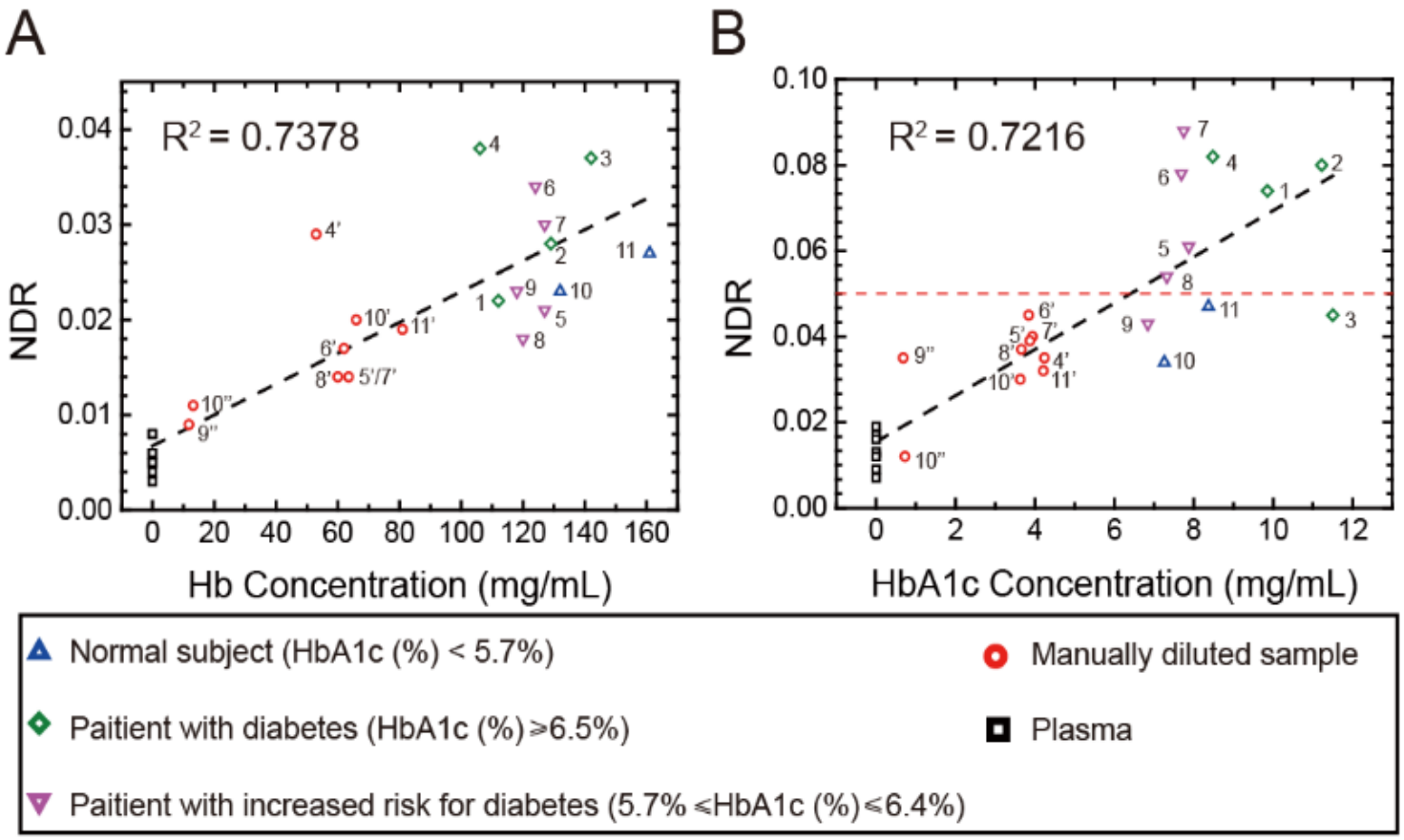
- 0.35μm two-polysilicon-four-metals (2P4M) CMOS standard fabrication technology



(Kuan et. al., Lab on Chip, 2016)

Detection of Hb and HbA1c Concentrations in Clinical Samples

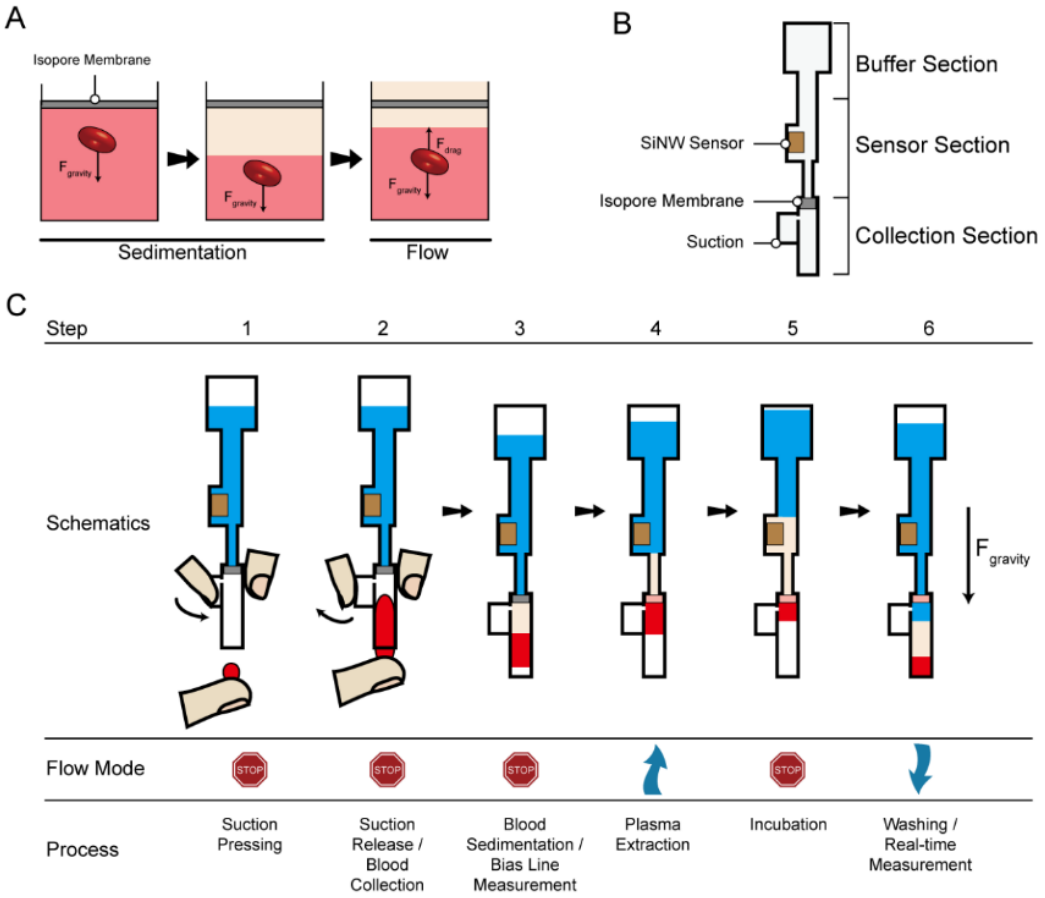
- Three clinical sample groups with different diabetes risk level

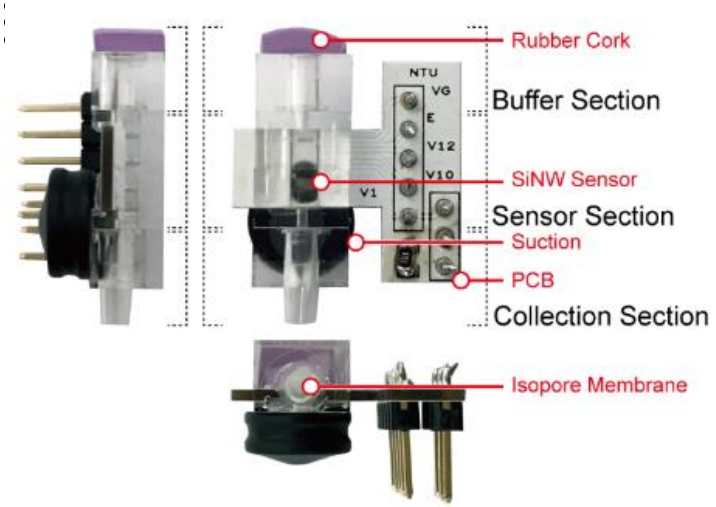


(Kuan et. al., Lab on Chip, 2016)

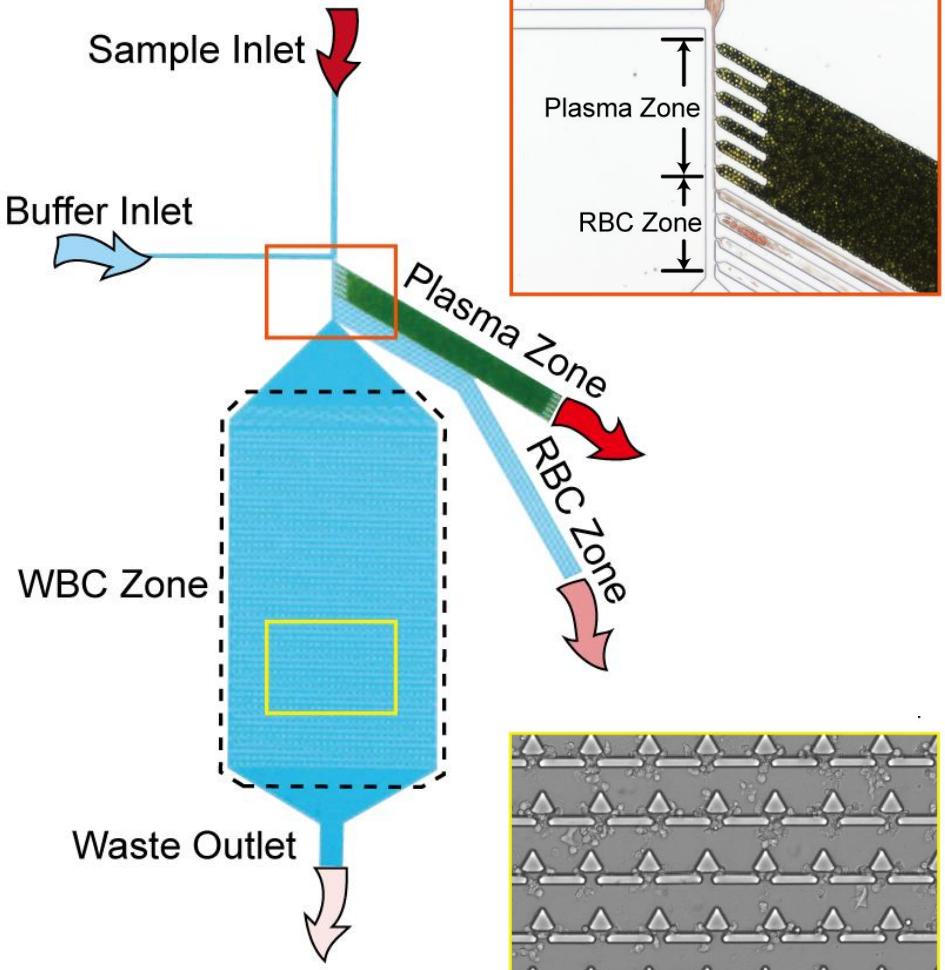
Microfluidic Platform for Heart Failure Diagnosis

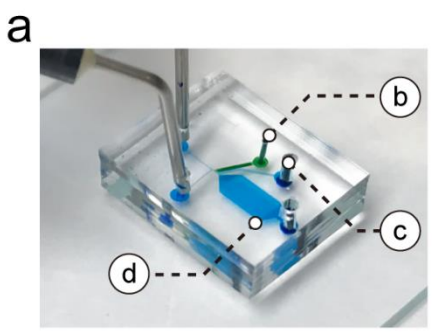
- Integration of sedimentation and porous membrane to enable dilution-free whole blood processing

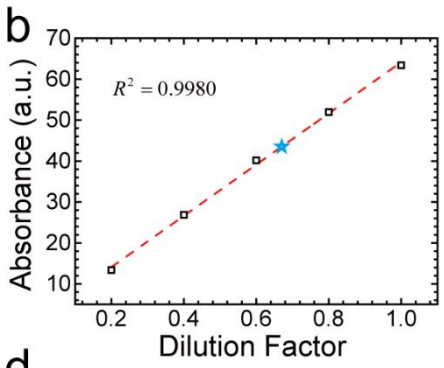


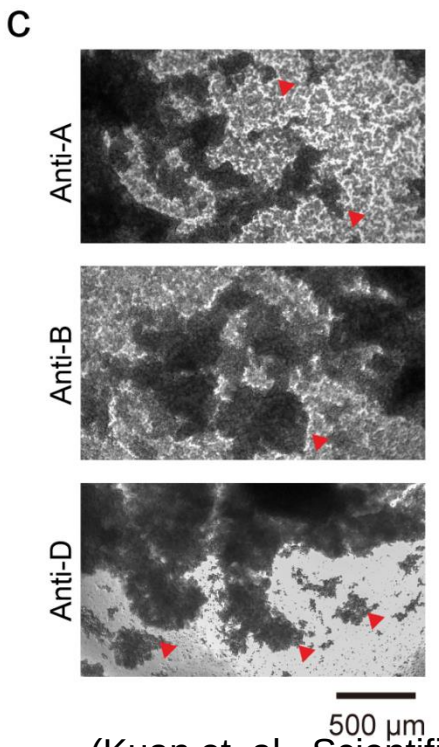


The Microfluidic Device for Plasma Extraction, RBC, and WBC Trapping









(Kuan et. al., Scientific Reports, 2018)

Summary

- We developed various microfluidic platforms for whole blood processing and in-situ analyte detection
  - on-chip whole blood processing and in-situ Hb/HbA1c detection
  - dilution-free plasma extraction
  - simultaneously plasma extraction, RBC and WBC trapping

- The microfluidic platforms for whole blood processing could
  - improve the accuracy and sensitivity of biosample analysis
  - eliminates the cost and time of sample preparation process
  - less blood sample volume, important for infants and elderly people

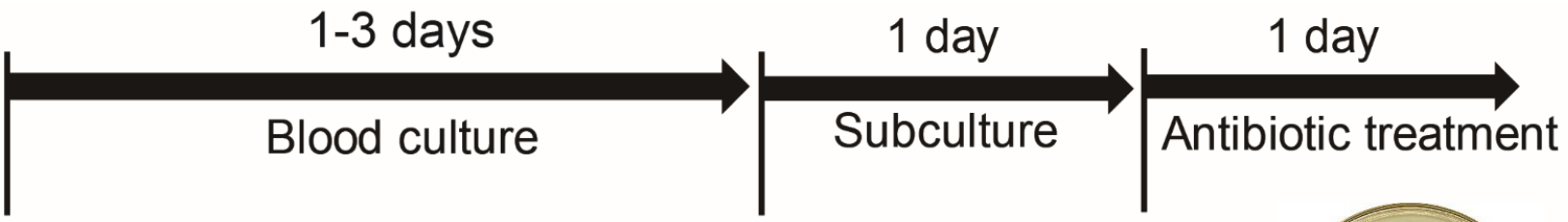
Microfluidics for bacteria isolation and detection

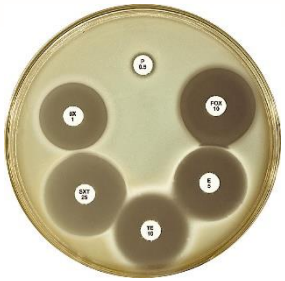
Bacteria Infection Diagnosis Method

- Current bacteria antibiotic susceptibility test (AST)

- Problems:
  1. complicated procedures and bulky instruments
  2. prolonged bacteria culture and sample process time

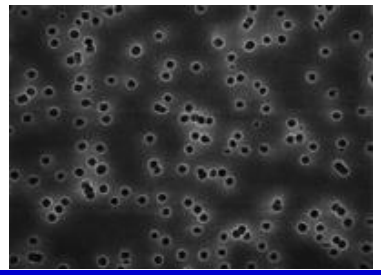
Clinical AST procedures





- Solution: the use of broad band antibiotics

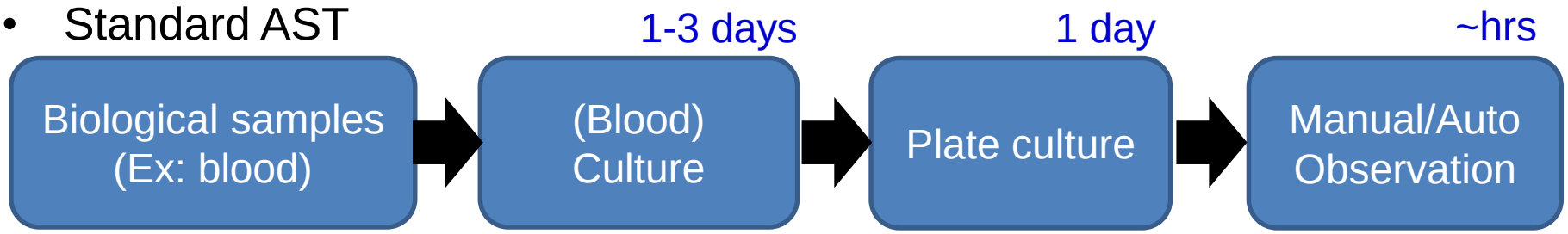
- Challenges of AST :
  - Extremely low bacteria in blood
  - Rapid and efficient diagnosis
  - Suitable antibiotics



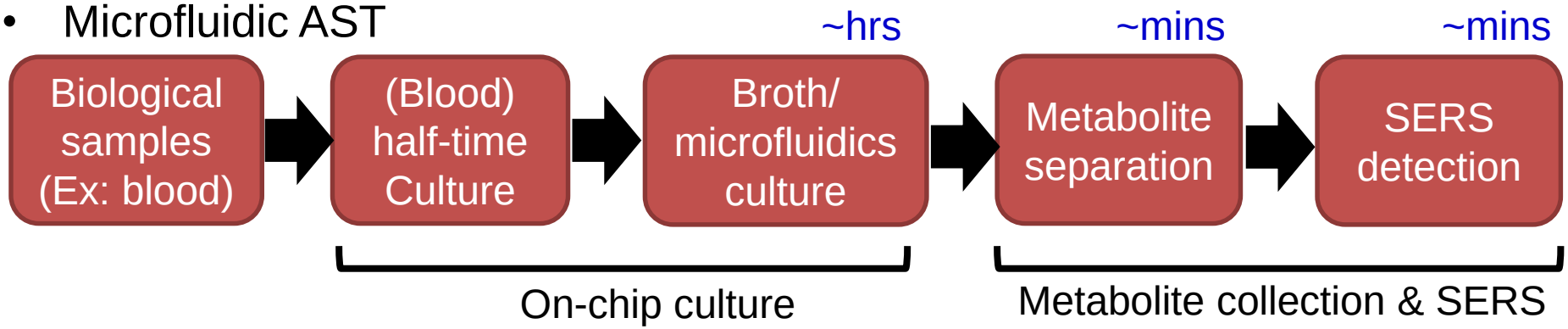
A rapid, highly-sensitive bacteria phenotypical analysis is required

Microfluidics for bacteria AST

- Standard AST

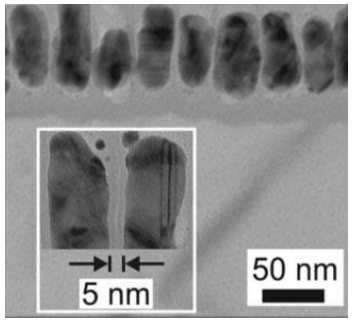


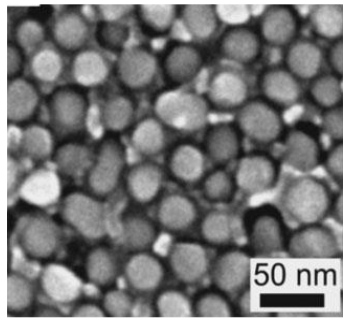
- Microfluidic AST



- System integration
  - Less manipulation error
  - Lower sample volume & process time
- Surface-Enhanced Raman Scattering (SERS)
  - Label-free and rapid detection

D=25nm W=5nm

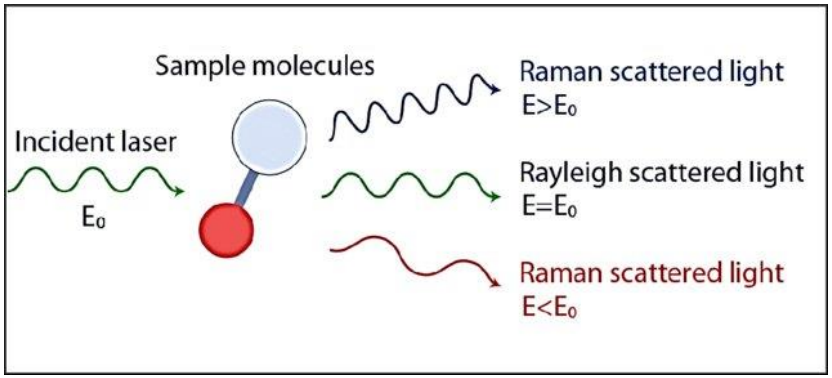




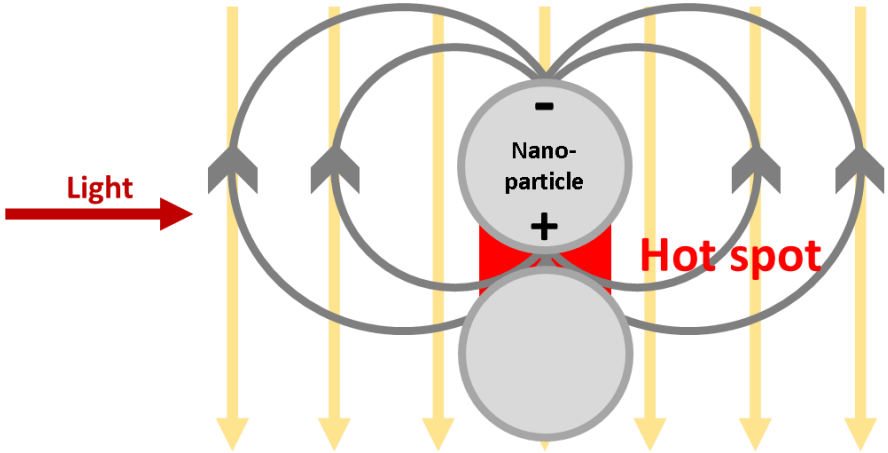
Surface-Enhanced Raman Scattering (SERS)

- **Raman scattering (RS)**
  - Inelastic scattering
  - 1 out of 10 million photons
  - Fingerprints of molecule

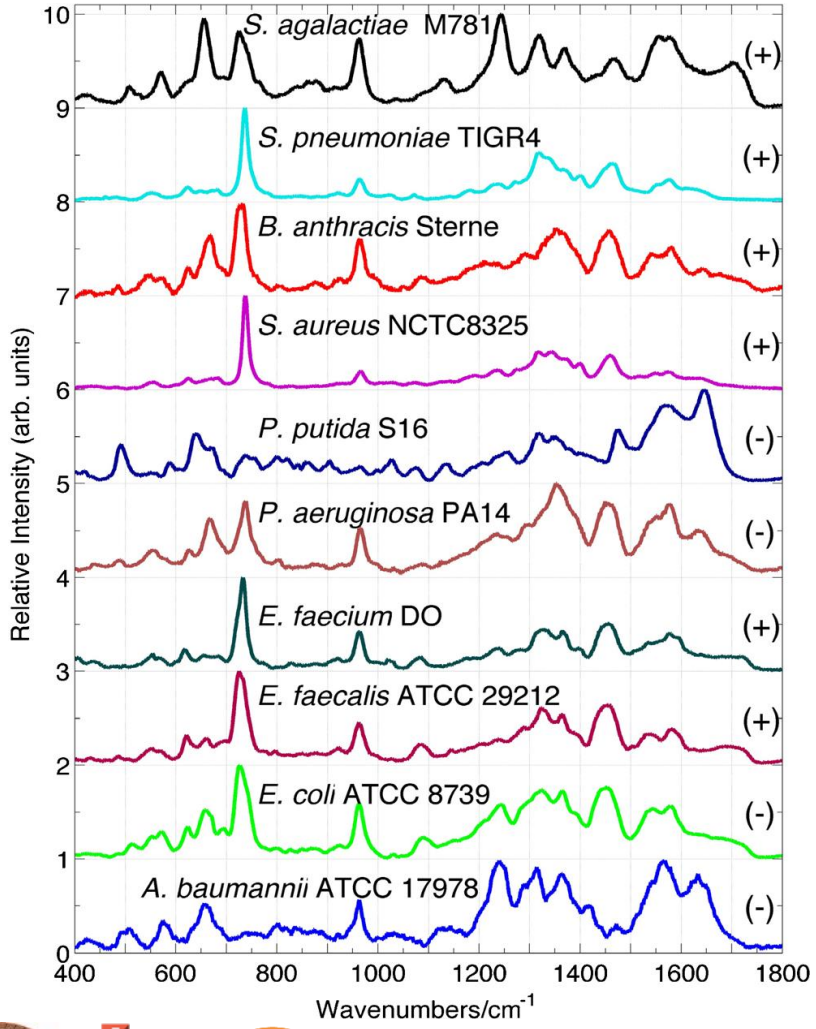
- **Surface-enhanced RS**
  - Rough silver or gold surfaces
  - Surface plasmon resonance
  - Enhance intensity (10^{10} ~10^{14})

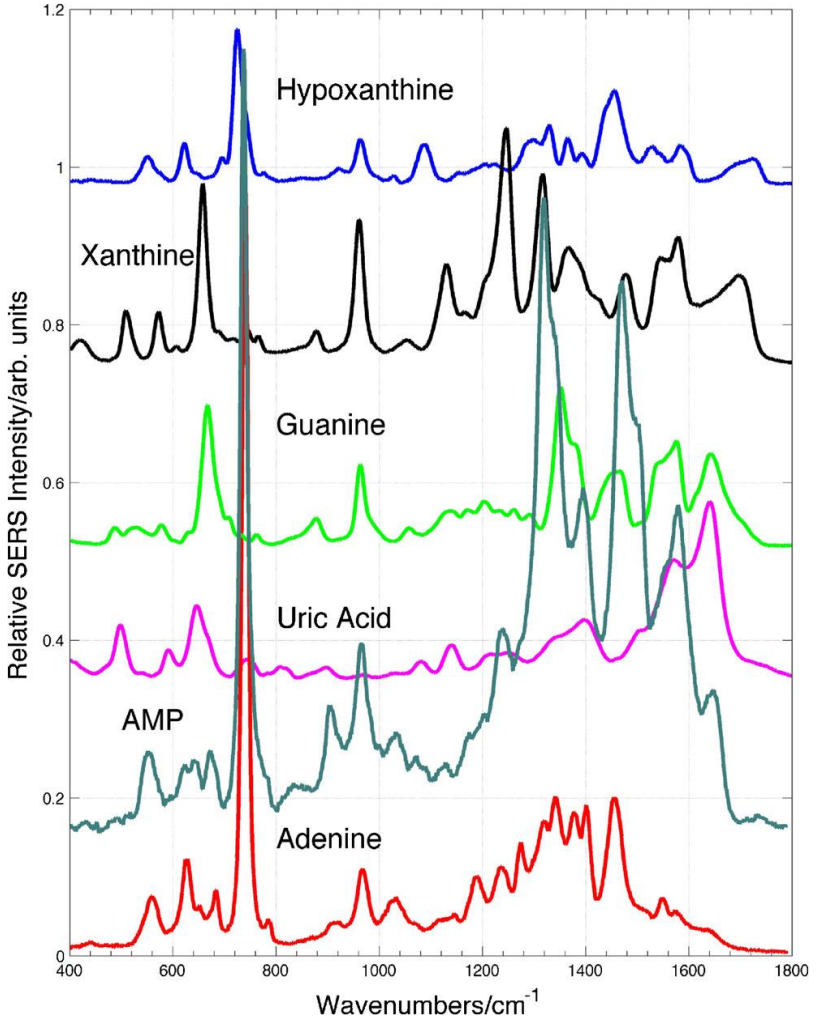


(Smith and Dent, 2005)



SERS Spectrum of Bacteria Strains

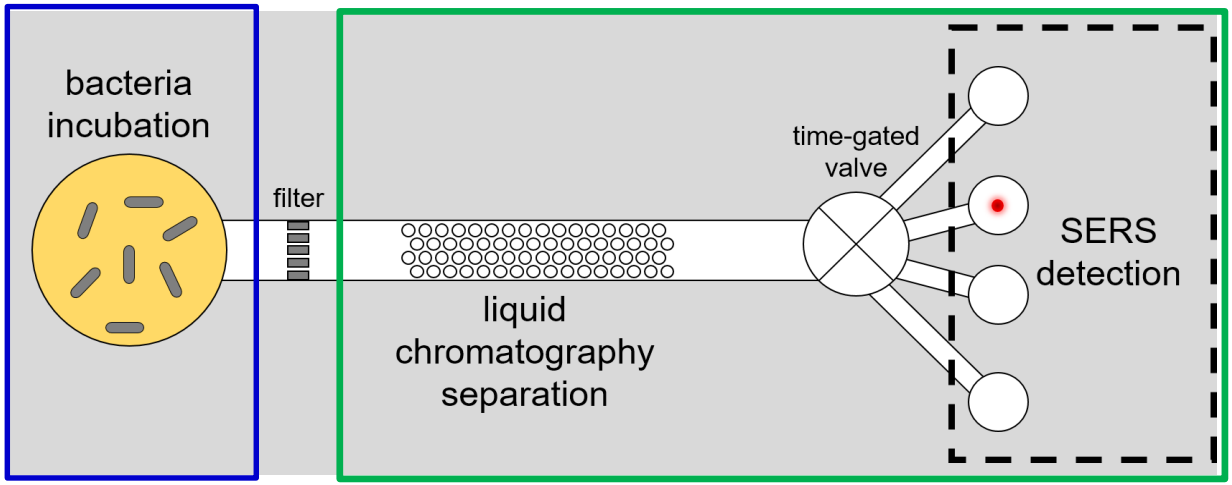




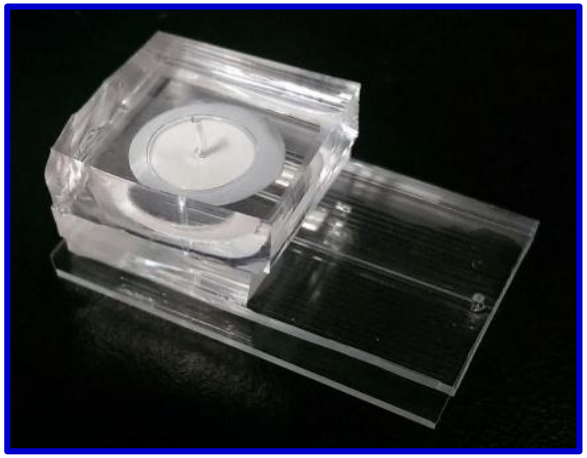
(W. R. Premasiri, et al, Anal Bioanal Chem, 2017)

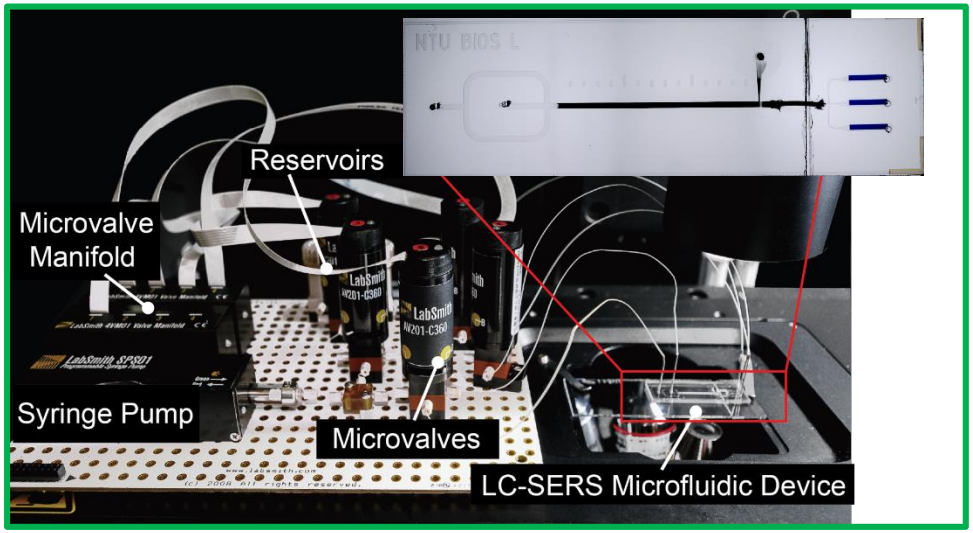
Prof. Ziegler in Boston University

Microfluidics for bacteria AST



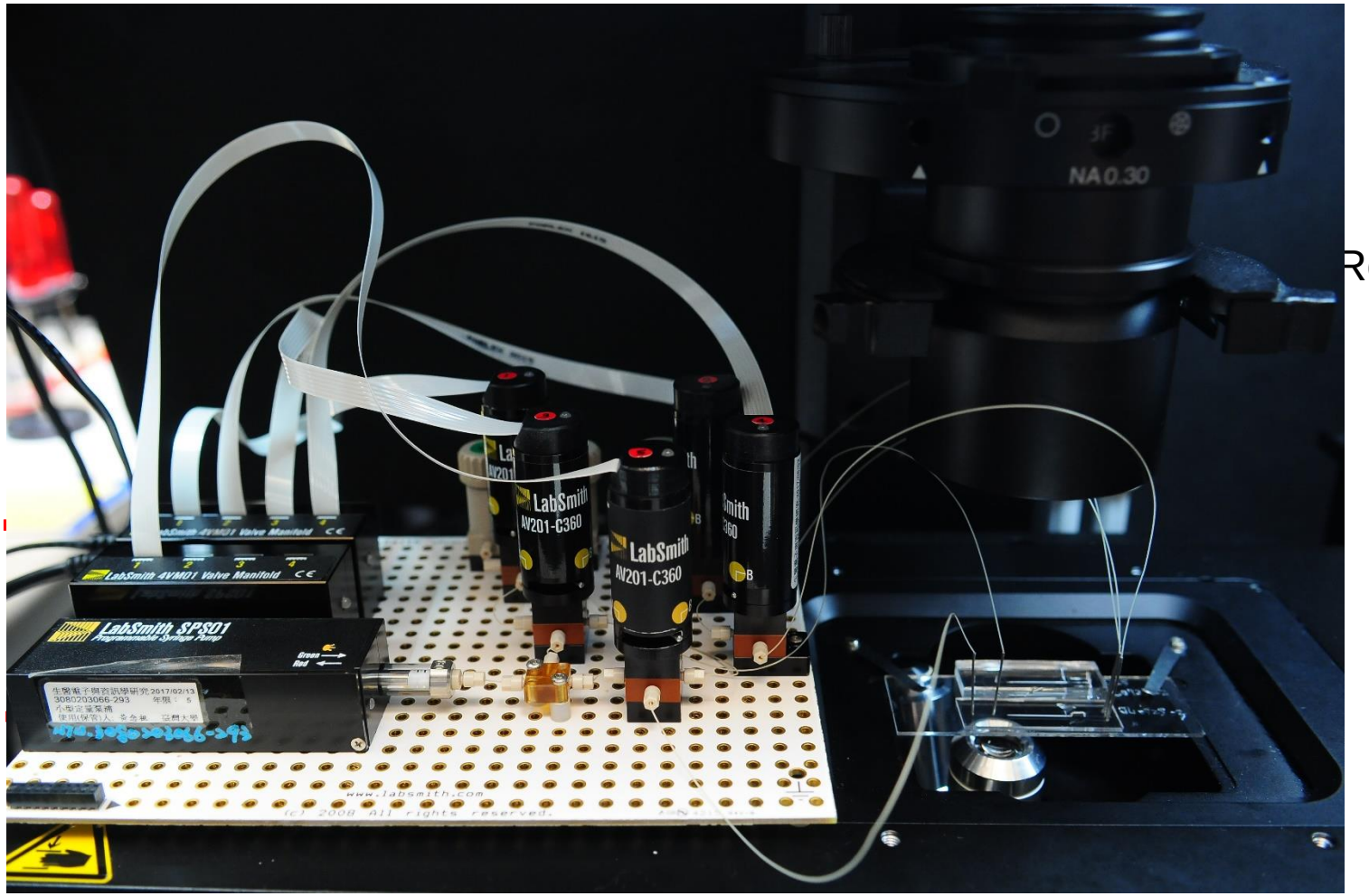
Sample preparation + Metabolite detection





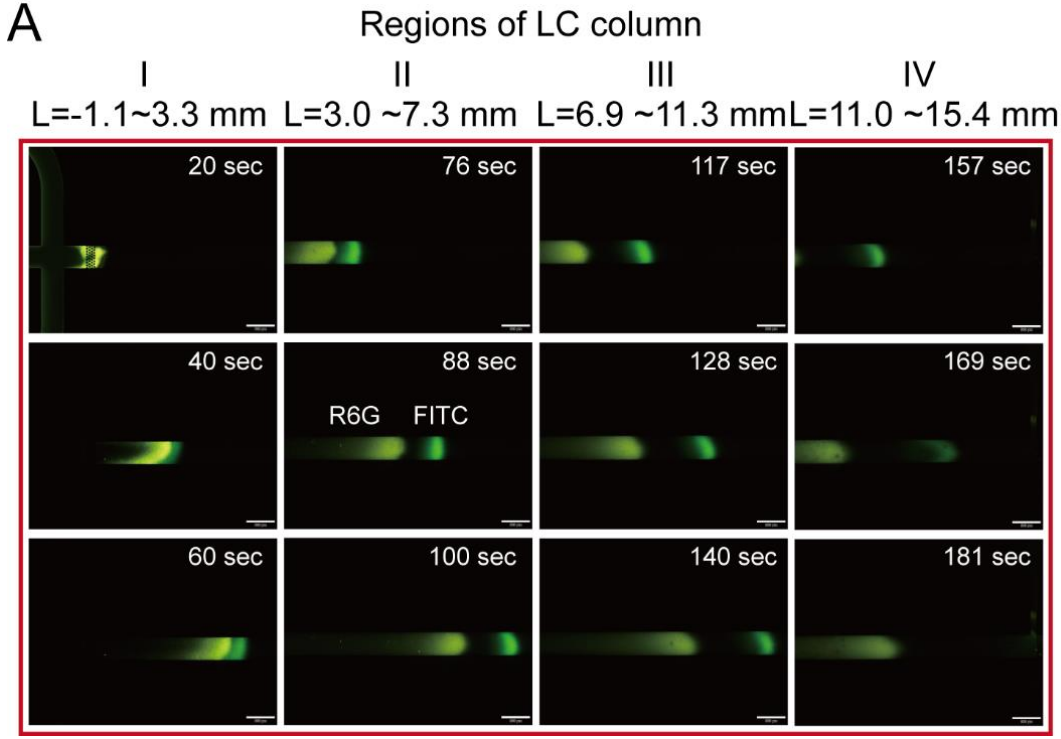
Automated microfluidic system

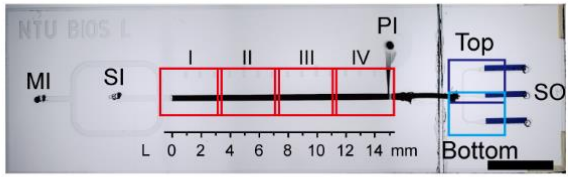
185 mm X 133 mm

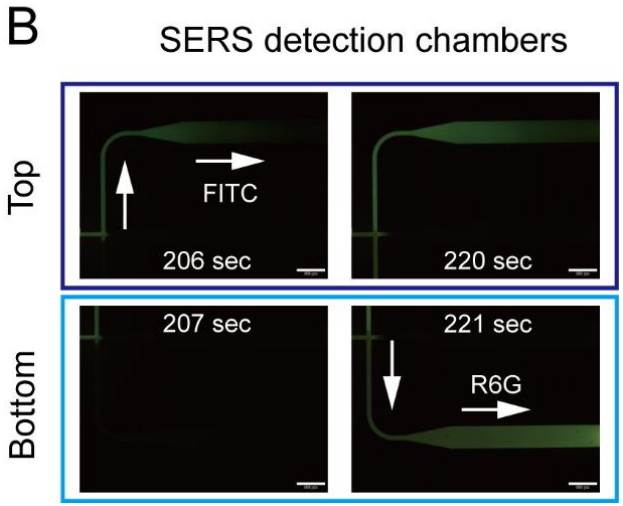


(Wang et. al., Microfluid. and Nanofluid., 2019)

On-chip LC separation of FITC and R6G



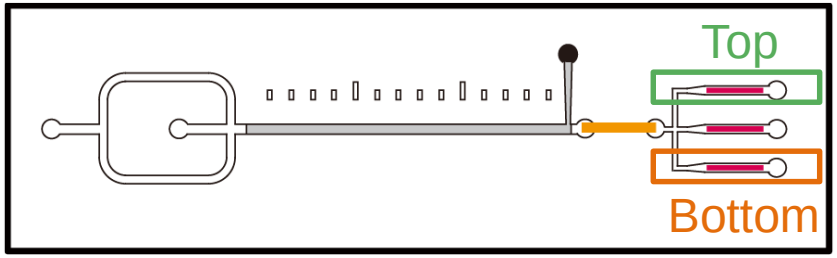


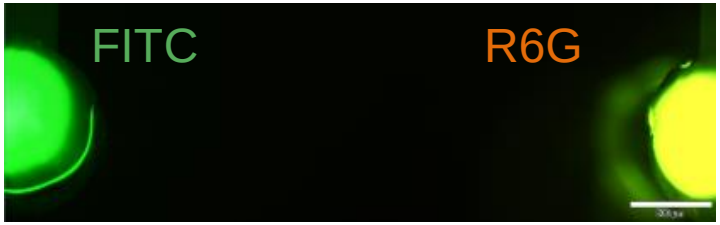


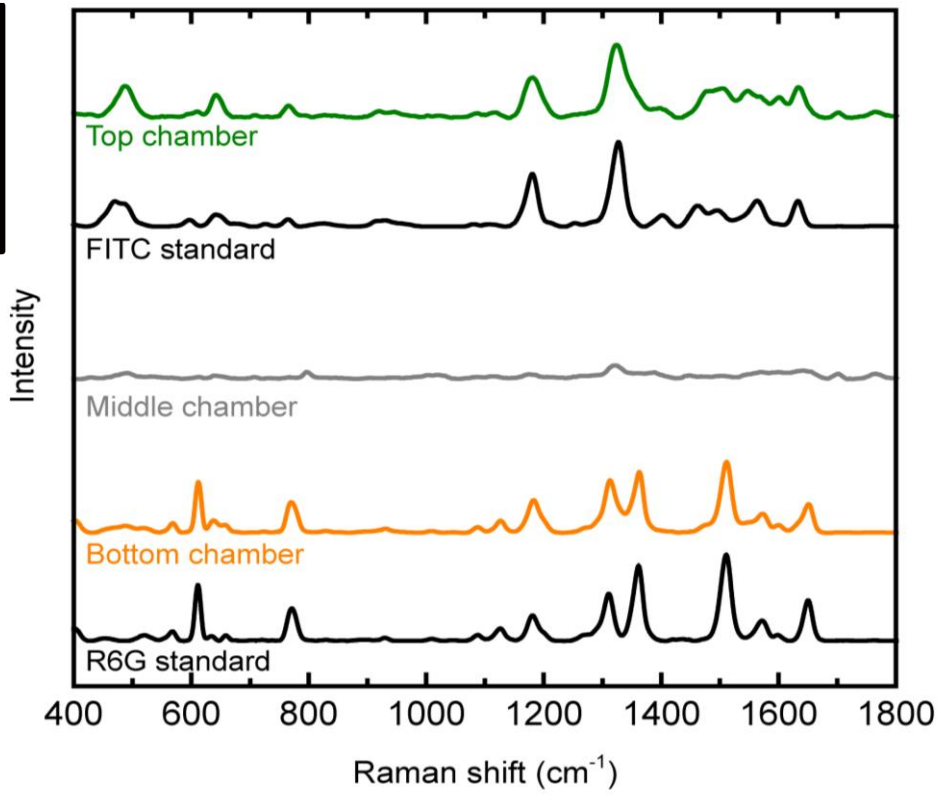
(Wang et. al., under review)

On-chip LC separation and SERS Detection

- Two fluorescent molecule (FITC and R6G) separation and in-situ SERS detection

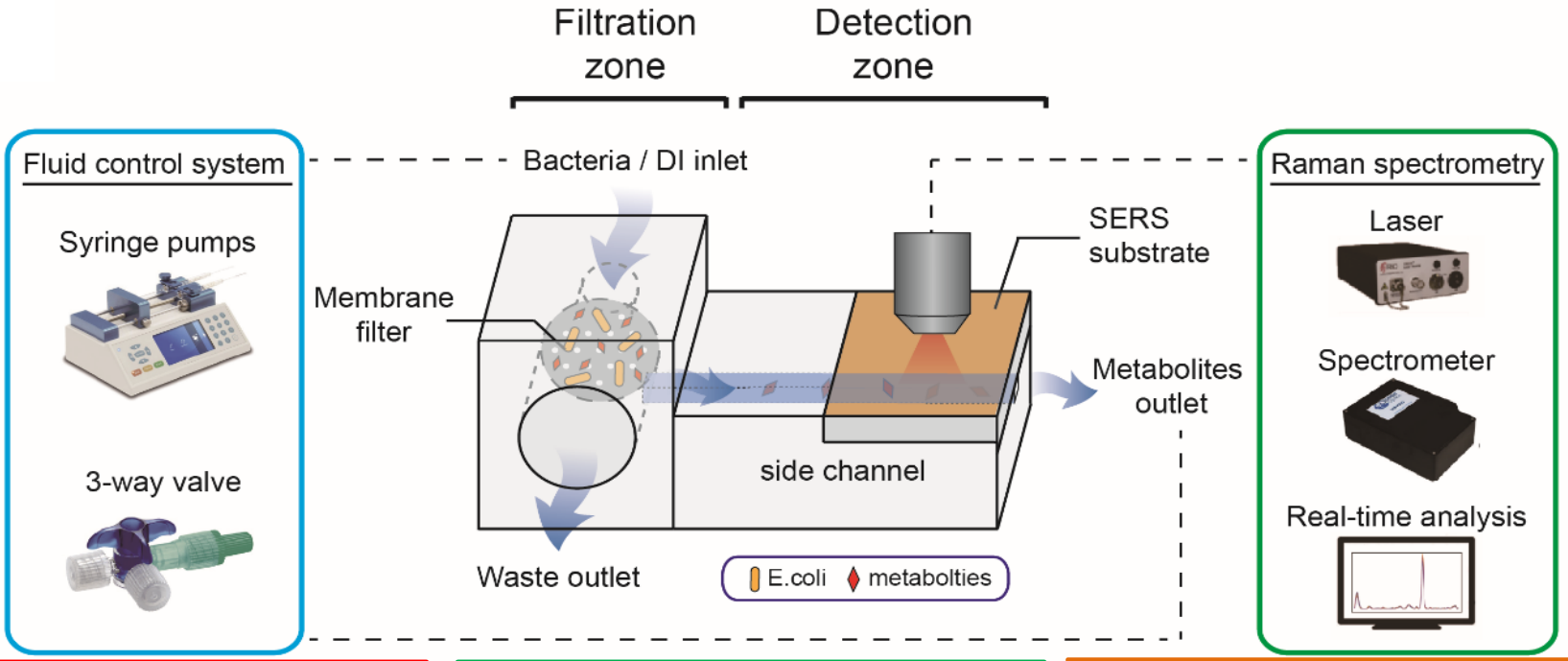


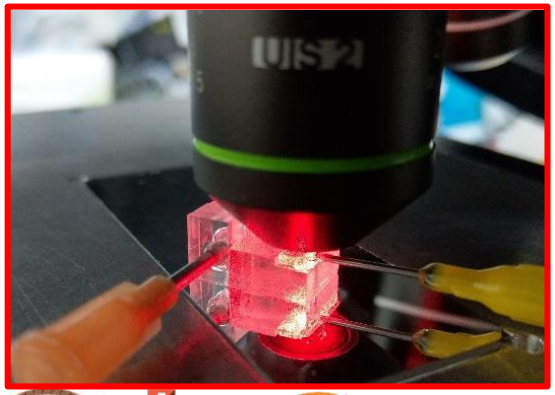


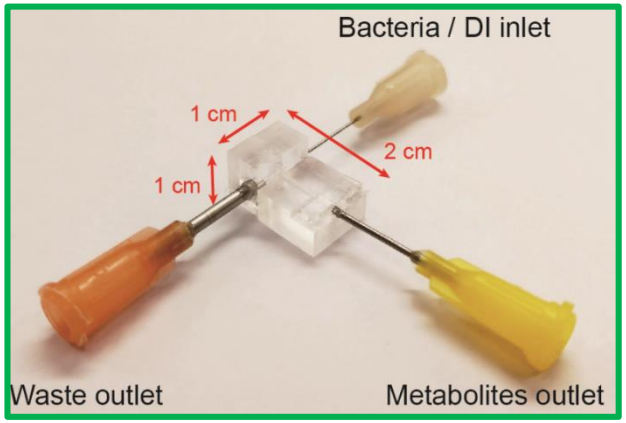


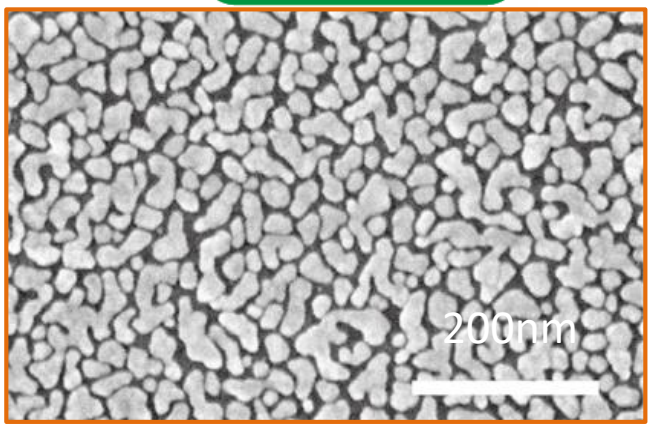
(Wang et. al., under review)

The microfluidic system integrating membrane filtration and SERS





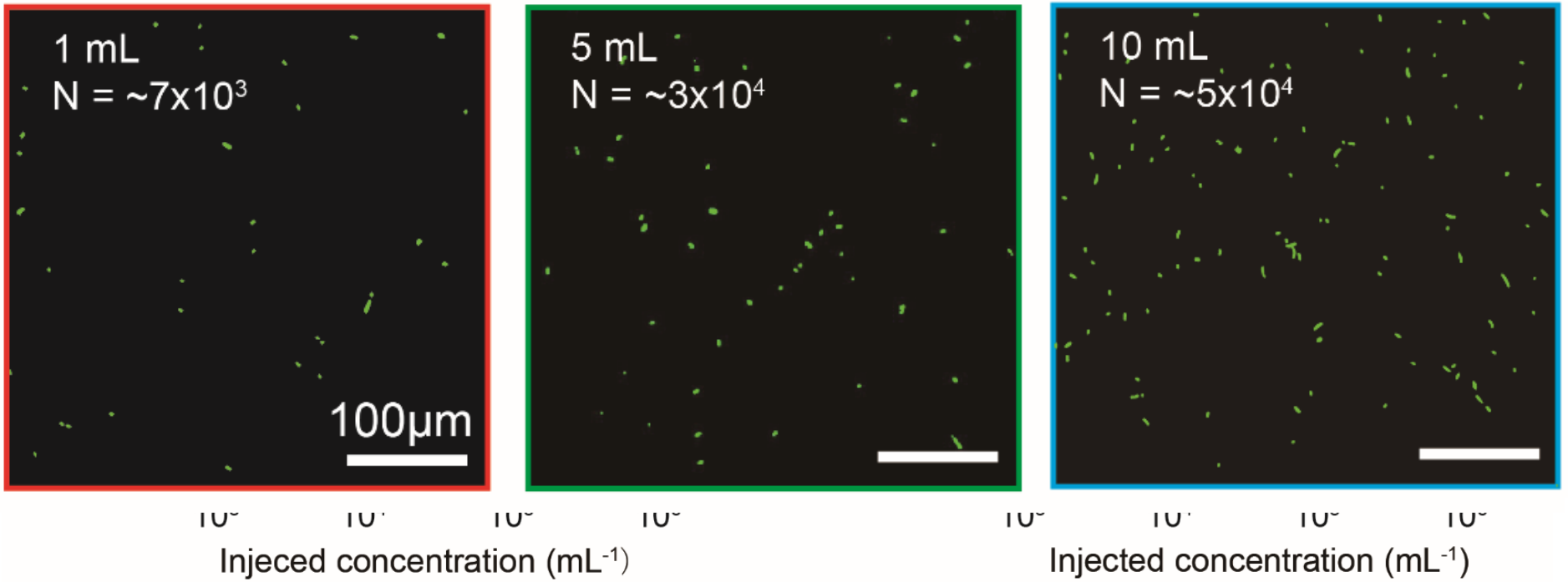




(Chang et. al., Anal. Chem. 2019)

Bacteria Filtration Capability

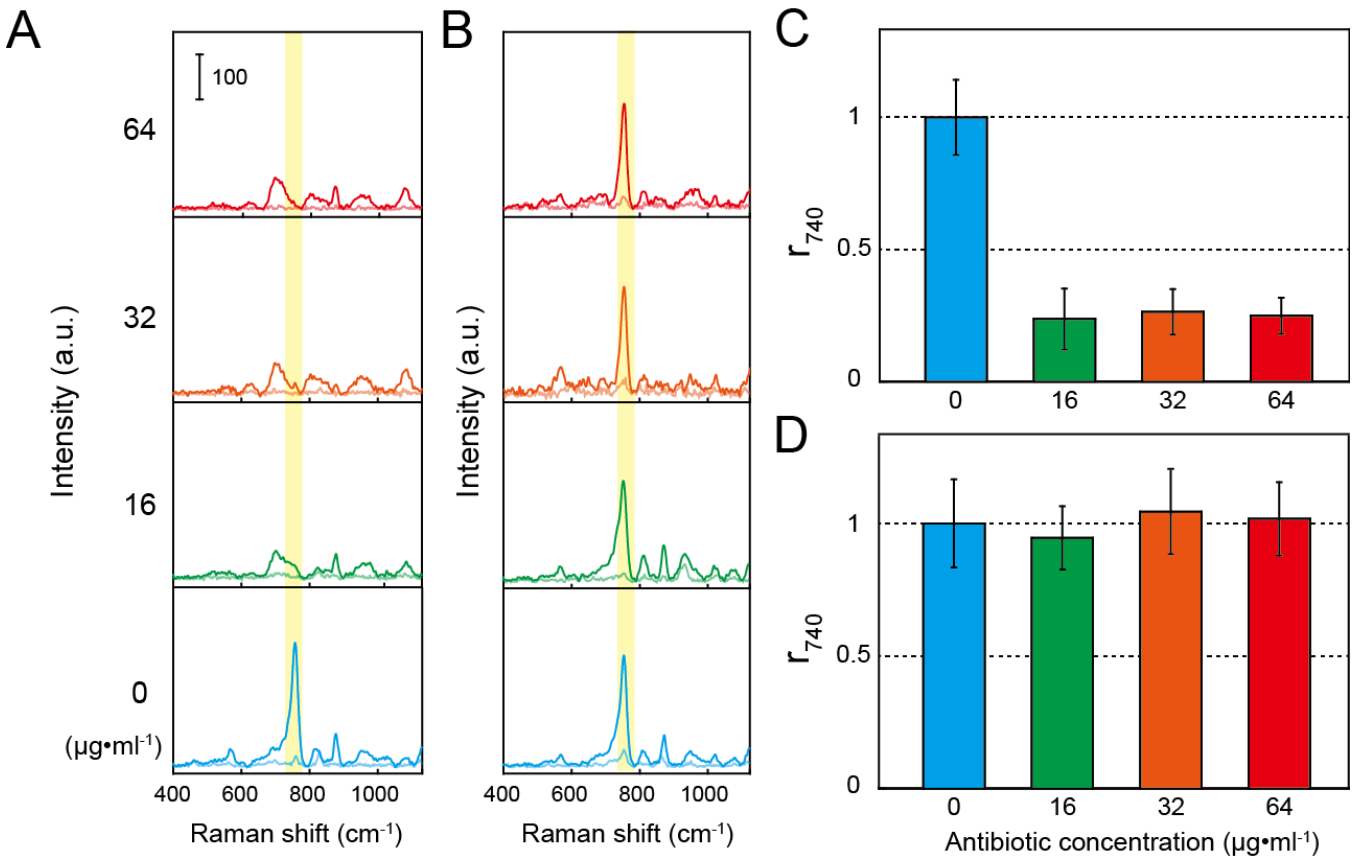
- Membrane filter: Polycarbonate membrane (pore size=0.22μm)
- Bacteria strain: Fluorescent E. coli (ATCC 25922 transfected with GFP)
- Volume: 1, 5, 10 mL & Concentration: 10^3 – 10^6 mL^{-1}



(Chang et. al., Anal. Chem. 2019)

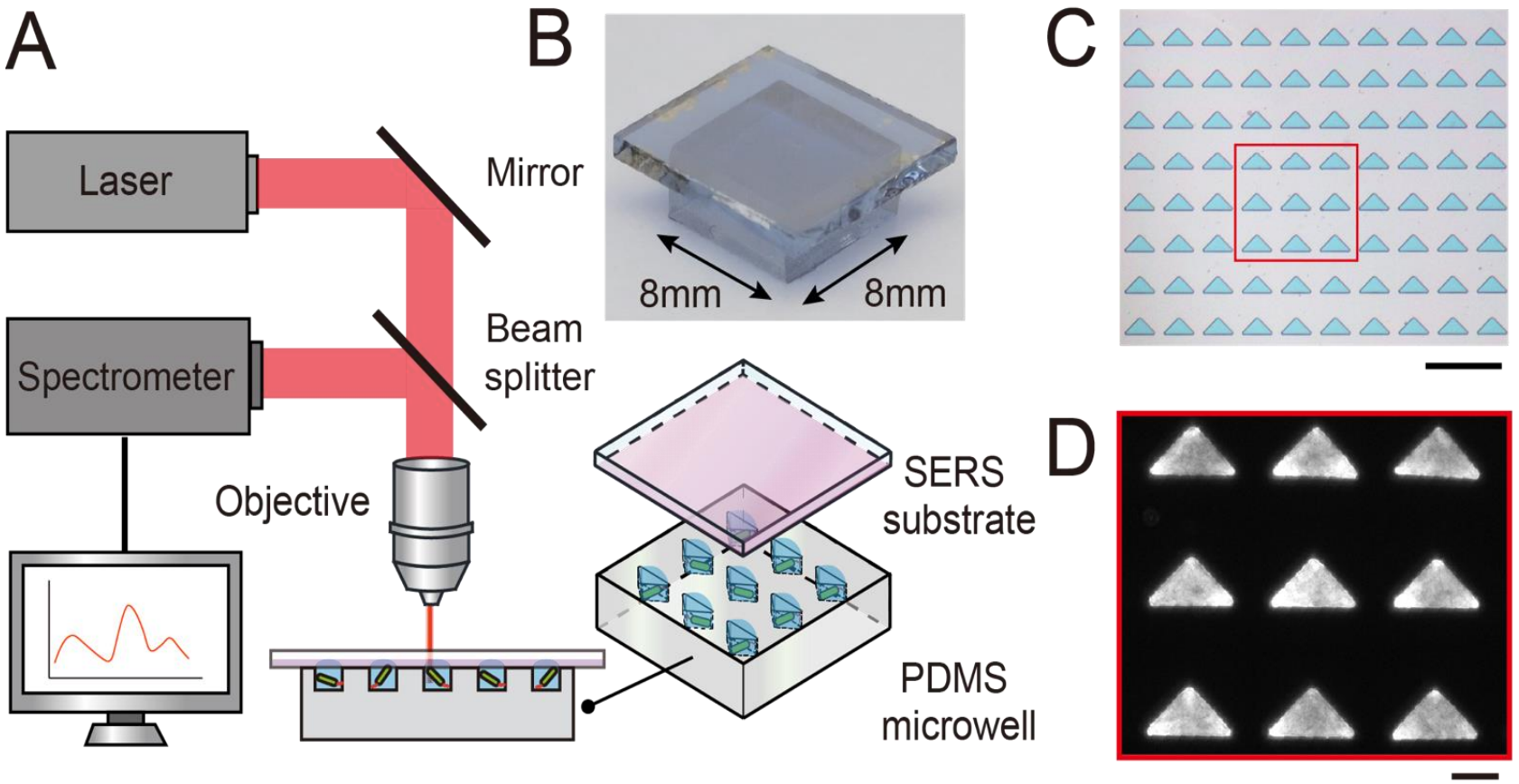
On-chip AST

- Susceptible E. coli (ATCC 25922)
- Resistant E. coli (DH5-alpha transfected with kanamycin resistance)



(Chang et. al., Anal. Chem. 2019)

The microwell-SERS system



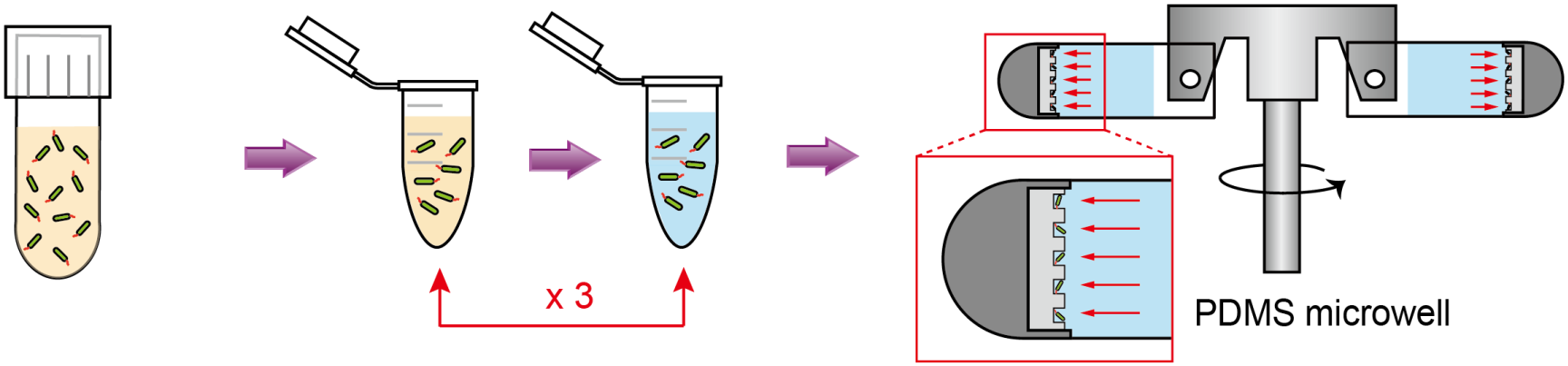
(Huang et. al., Lab on Chip, 2020)

Operation protocol of Microwell-SERS system

1. Incubation with antibiotic

2. Removal of culture medium

3. Bacteria dispersed inside microwells

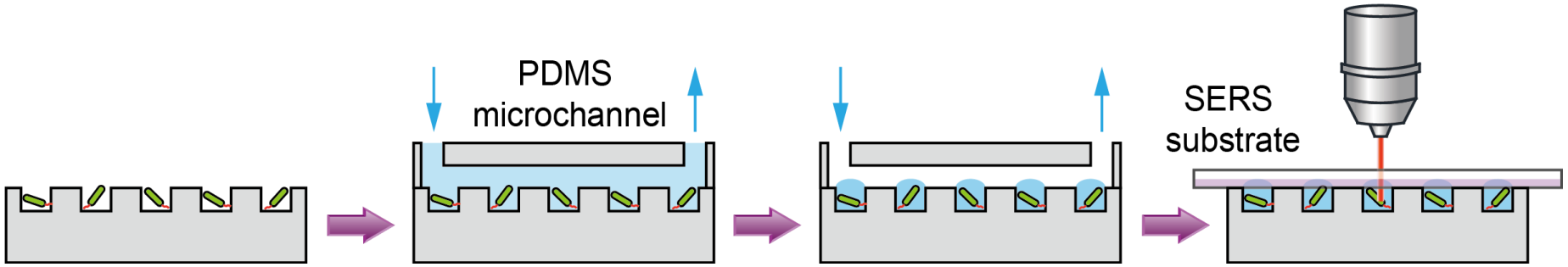


4. Air-dry

5. Bacteria hydration

6. Removal of water in microchannel

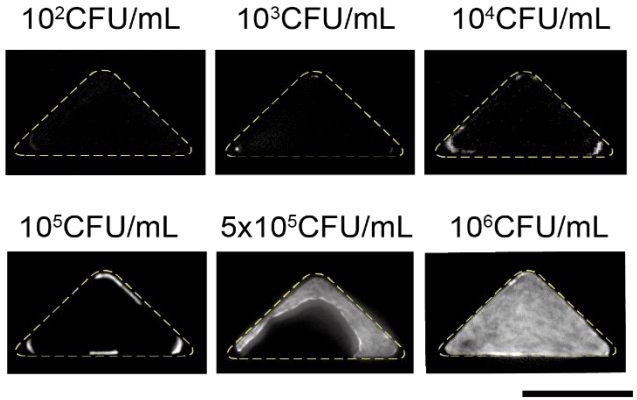
7. SERS detection



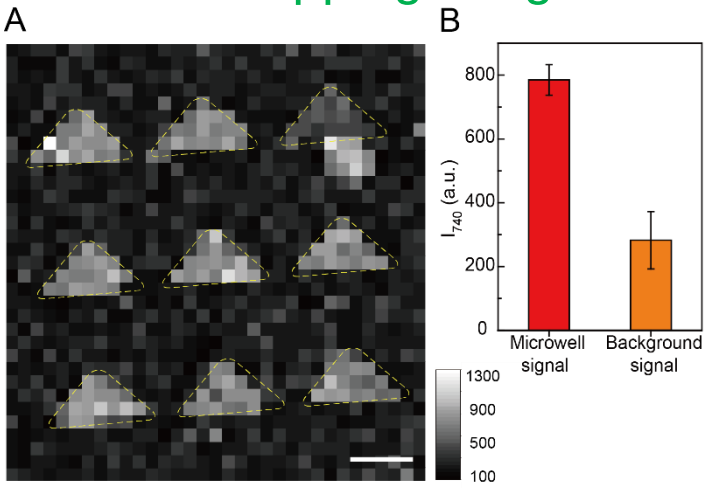
(Huang et. al., Lab on Chip, 2020)

The microwell-SERS system for AST

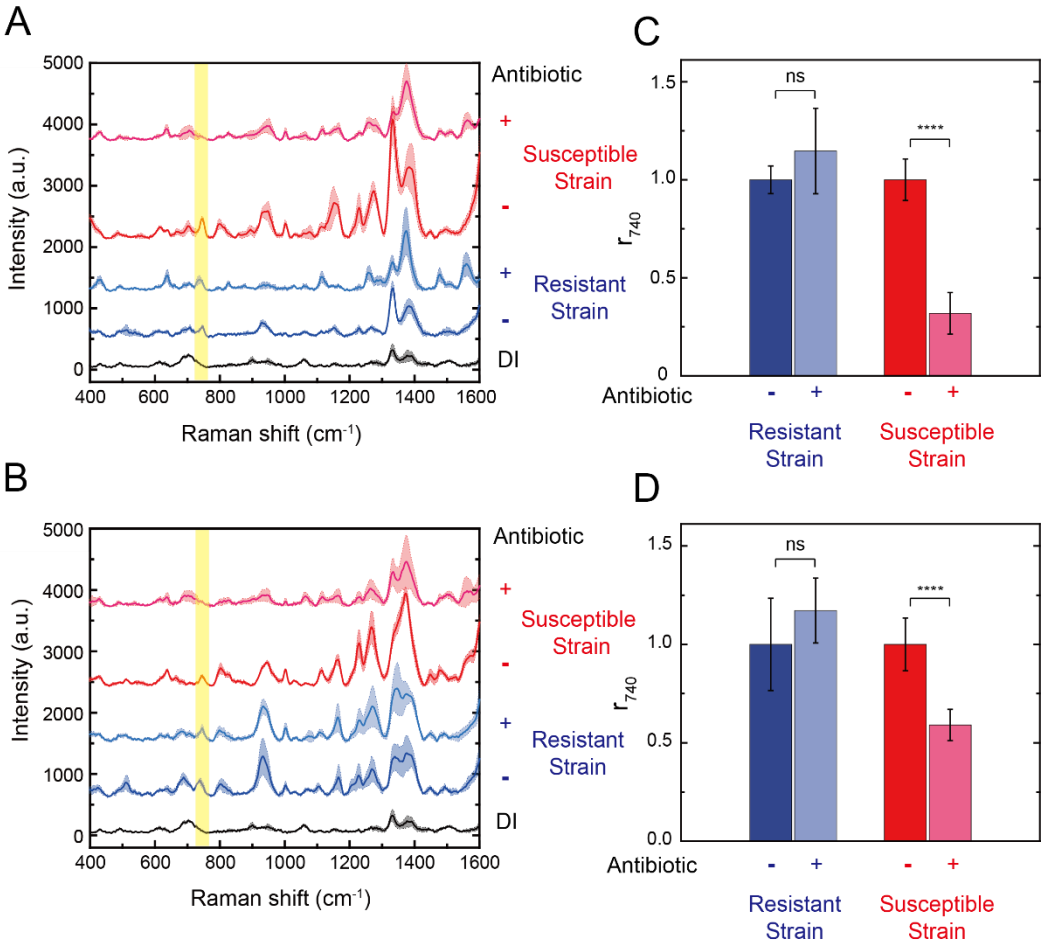
- Bacteria encapsulation images



- Raman mapping image

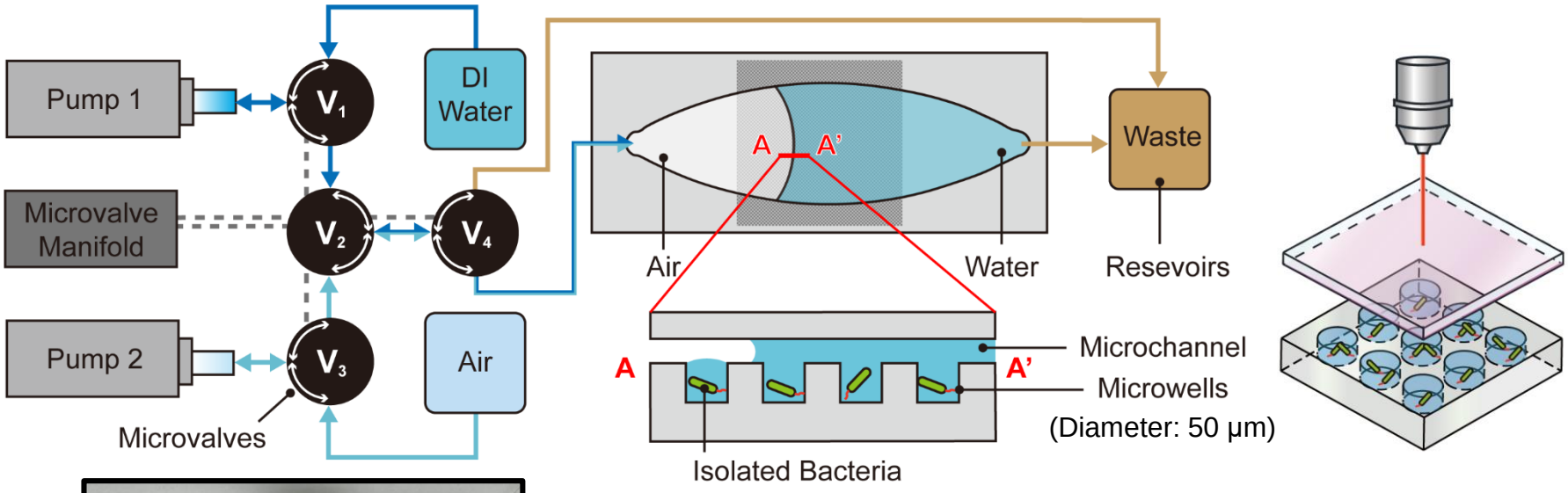


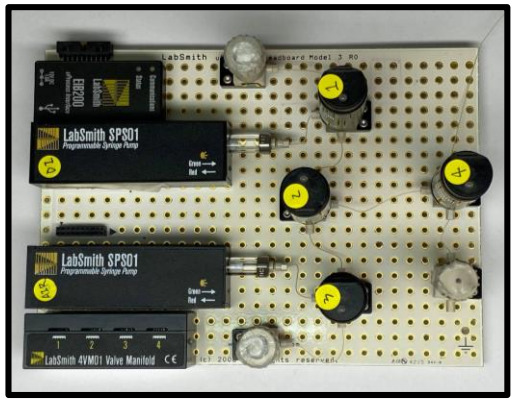
- Antibiotic susceptibility test results of *E. coli* and *S. aureus* treated with antibiotic



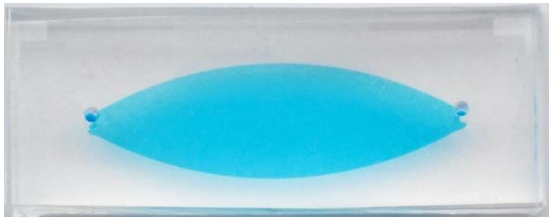
(Huang et. al., Lab on Chip, 2020)

The “automated” microwell-SERS system

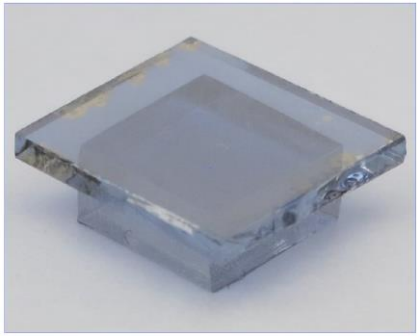




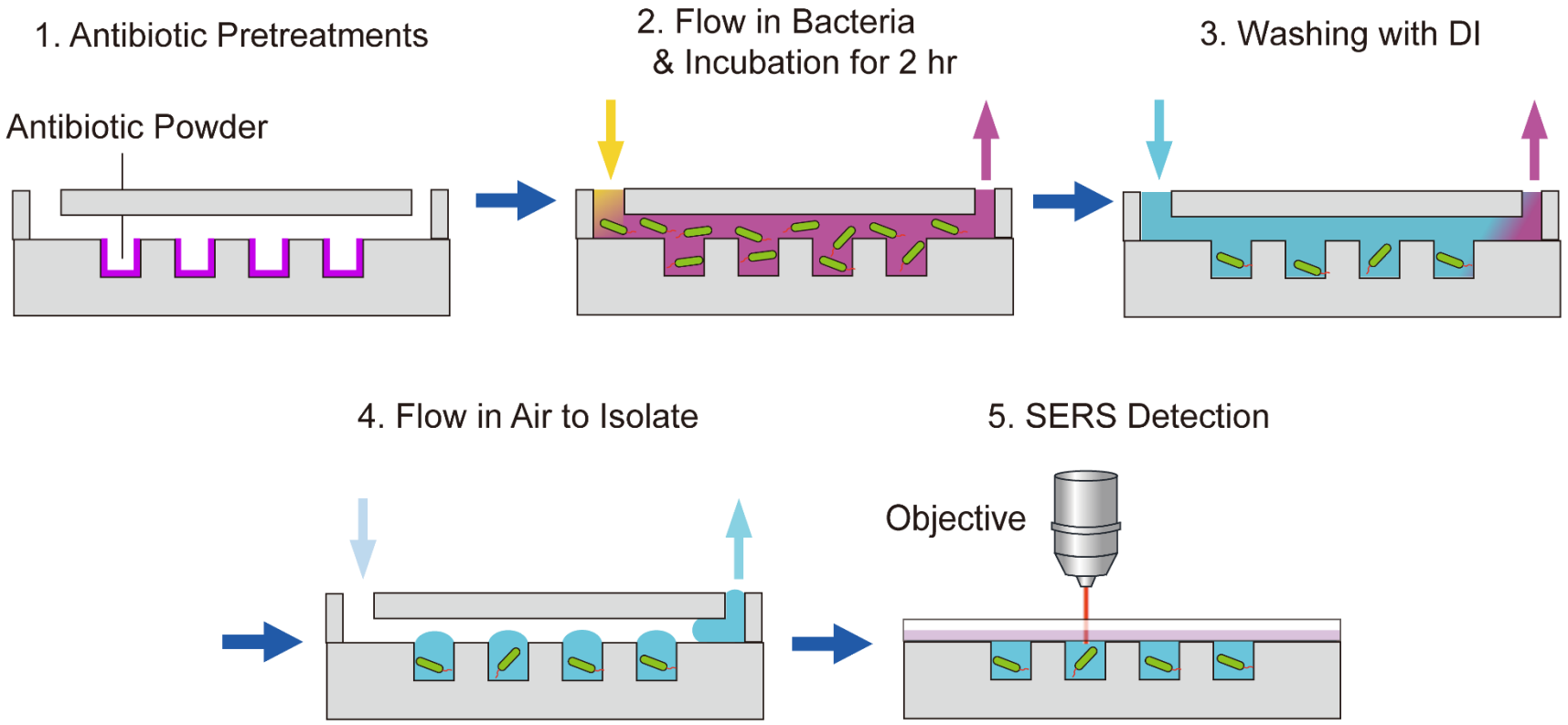
LabSmith System



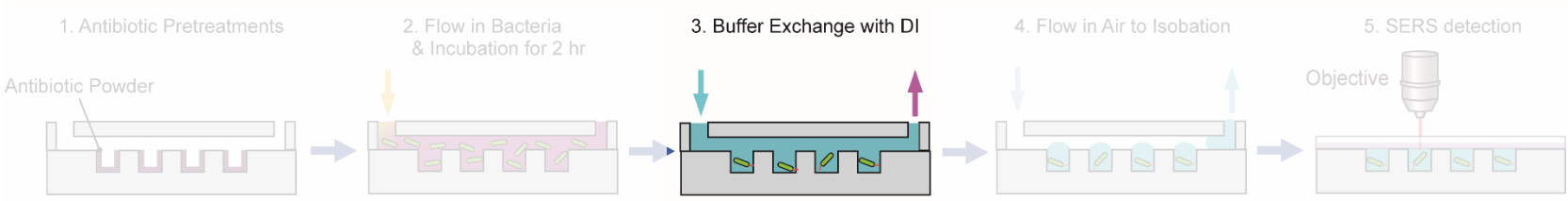
PDMS Microchip 1 mm



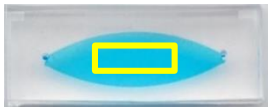
Automated bacteria isolation and washing process

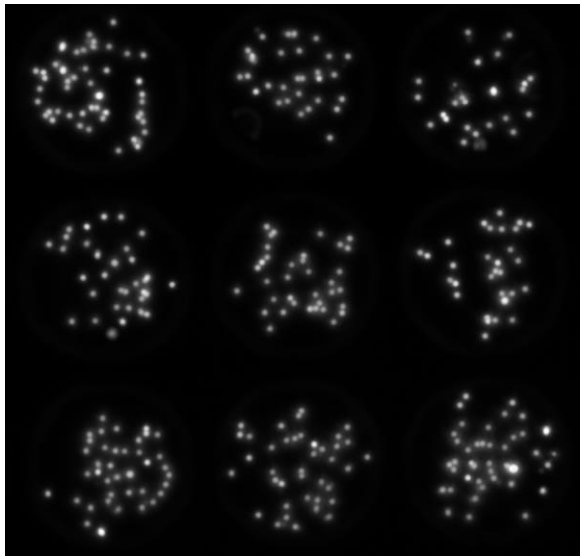


Washing flow rate optimization

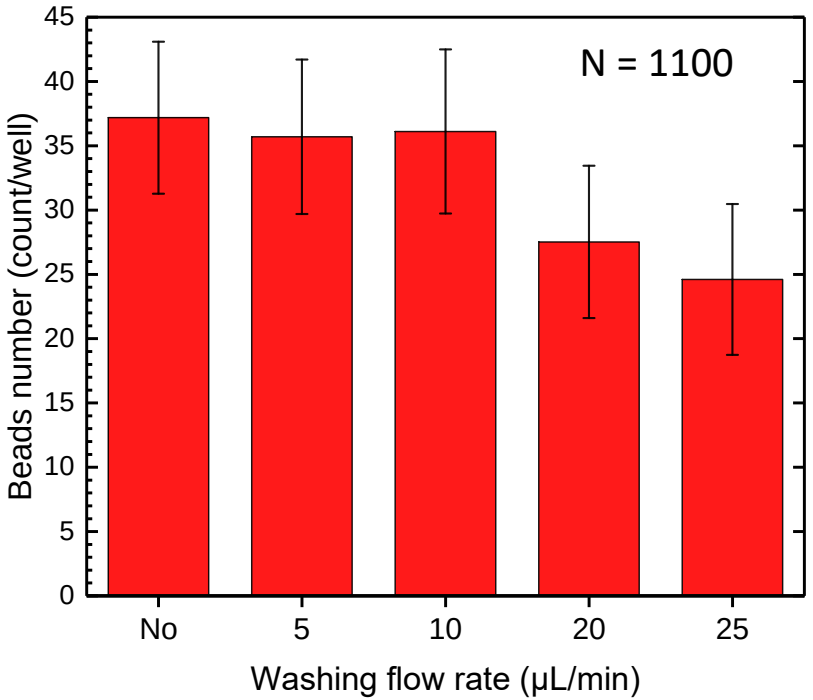


Fluorescence beads concentration:
5 x 10^7 particles/mL (diameter: 2 μm)
Automatic wash 3 min

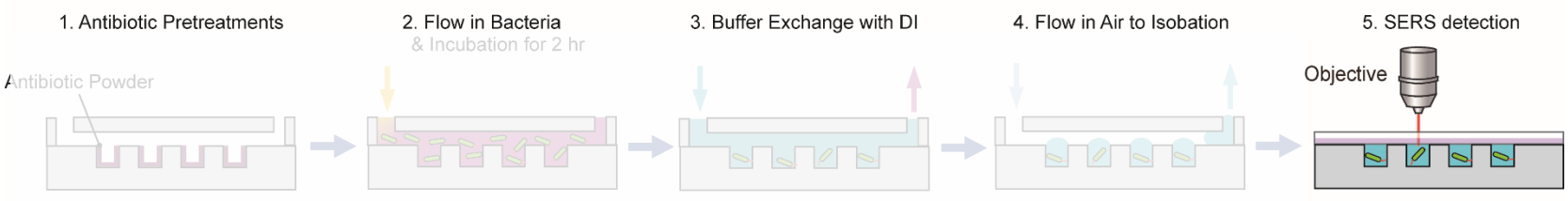


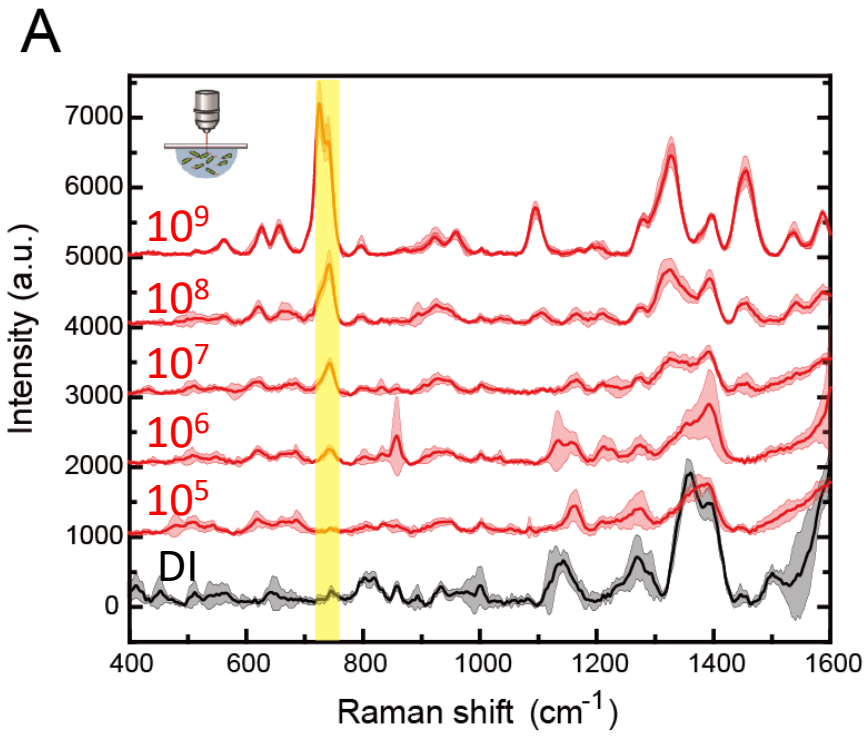


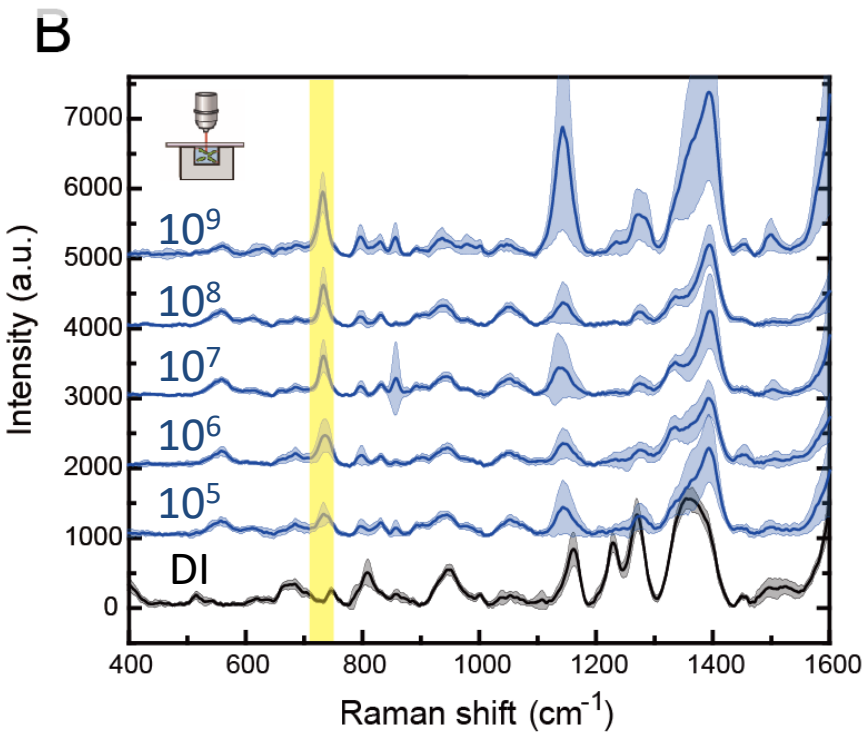
Before wash



The SERS spectrum at various bacteria concentration

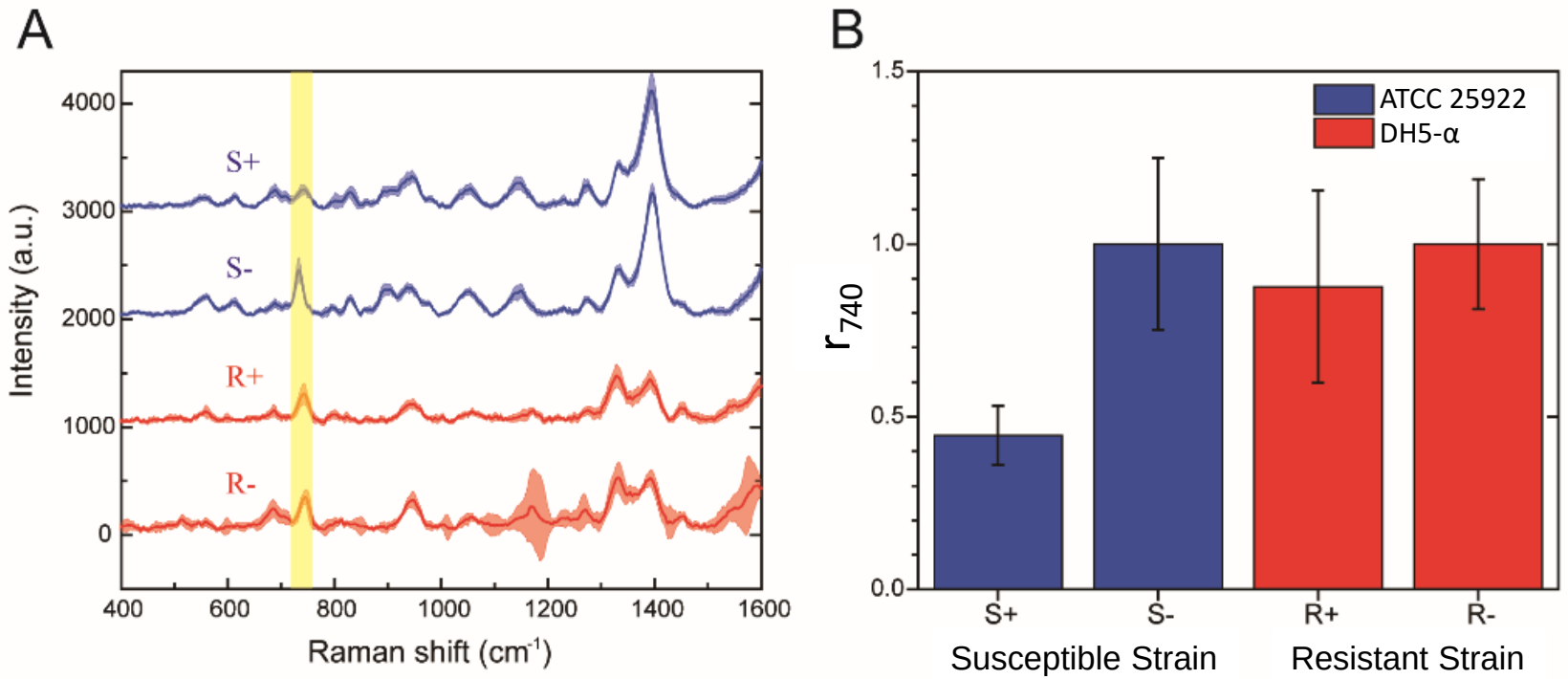






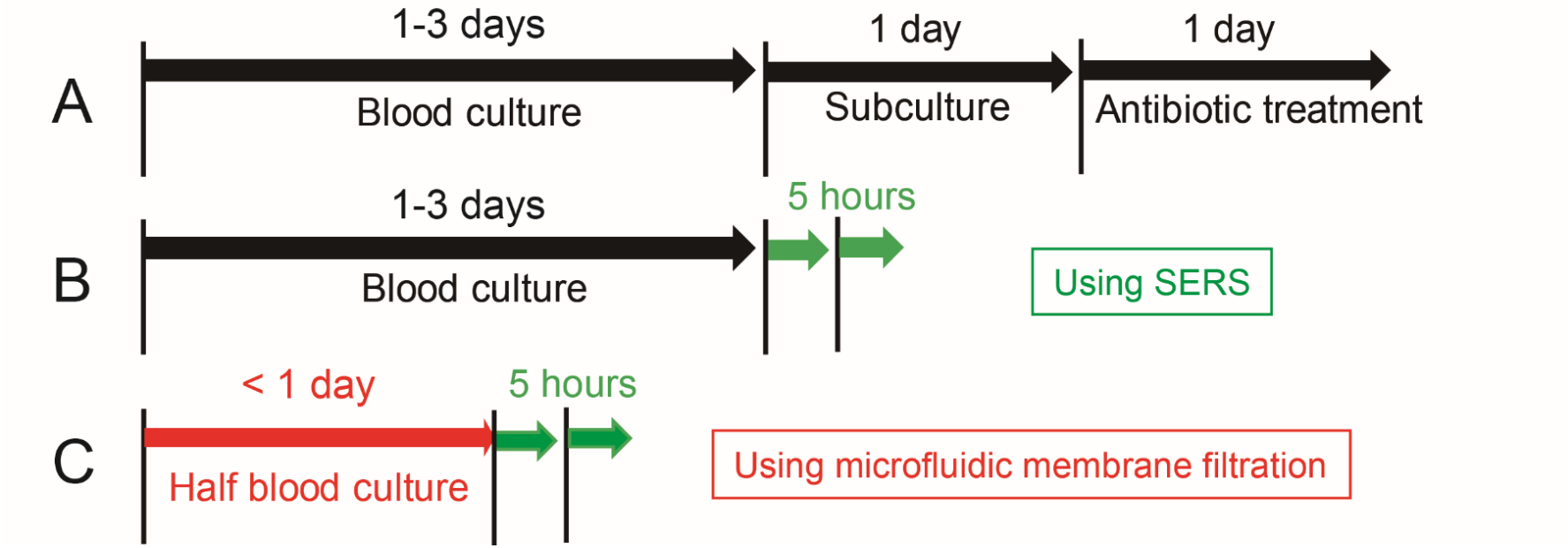
AST results using automated microwell-SERS system

- **Sample:** *E. coli. (ATCC 25922)* - susceptible
  E. coli. (DH5-α) - resistant
- **Antibiotic:** kanamycin 16 μg/mL
- **Bacteria concentration:** 10^8 CFU/mL
- **Antibiotic treating time:** 2 hours



Summary

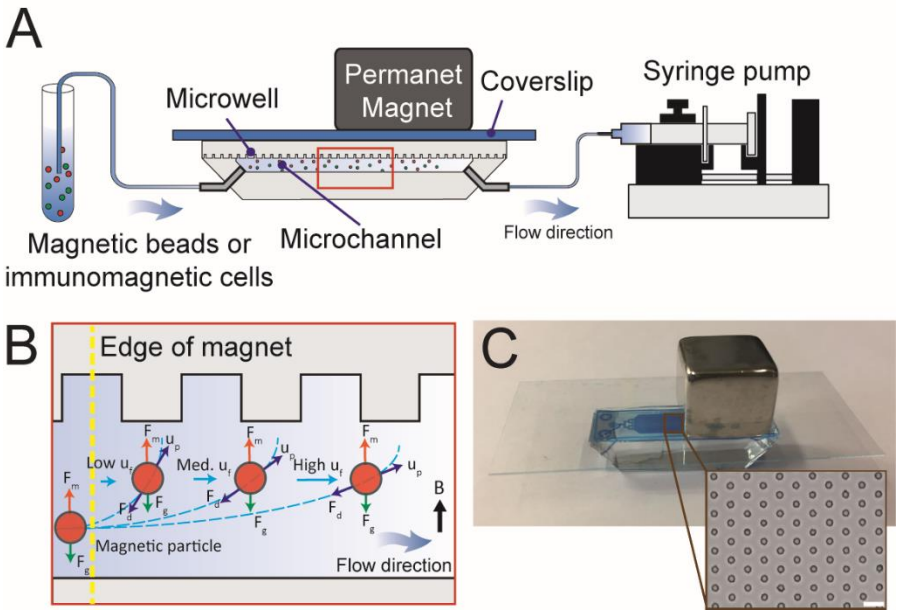
- A microfluidic device integrating **membrane filtration with SERS substrate** for rapid bacteria detection and AST.
- The device enables a **high-throughput (~2mL/min)**, **label-free**, **real time** and ***in-situ*** detection with much **less manual error**.
- The device currently achieves **~10000X** bacteria enrichment.



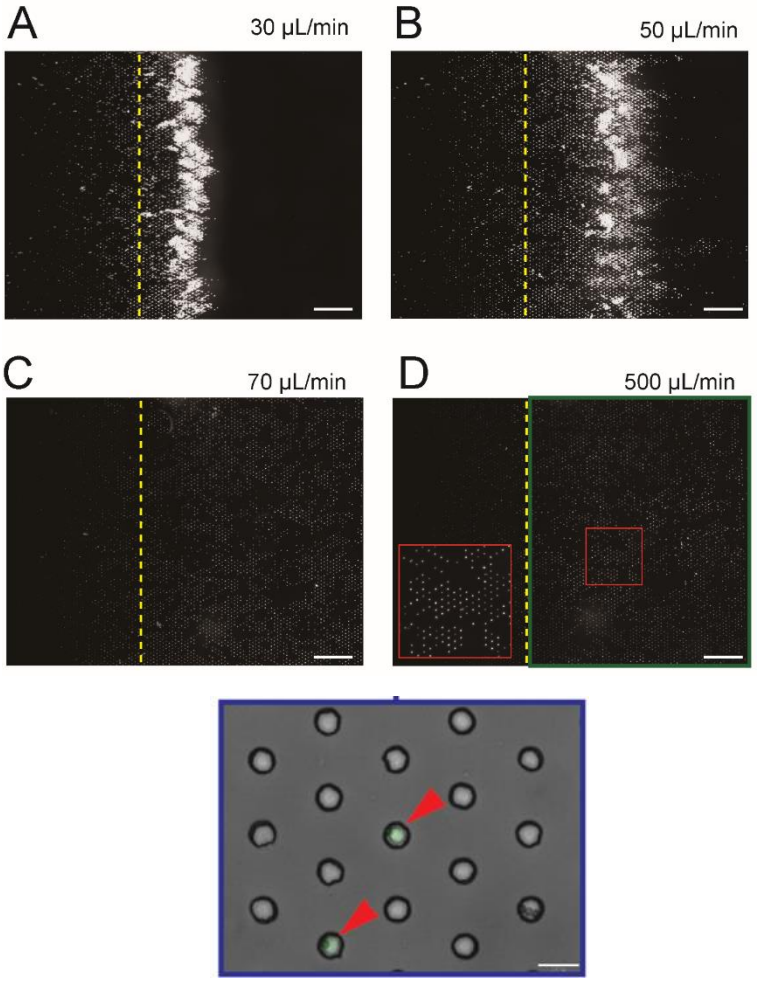
Microfluidics for cell trapping and detection

The microfluidic microwell device for immunomagnetic single cell trapping

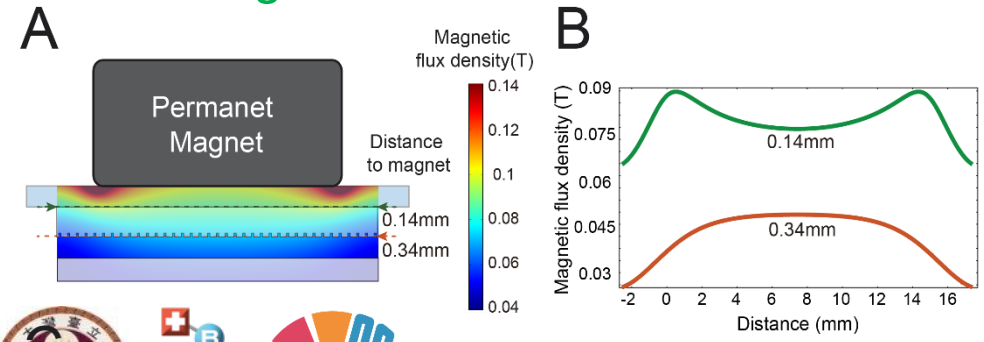
- The microfluidic microwell device



- The sweeping process to enable single particle trapping



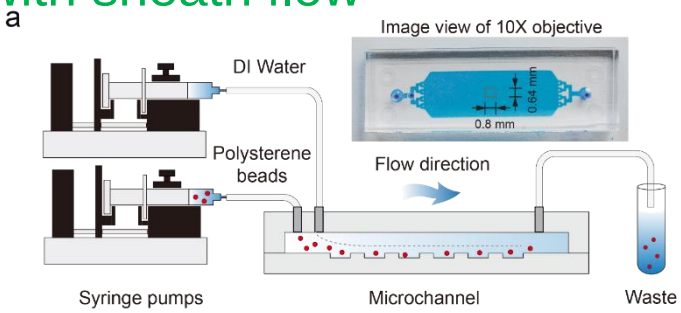
- The magnetic field distribution



(Huang et. al., Micro. & Nanofluidics, 2018)

The microfluidic microwell device for hydrodynamic particle trapping

- The microfluidic microwell device with sheath flow



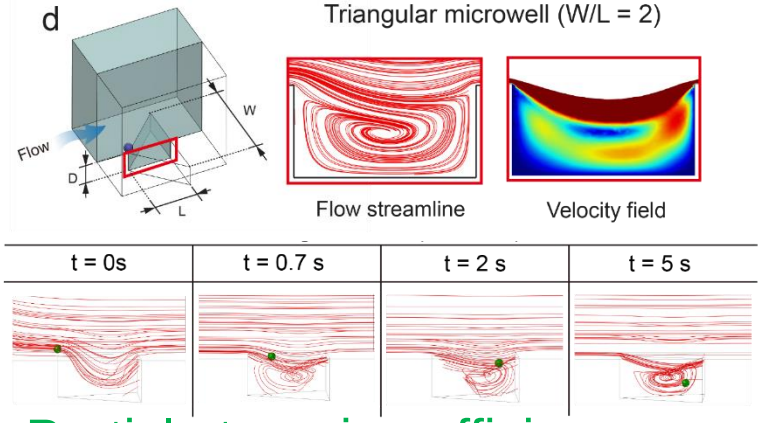
b

	Circular microwell (W = 40 μm, L = 40 μm)	Triangular microwell (W = 40 μm, L = 40 μm)	Triangular microwell (W = 80 μm, L = 40 μm)	
Photo				
Flow rate	15 μL/min	15 μL/min	15 μL/min	15 μL/min
Re	0.05	0.05	0.05	0.05
W/L	1	1	2	2
R/L	0.075	0.075	0.075	0.125
Occupancy	7.1%	28.6%	52.4%	35.7%

c

	Triangular microwell (W = 80 μm, L = 40 μm)	Triangular microwell (W = 40 μm, L = 20 μm)		
Photo				
Flow rate	15 μL/min	7.5 μL/min	15 μL/min	30 μL/min
Re	0.05	0.025	0.05	0.1
W/L	2	2	2	2
R/L	0.125	0.1125	0.1125	0.1125
Occupancy	35.7%	52.4%	30.1%	7.1%

- Flow stream and particle trajectory simulation



- Particle trapping efficiency w/ and w/o sheath flow

	with sheath flow	without sheath flow
Photo		
Flow rate	15 μL/min	15 μL/min
Re	0.05	0.05
W/L	2	2
R/L	0.075	0.075
Occupancy	52.4%	14.3%

(Lai et. al., Micro. & Nanofluidics, 2019)

Tele-health Care for Chronic Renal Failure Patients

- 1.7 million patients suffer from chronic renal failure in the world.
- In Taiwan, there are 70,000 patients
  - >90% patients take hemodialysis in the hospital three times per week => USD 100 per treatment
  - ~9% patients take peritoneal dialysis (PD) at home => cost and time efficient
- The bottleneck of promoting peritoneal dialysis is the early-stage inflammation

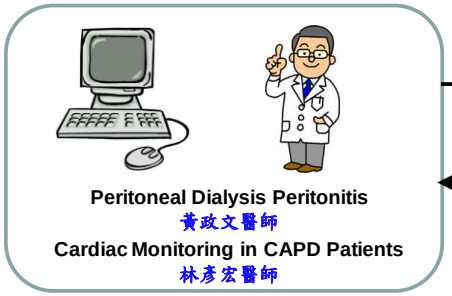
Hemodialysis

Peritoneal dialysis

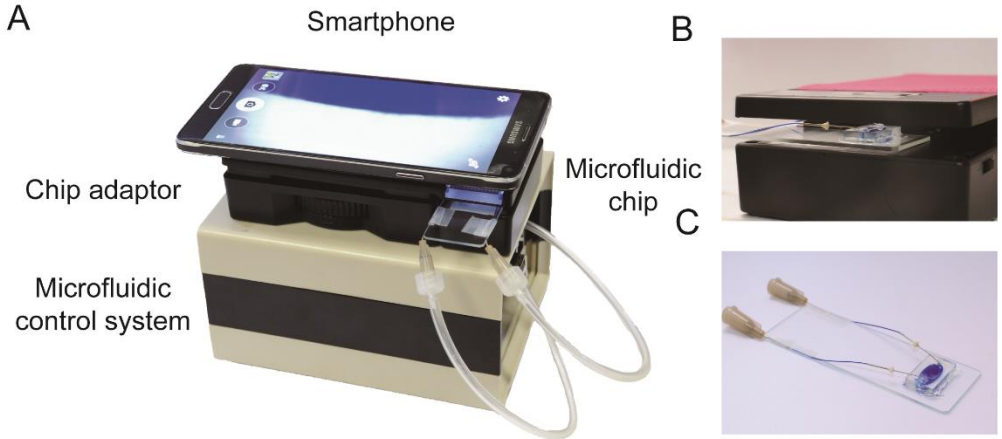
Develop a microfluidic platform to monitor the early-stage inflammation of PD patients

Microfluidics for White Blood Cell Counting

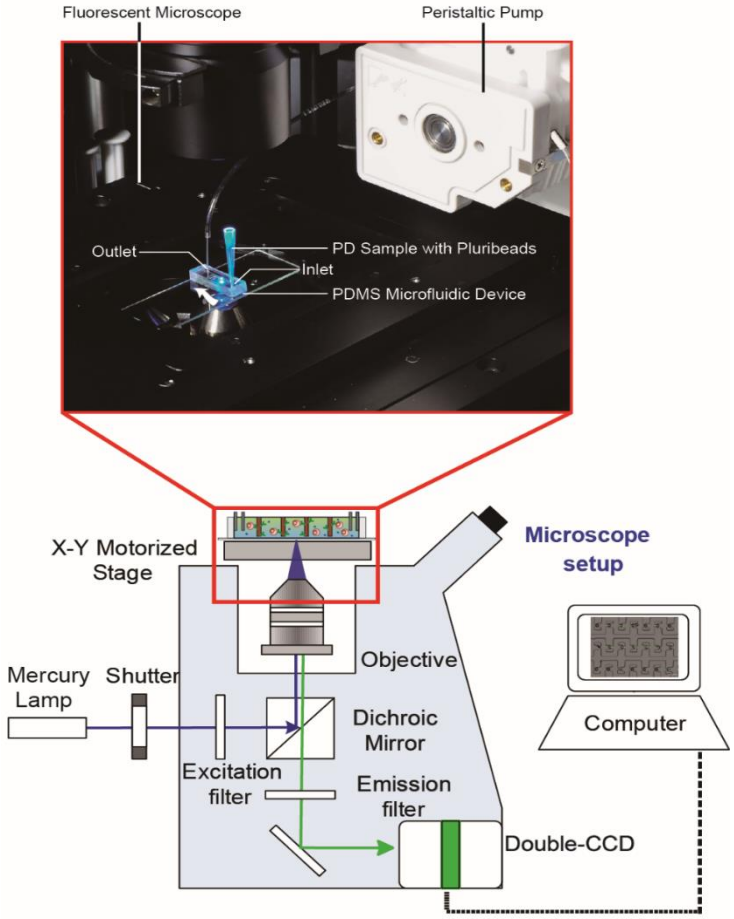
- Key parameters of early-stage inflammation of PD patients
  - 100 WBC/μl and >50% neutrophils in PD solution



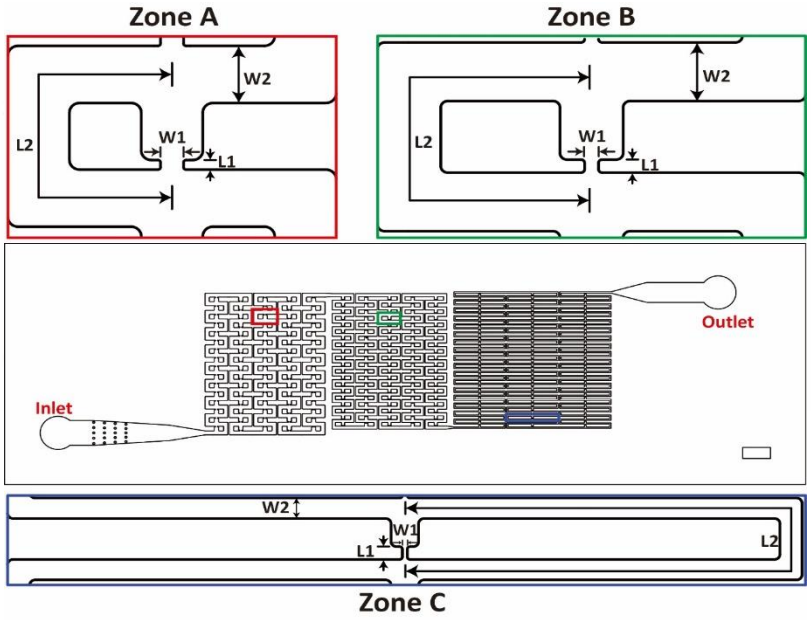
NTU Hospital

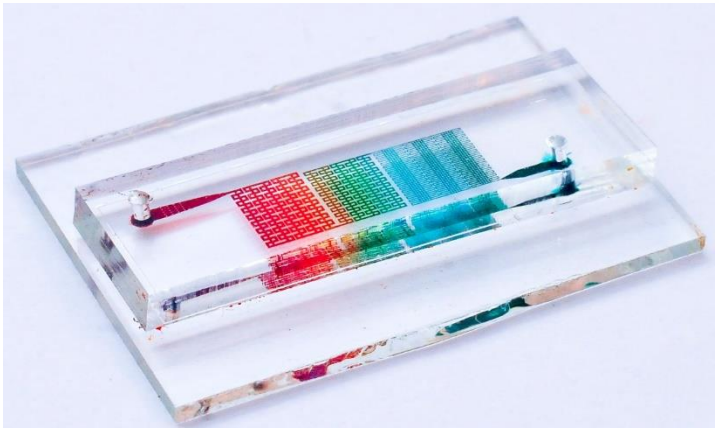


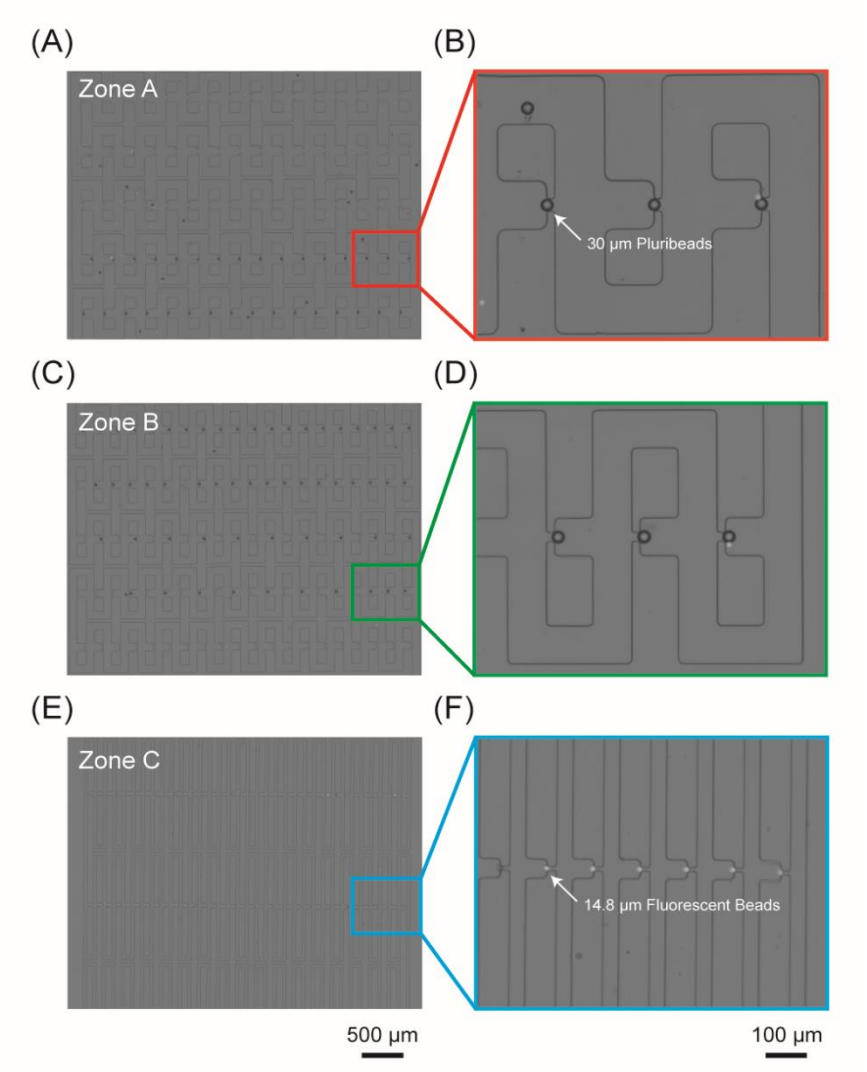
Mobile Device



The Microfluidic Device with Hydrodynamic Trap Arrays

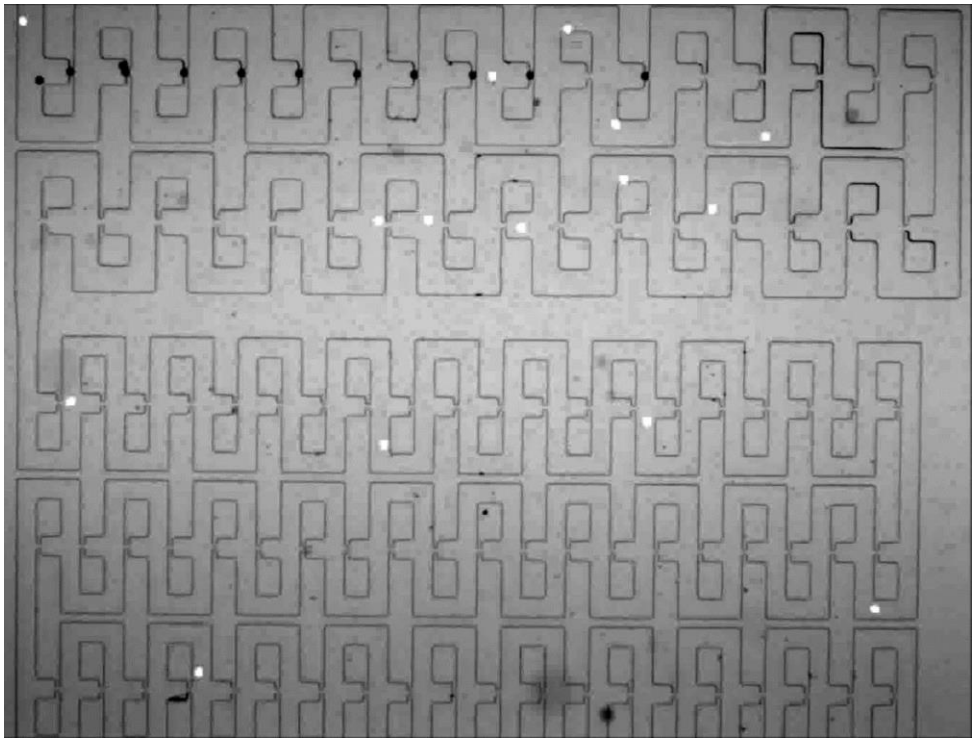






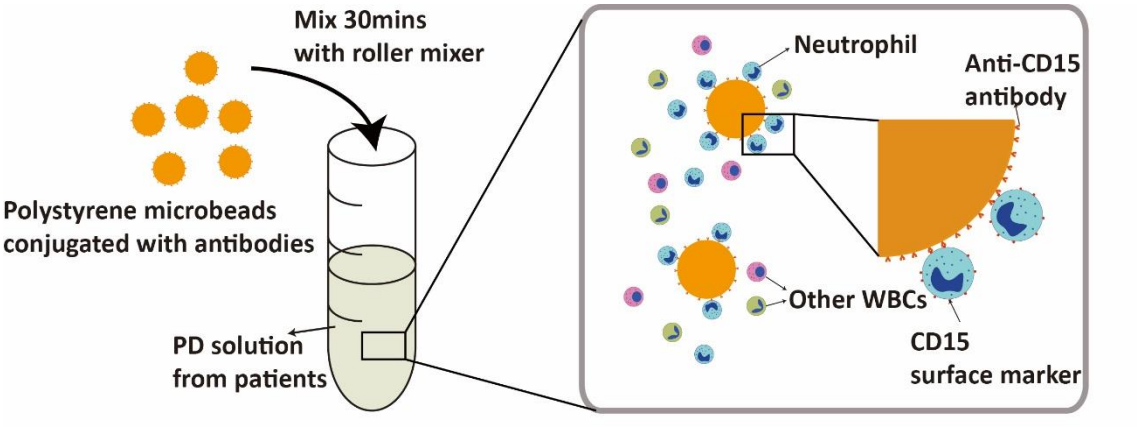
15 μm Fluorescent Beads and 30 μm Pluribeads Trapping

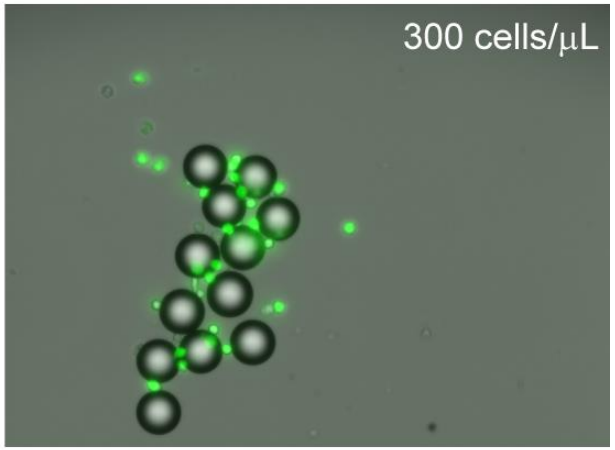
- Pluribeads: 30μm, concentration: 20/μL
- Fluorescent beads: 14.8μm, concentration: 50/μL
- Flow rate: 2μL/min
- Sample volume: 10μL

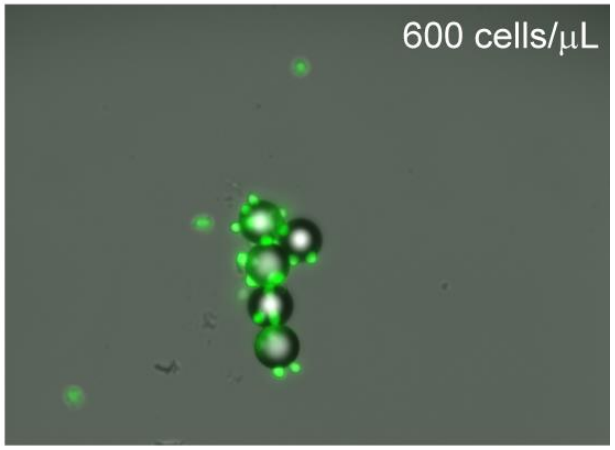


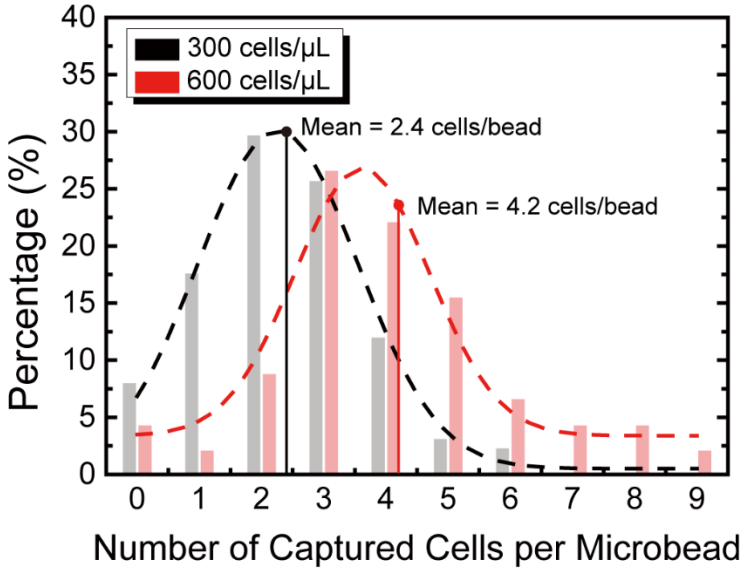
4x speed

Neutrophils Conjugated to 30μm Pluribeads





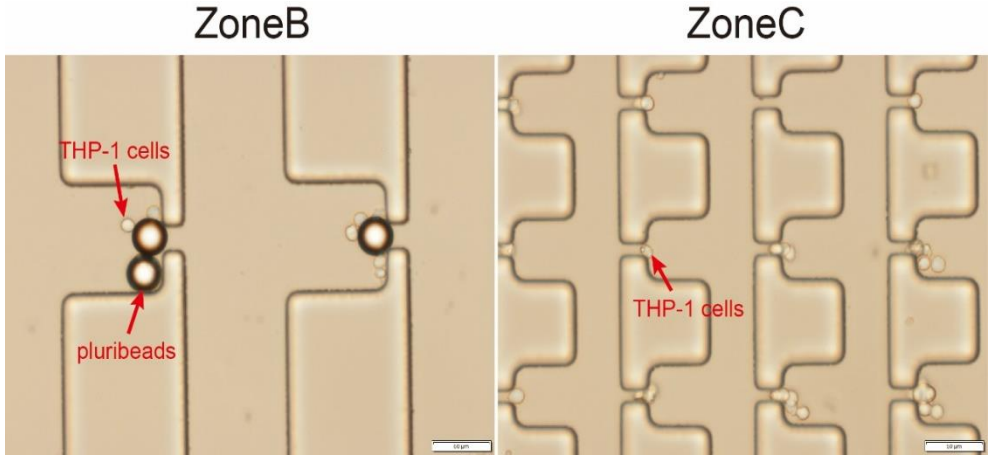


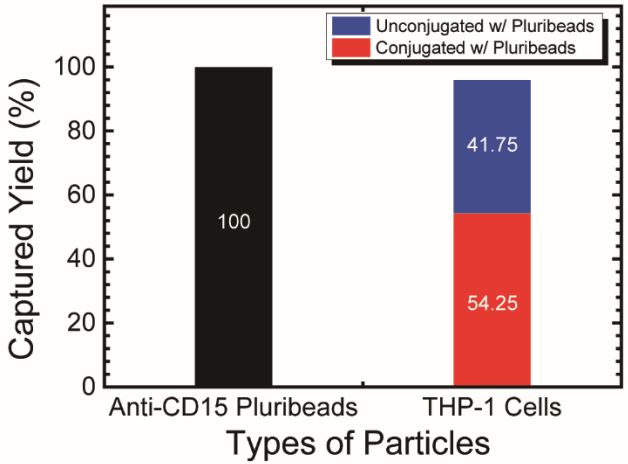


Microbeads capture more cells under higher concentration

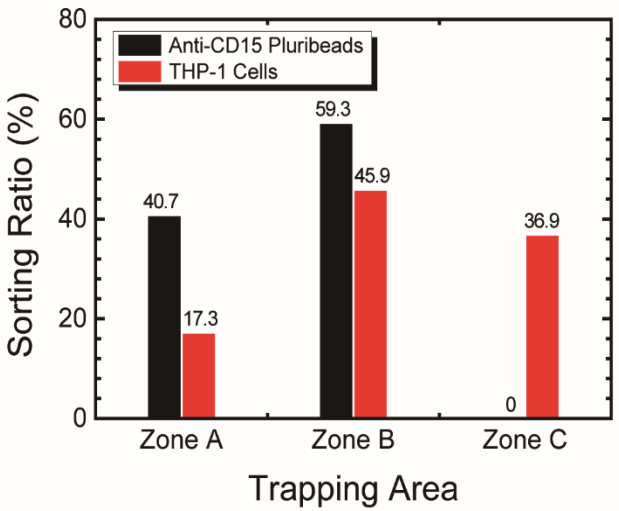
THP-1 cells conjugated to 30 μm Pluribeads

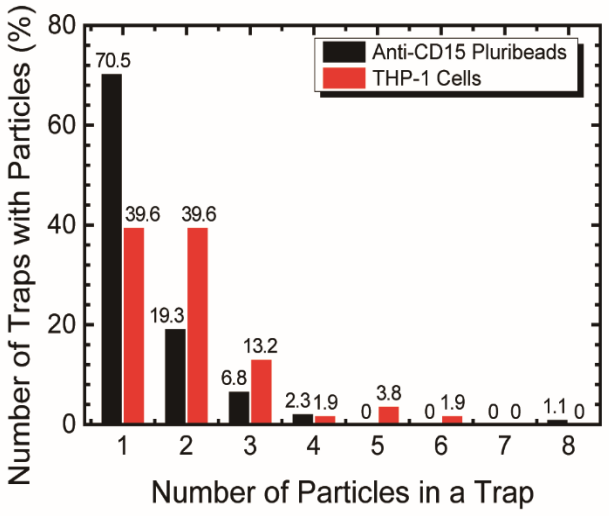
- Pluribeads and THP-1 cells trapping image under flow rate: 0.05μL/min





- 100% pluribeads trapping rate
- 96% THP-1 cells trapping rate
- THP-1 cell has CD15 expression

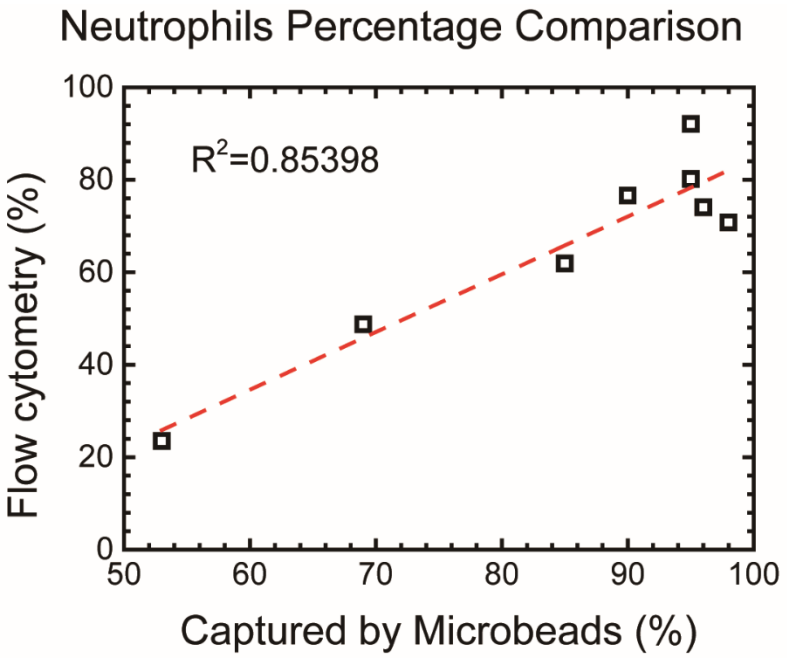




Neutrophils Concentration in PD Solution

- The neutrophils percentage comparison between flow cytometry and microfluidics

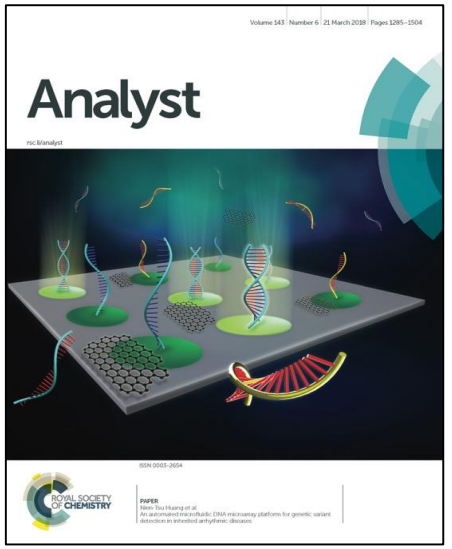
Patients Number	Neutrophils Percentage		Total WBC Concentration (μL)	
	Flow Cytometry	Captured by Microbeads	Flow Cytometry	Counting Chamber
Patients 1	96%	74.03%	2567	
Patients 2	85%	61.88%	341	
Patients 3	95%	80.18%	1517	
Patients 4	90%	76.59%	6395	
Patients 5	98%	70.77%	3792	
Patients 6	95%	92.60%	4166	
Patients 7	69%	48.75%	735	
Patients 8	53%	23.50%	15273	11700

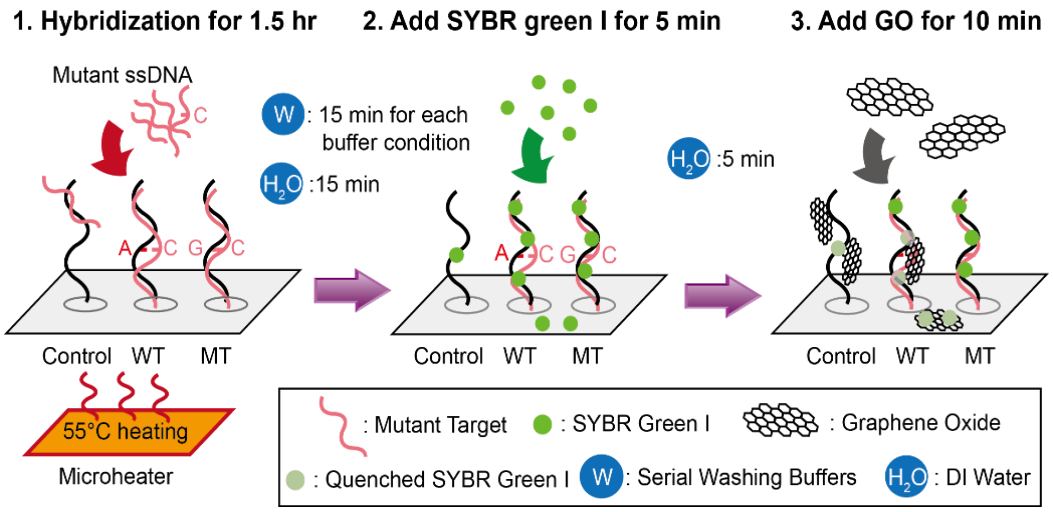


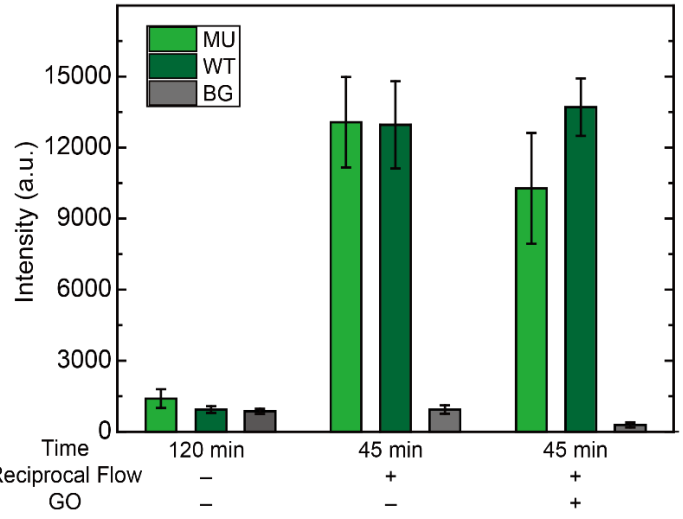
Microfluidics for DNA microarray hybridization

Microfluidics for Personalized Medicine

- Conventional genetic tests are usually time-consuming (~4-8 weeks) and expensive (USD 3000–5500)
- The microfluidic DNA microarray platform for genetic variants detection in inherited arrhythmic diseases, such as long QT syndrome (LQTS), Brugada syndrome (BrS)
  - Single nucleotide polymorphism (SNP)
  - Graphene Oxide (GO) to inhibit non-perfectly matched DNA



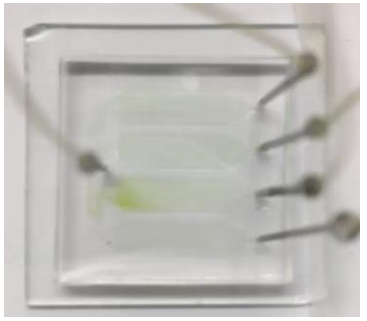


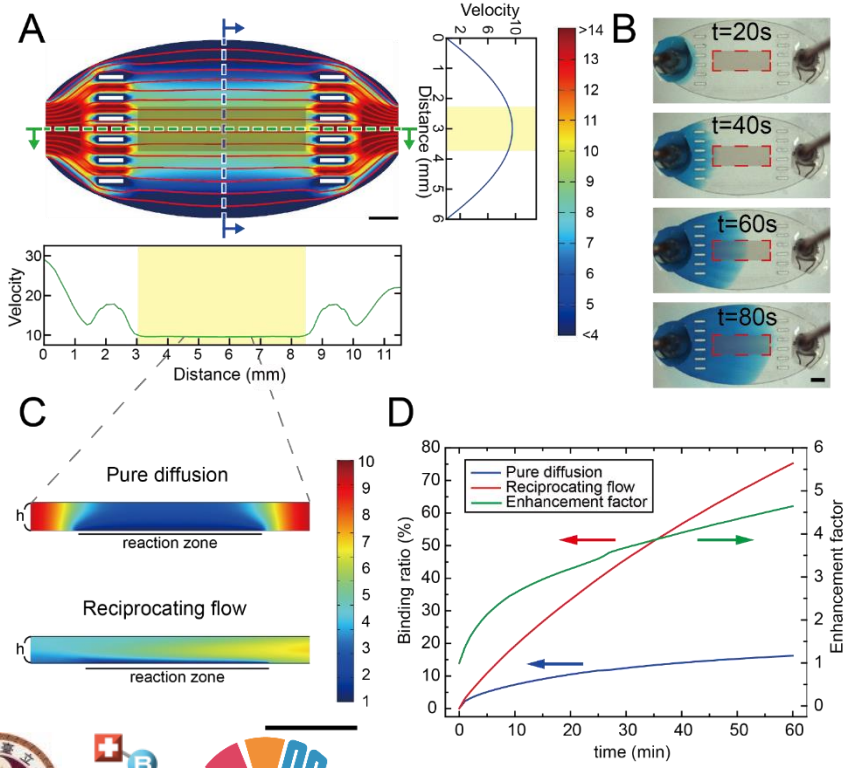


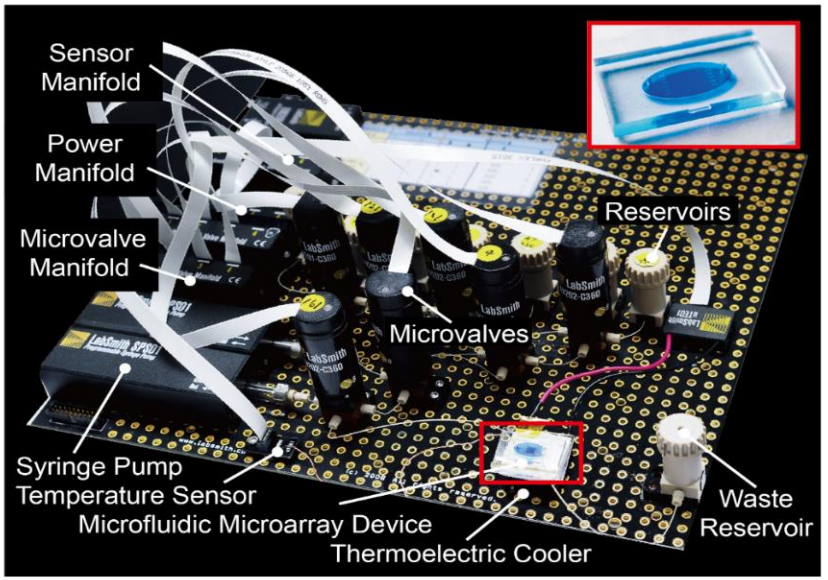
(Huang et. al., Analyst, 2018)

Automated DNA hybridization process

- Automated and precise fluidic control
  - Active mixing, temperature control
  - Total assay time: <3 hours
  - Required blood volume: 20 μL





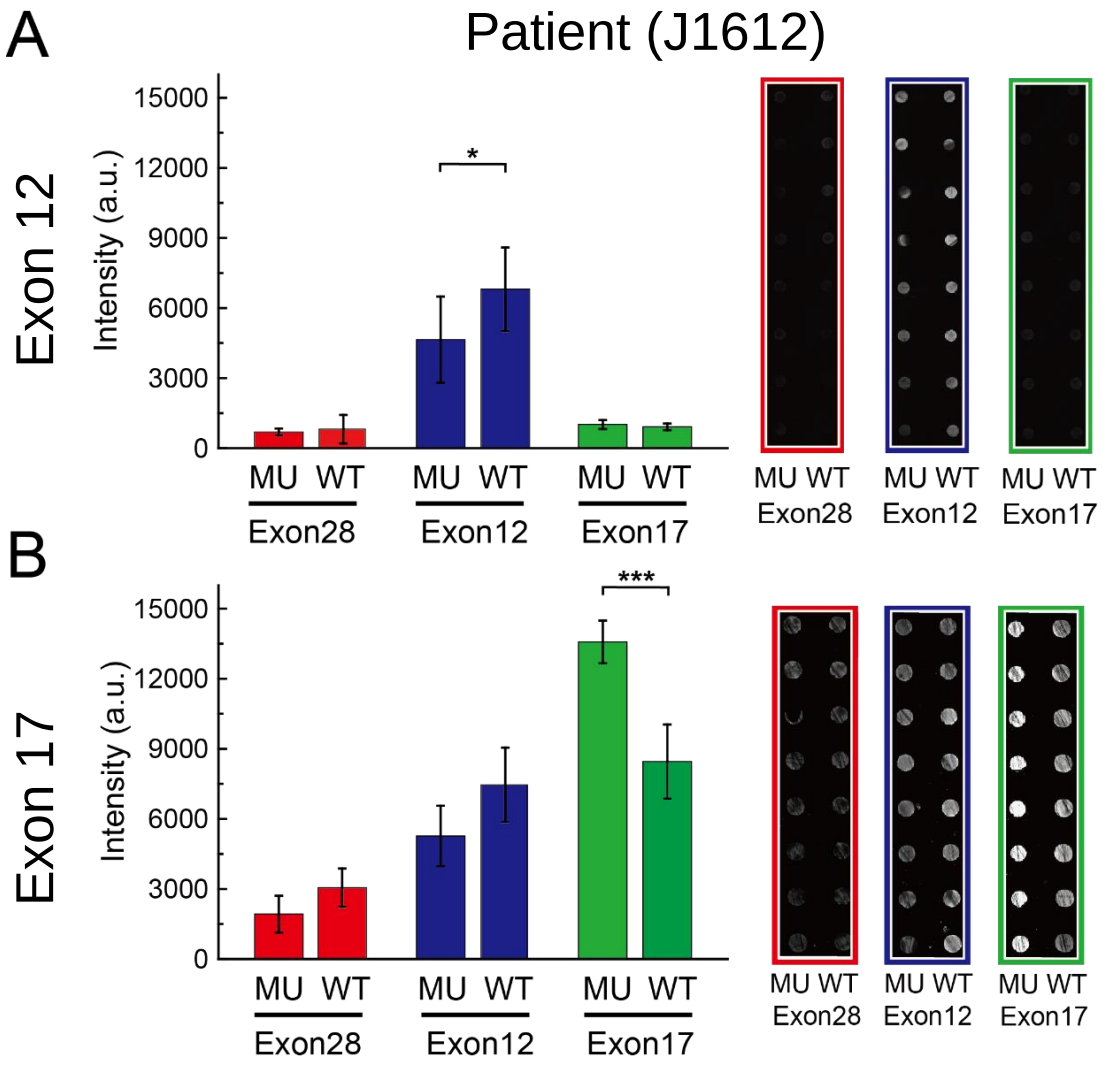


(Huang et. al., Analyst, 2018)

LQTS Clinical Sample Validation

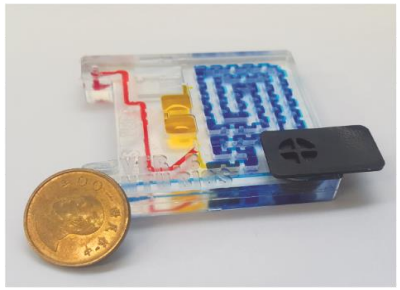
- The microfluidic DNA hybridization device
  - SNP detection by GO + SYBR green I
  - Automatic reagent control and rapid DNA hybridization
  - Distinguish three clinical samples with specific exon mutation

Exon	Sanger result
12	WT
17	MU



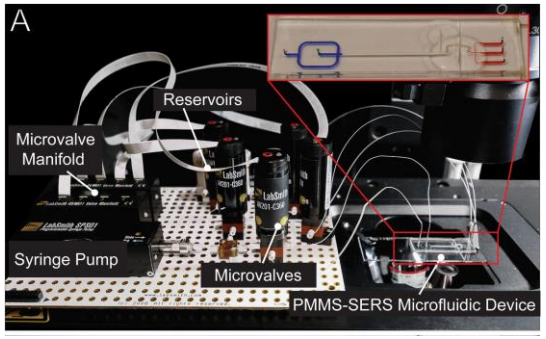
(Huang et. al., Analyst, 2018)

Disease	Diabetes	Periodontal dialysis	Leukemia	Bacterial Infection	Genetic Diseases	Blood Counting
Markers	Glycated Hemoglobin (HbA1C)	White Blood Cells (WBC)		Bacteria & Metabolites	Primary cilia	Mutated DNA sequence	WBC, RBC, Plasma

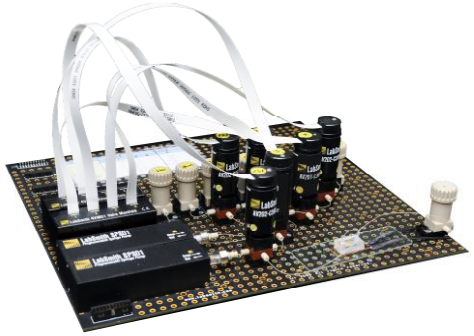


(Kuan et. al., Lab Chip, 2016)

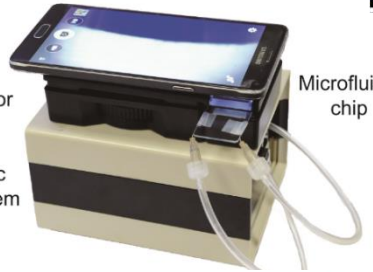
Smartphone

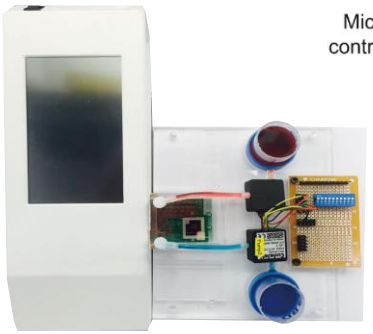


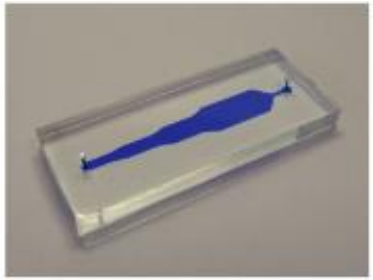
(Wang et. al., Microfluid. and Nanofluid., 2019)



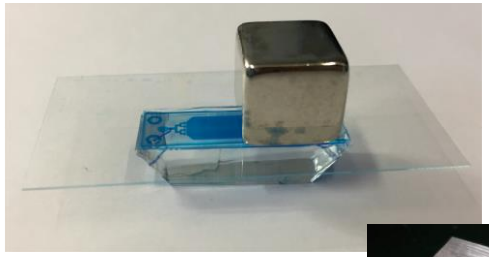
(Huang et. al., Analyst, 2018)





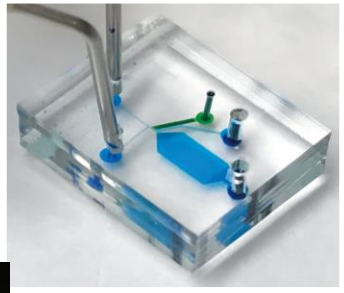


(Chu et. al., Biomicrofluidics, 2019)



(Huang et. al., Microfluid. and Nanofluid., 2018)

(Chang et. al., Anal. Chem. 2019)



(Kuan et. al., Scientific Report, 2018)

Conclusions

- We developed various microfluidic platforms for personalized medicine and healthcare
  - whole blood processing and in-situ analyte detection
  - bacteria enrichment followed by antibiotic susceptibility test (AST)
  - Cell trapping and counting for disease diagnosis
  - rapid and automated DNA hybridization process and SNP analysis

- The microfluidic platforms could potentially
  - improve the quality of medical care and enables long-term health monitoring in point-of-care settings
  - eliminates the cost and time of sample preparation process




Thank you !!!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