Lecture #9

Fundamentals of microfluidics and microfluidic chips

Aims:

- Understand the behavior of liquids at the microscale
- Learn the basics about continuous flow and droplet microfluidics
- Get an overview on the basic chip manufacturing principles
- Get an overview on the different droplet modules and functionalities
- Prominent applications

Lectures (PHH 331)	Date & Topic	Details	Practical
1	12.09 General Intro	Get to know teachers, TAs, students and aims of the course	16.09 Measure temperature using thermistor (using M&A explorer) TL
2	19.09 Lecture LabVIEW TL Group formation (4 students, each)	Some first basic steps in LabVIEW programming	23.09 Brief intr… / thermistor program (input and output) TL
3	26.09 Case study FACS, similarities and differences to droplet microfluidics Selection of case study topics	1.) Property to measure? 2.) Device? 3.) Working princip… 4.) Alternati…	…ion of bioinstrument case study
4	03.10 Preparation of bioinstrument case study		07.10 Tour through LBMM workstation labs, intro into Nature Protocols (all groups)
5	10.10 All groups presenting case study		No practical
6	17.10 Lecture optics Homework: Stu… …e laser/PMT blueprint (by 21.1…	Mirrors, filters, microscope setup, lenses, etc.	21.10 Holidays
	24.10 Holidays		28.10 Build workstation optics 1
7	31.10 Lecture electronics	FPGA, PMTs, amplifier, function generator	04.11 Build workstation 1 optics 2, laser alignment
8	07.11 Intro into enzyme concentration measurement experiment (kinetics, etc.) + task FP	Enzymes, kinetics, practical task	11.11 Build workstation electronics
9	14.11 No lecture	Software similar to Thermistor program, pdf on installation	18.11 Build workstation software: Add output LED (mimicking sorting trigger) into analysis software
10	21.11 Fundamentals of microfluidics and microfluidic chips	Flow at the microscale, microfluidic chips (manufacturing), droplet microfluidic modules	25.11 Microfluidics practice session
11	28.11 Prepare presentation		02.12 Determine enzyme concentration in microfluidic droplets
12	05.12 Prepare presentation		09.12 Sorting Demo on LBMM workstation1 (all groups)
13	12.12 Groups presenting results (all groups) 15.12 Submit report (all!)		**16.12 – TUESDAY! - Individual Q & A sessions (10min, each)**

Last lecture before YOUR microfluidics experiment!

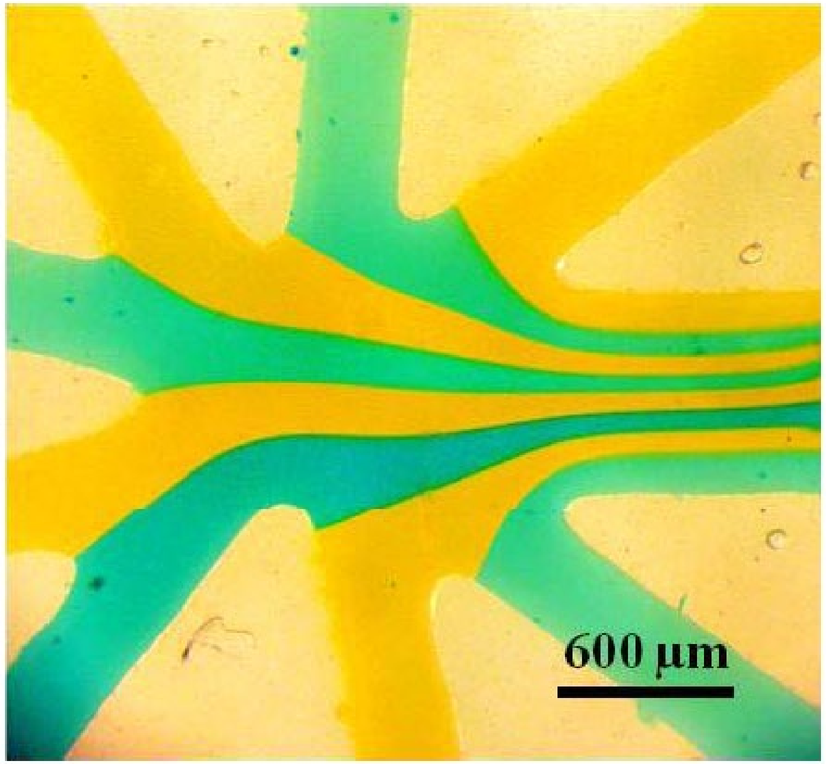
Green shading: Single seminar/practical with all 18 students
Red shading: Individual seminar/practical with only 6 students required (= 3 sequential 90min slots, 4.5h in total)

A general difference between macroscopic and microscopic flow

What is happening here?

https://www.azom.com/article.aspx?ArticleID=14131

Macroscopic flow is turbulent



http://alcheme.tamu.edu/?page_id=6720

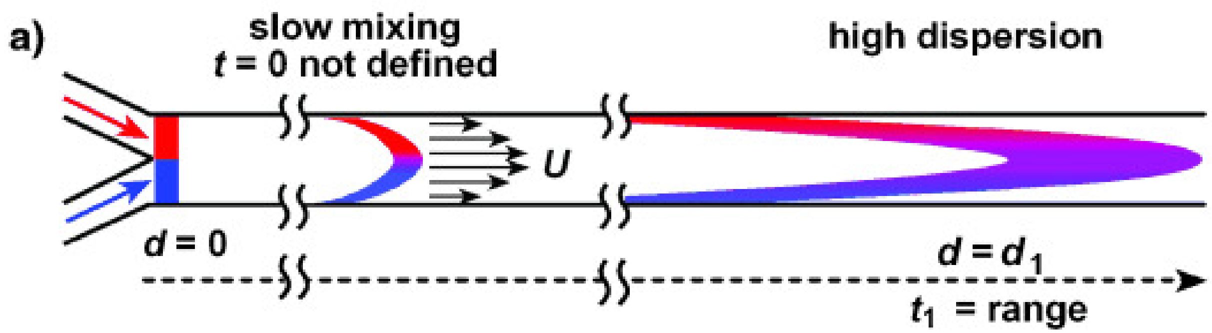
Microscopic flow is laminar

A general difference between macroscopic and microscopic flow

“Consider the act of mixing sugar cubes in tea: waiting for the sugar to diffuse and mix is too slow, and rapid mixing is achieved by stirring which induces turbulent flow (convectional mixing). If the tea is replaced by a viscous liquid, stirring becomes harder, because viscosity damps any motion reducing turbulence and diffusive mixing dominates. ***In microfluidic devices, viscous forces dominate****. But instead of changing the liquid, the same effect is obtained by reducing size. Scaling a tea cup to less than a millimeter gives an approximation of the fluid physics in a microfluidic system.* ***This fluid flow regimen in microfluidics is laminar and not turbulent****.”*

Vyawahare S. et al., Chem Biol. 2010

A general difference between macroscopic and microscopic flow



Small channel diameters imply that **fluid moves very close to the static walls**
=> friction and viscosity become more relevant

Song, H., Tice, J. D., & Ismagilov, R. F. (2003). A Microfluidic System for Controlling Reaction Networks in Time. Angewandte Chemie International Edition, 42(7), 768–772.

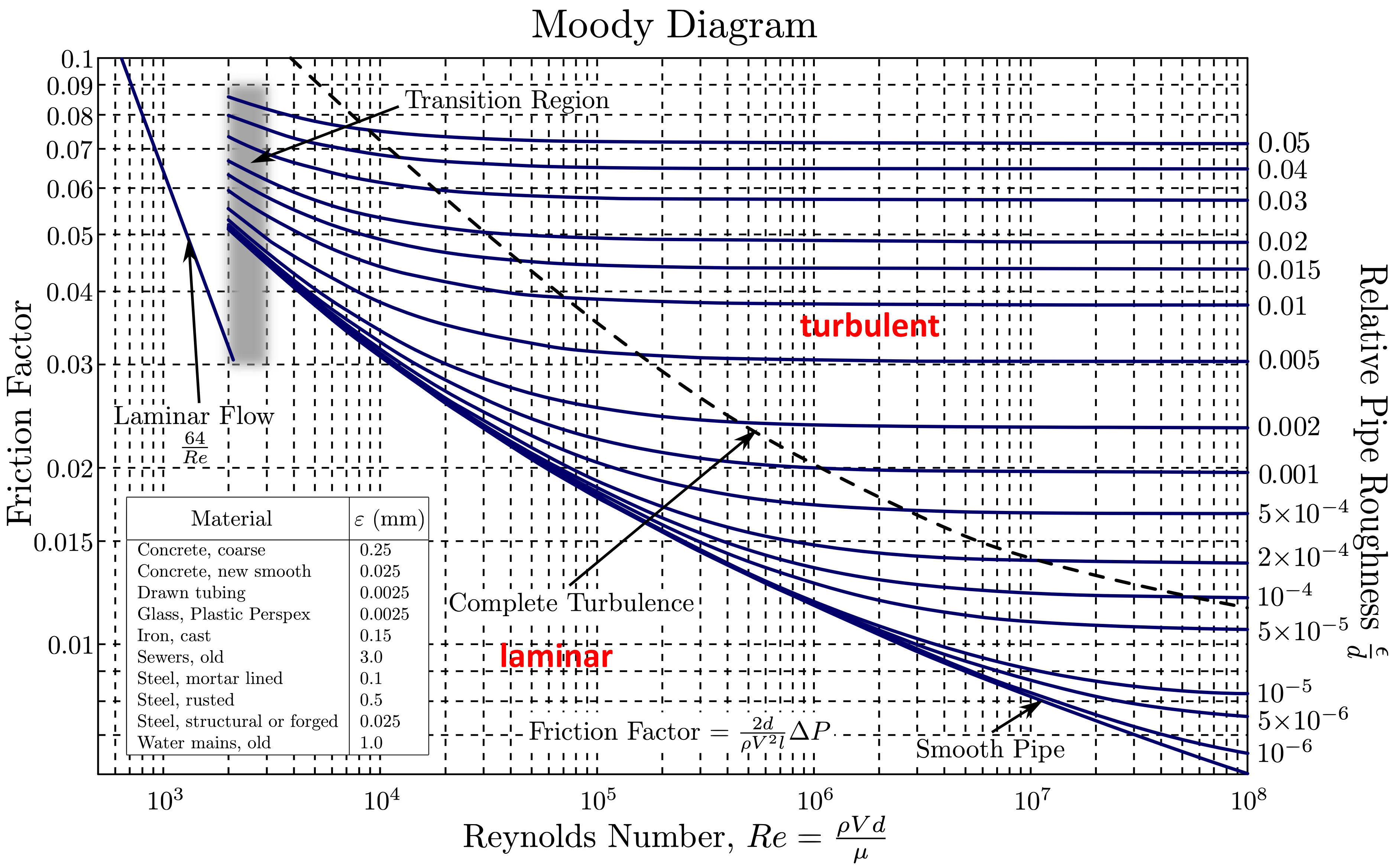
Reynolds number – is the flow laminar or turbulent?

$$Re = \frac{\rho V L}{\mu}$$

With ρ = density of the fluid, V = velocity of the fluid, L = length of the channel and μ = viscosity of the liquid.

For Re < 2300, flow is laminar, above 4000 it becomes entirely turbulent. Exact numbers depending on roughness of channel wall (see Moody diagram)

Reynolds number – is the flow laminar or turbulent?



https://commons.wikimedia.org/w/index.php?curid=4681366

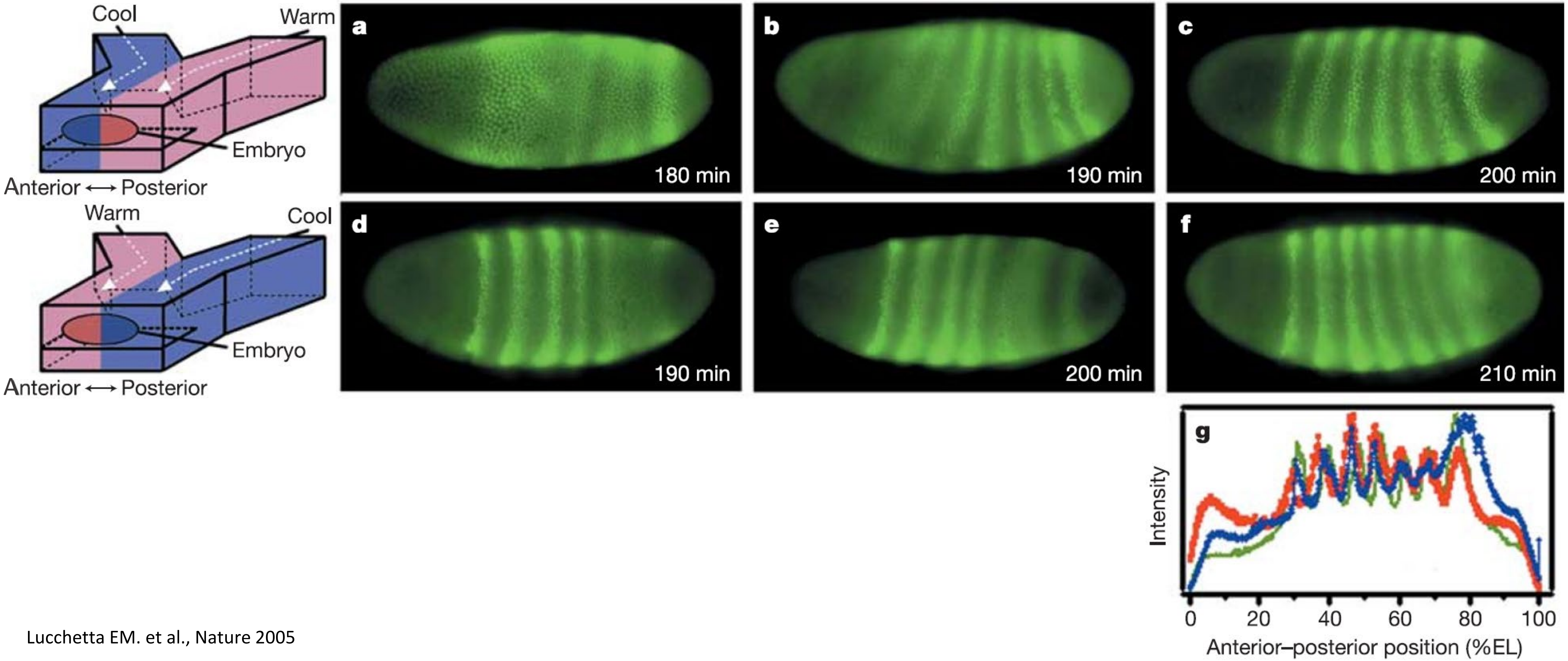
Peclet number – what drives mixing?

$$Pe = \frac{VL}{D}$$

where D is the diffusion constant, V the velocity of the fluid and L the length of the channel.

For low Pe (<1), mixing is only determined by diffusion, so very slow. This enables very special experimental setups:

Application: Measuring the effect of temperature on embryonic development, discovery of compensation mechanisms

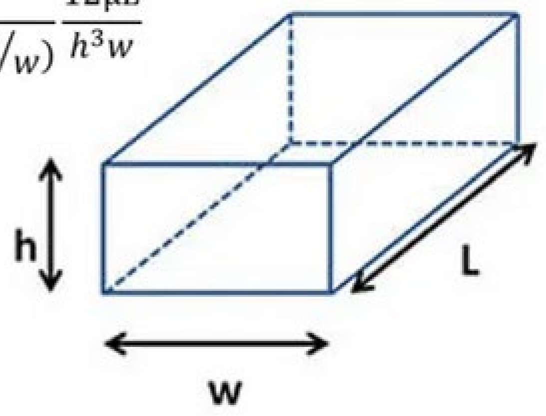


Lucchetta EM. et al., Nature 2005

Other relevant parameters in continuous flow at the microscale - resistance

Rectangular cross section

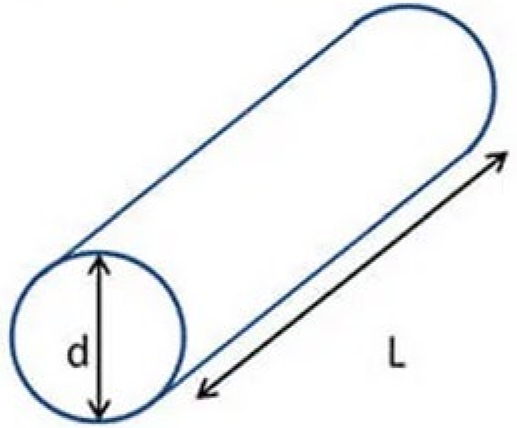
$$R = \frac{1}{1 - 0{,}63\left(h/w\right)} \frac{12\mu L}{h^3 w}$$



L = length of the channel (L>>w)
h = height of the channel
w = width of the channel (w>>h)
μ = dynamic viscosity

Circular cross section

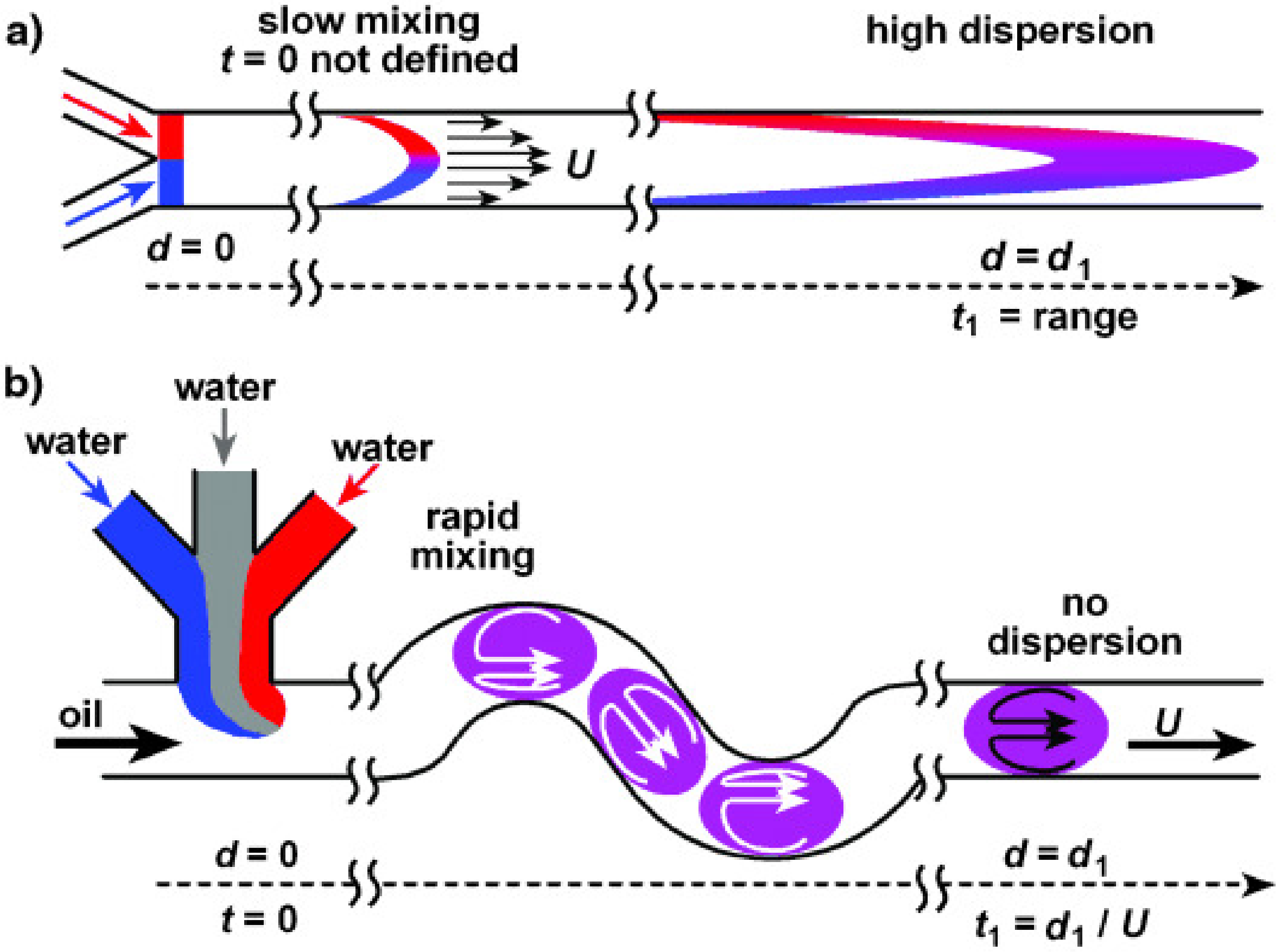
$$R = \frac{128\mu L}{\pi d^4}$$



L = length of the channel (L>>d)
d = diameter of the channel
μ = dynamic viscosity

Modified from https://www.fluigent.com/de/resources-support/expertise-de/expertise-reviews-de/concepts-and-physics-of-microfluidics-de/microfluidic-resistance/

How do things change in multiphase systems?



Song, H., Tice, J. D., & Ismagilov, R. F. (2003). A Microfluidic System for Controlling Reaction Networks in Time. Angewandte Chemie International Edition, 42(7), 768–772.

Capillary number – stable or unstable droplets?

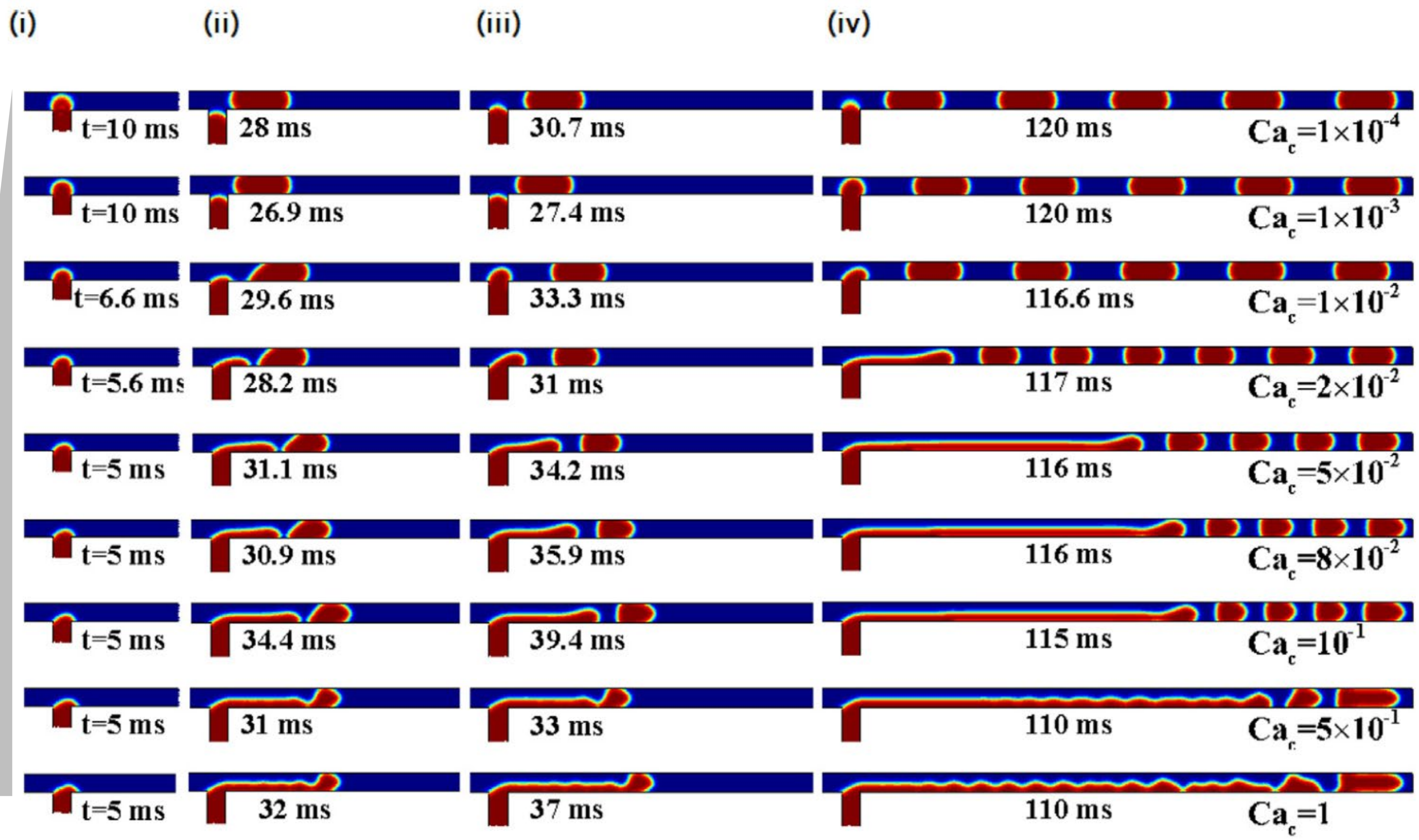
The capillary number Ca describes the ratio between viscous forces and surface tension between two immiscible liquids (e.g. water and oil):

$$Ca = \frac{\mu V}{\gamma}$$

μ = viscosity of the dispersed liquid
V = velocity
γ = surface tension between two fluid phases

In microfluidics the capillary number is usually between 10^{-3} – 10^{-1}

Capillary number – stable or unstable droplets?



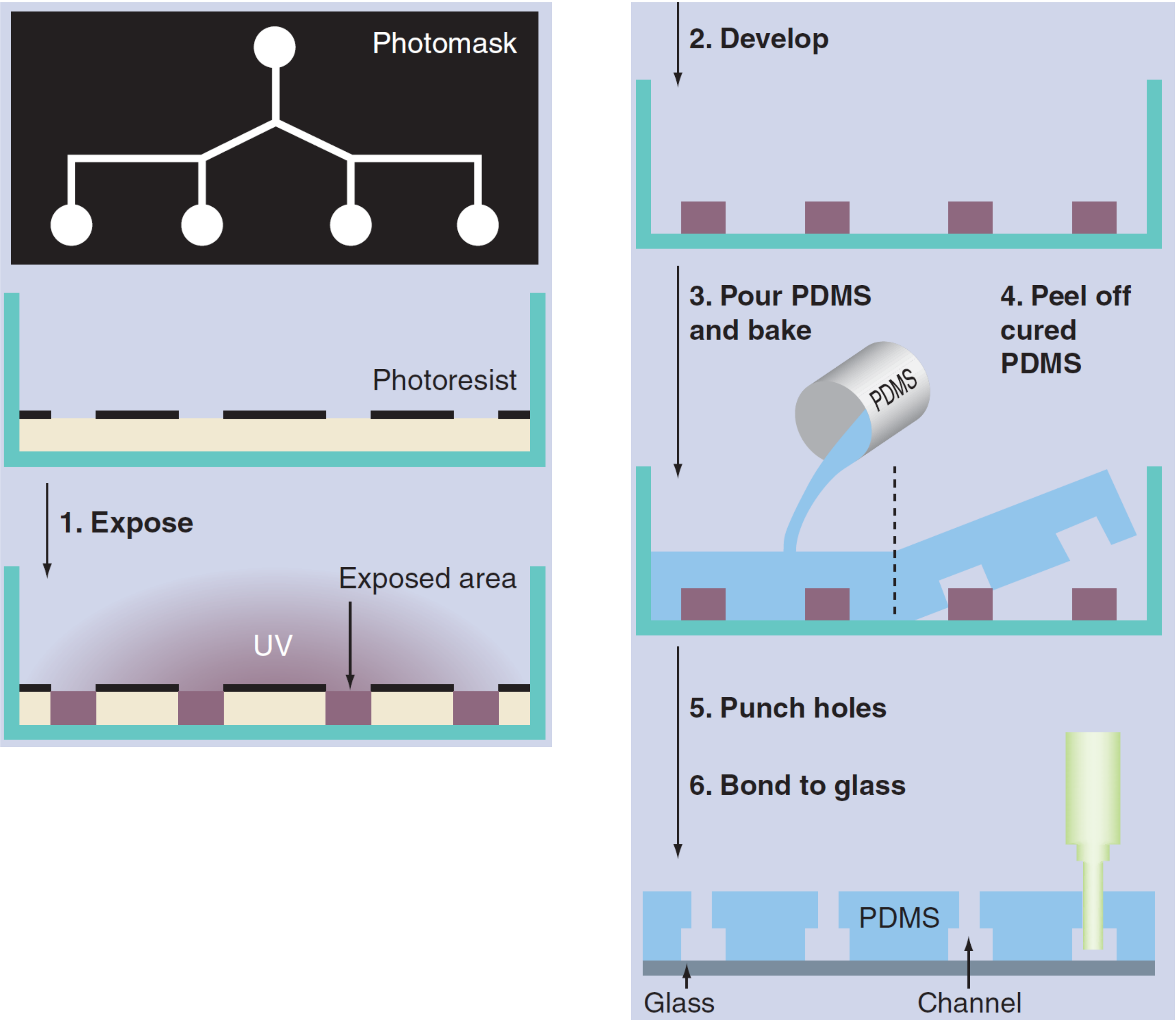
surface tension

viscosity,
flow rate

ratio of aqueous to oil flow = 1

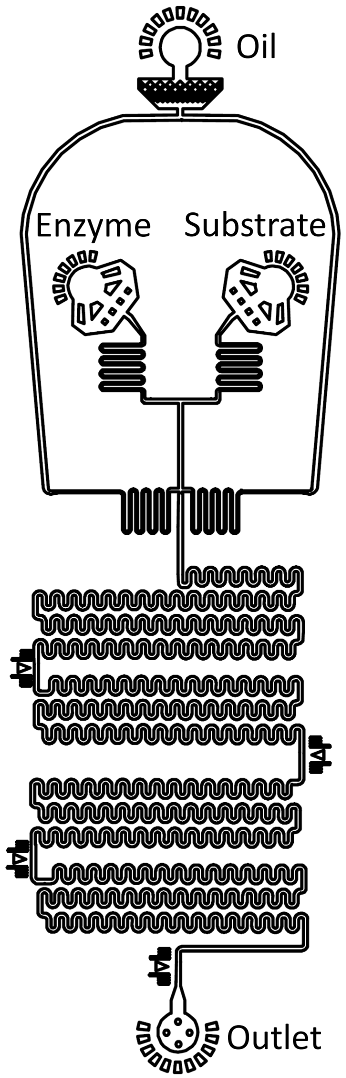
Modified from A. Venkateshwarlu and R.P. Bharti / Journal of the Taiwan Institute of Chemical Engineers 129 (2021) 6479

How to manufacture microfluidic chips?

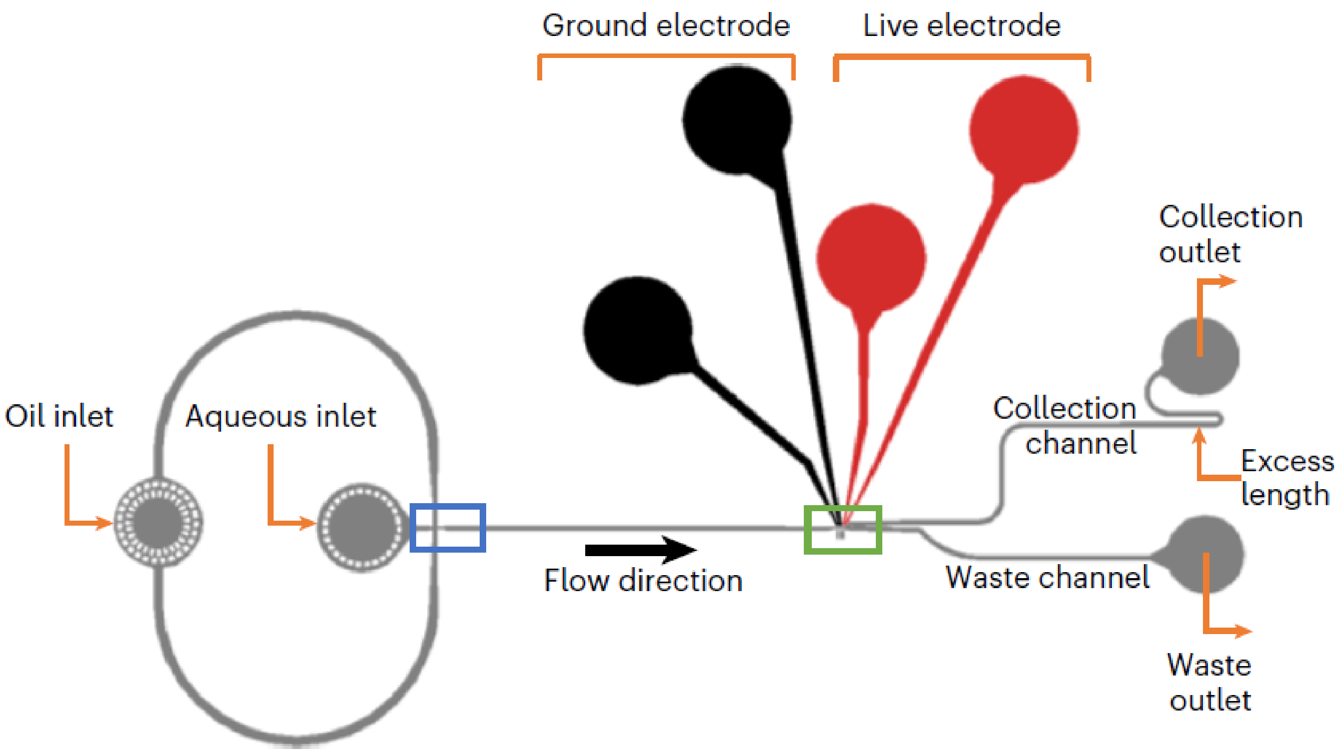


BIOENG-421 modules

Determination of enzyme concentrations (02.12.2025)



LBMM sorting demo (09.12.2025)

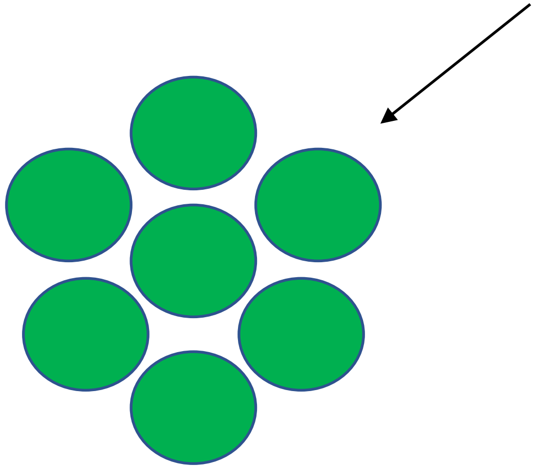


Some practical benefits of microfluidic systems

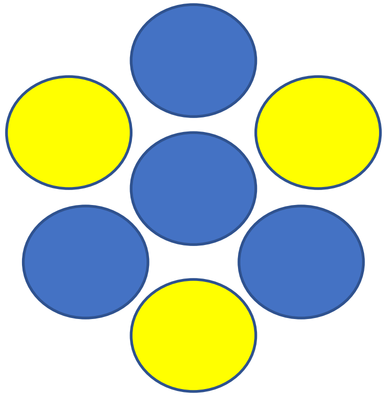
- Large surface to volume ratio = **very rapid heat exchange** (on-chip PCR, etc.)
- Very small volumes enabling to obtain **detectable concentrations of analytes from single cells** (single cell sequencing, phenotypic antibody screens, directed evolution, etc.)
- Low volumes reduce reagent cost and facilitate the **use of very limited material such as patient cells**
- Droplets as assay compartments (“miniaturized test tubes”) can be generated at kHz frequencies, **enabling uHTS**

Single cell RNAseq

This genotype does NOT exist!

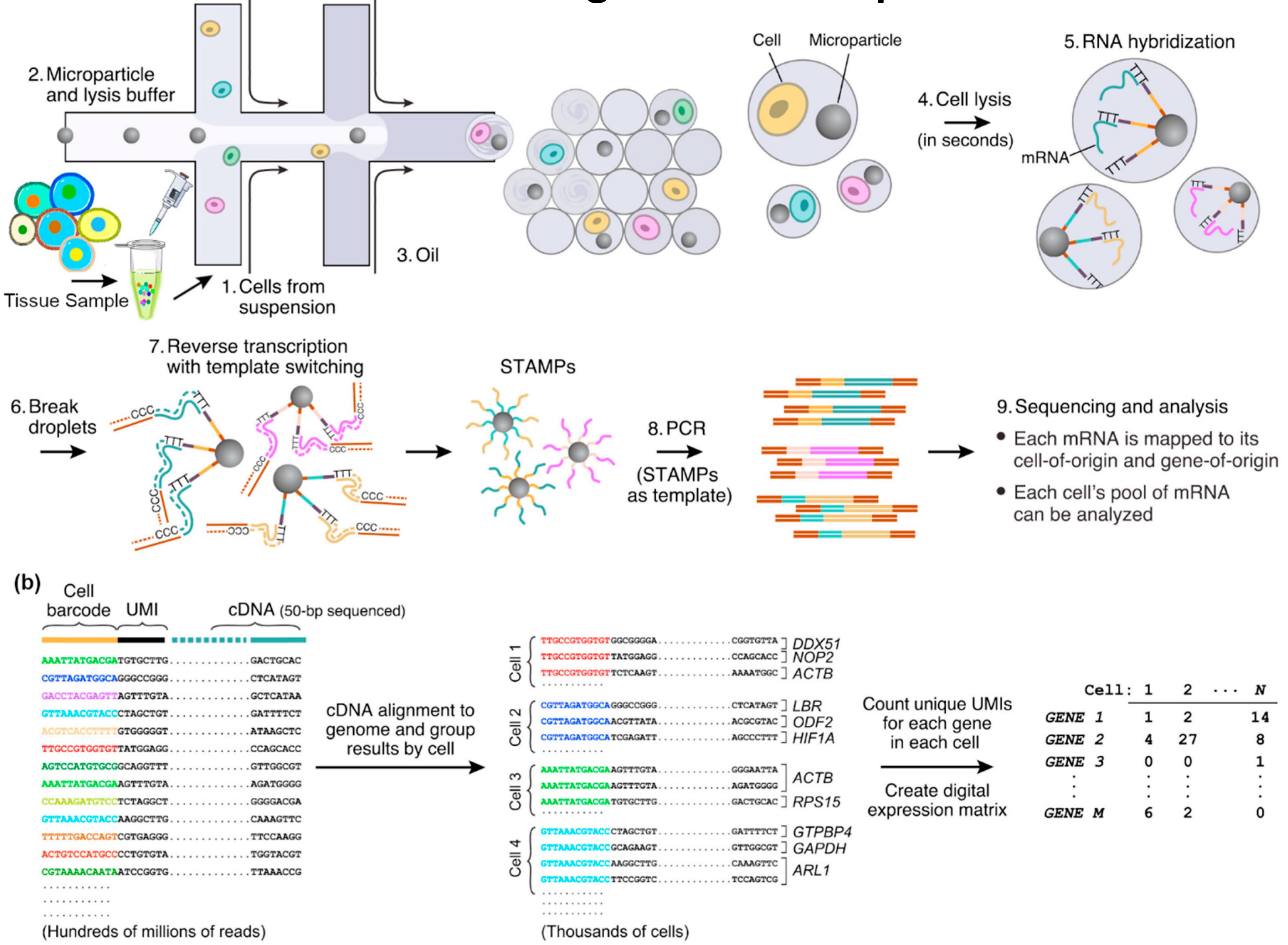


Averaged bulk assays



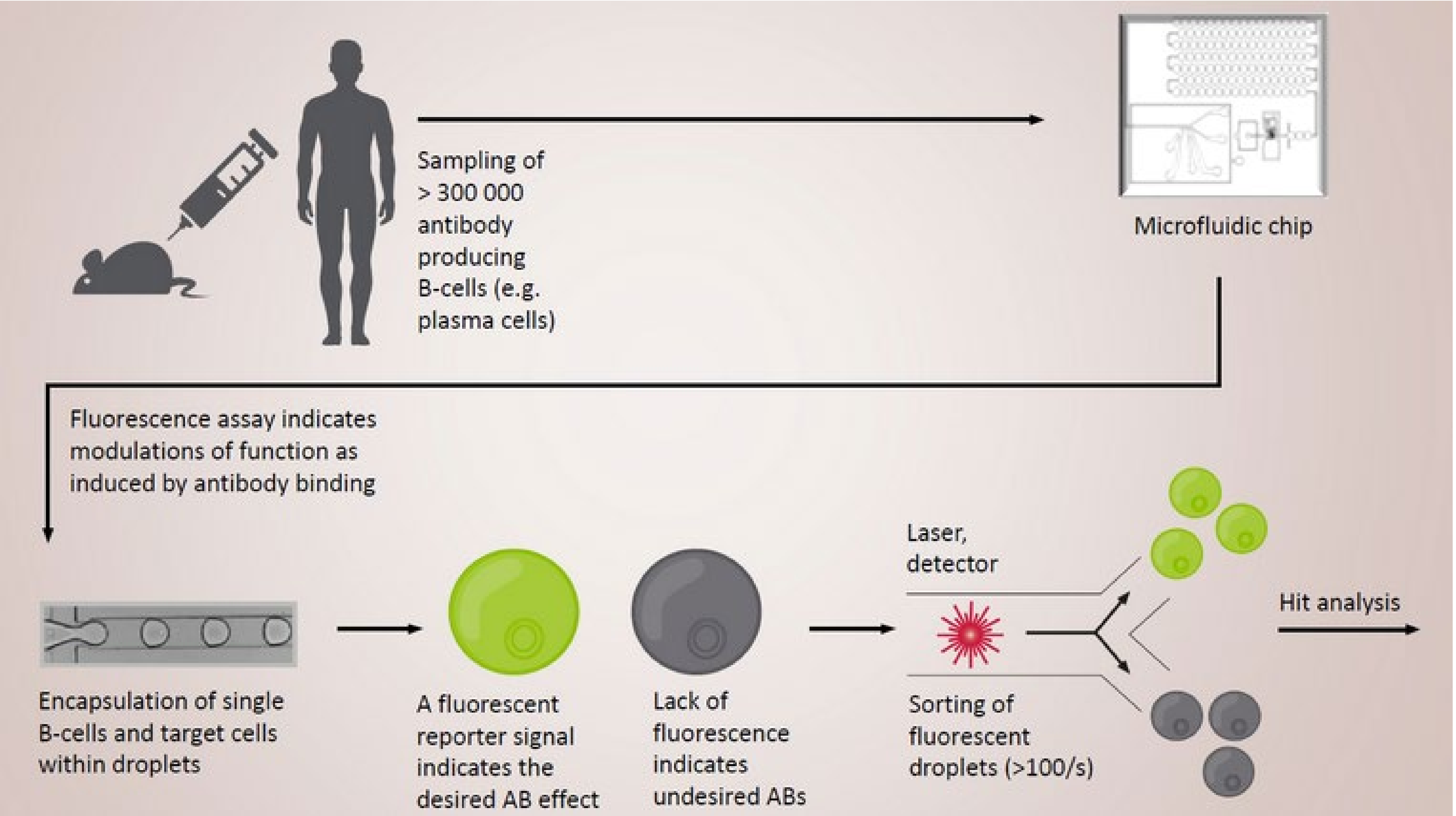
Single cell resolution

Single cell RNAseq

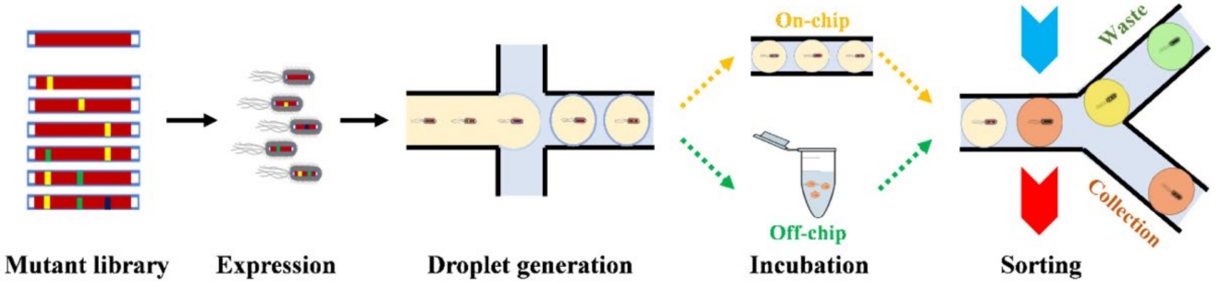


Roozbeh Abedini-Nassab et al., *Biosensors* **2024**, *14*(4), 189; https://doi.org/10.3390/bios14040189

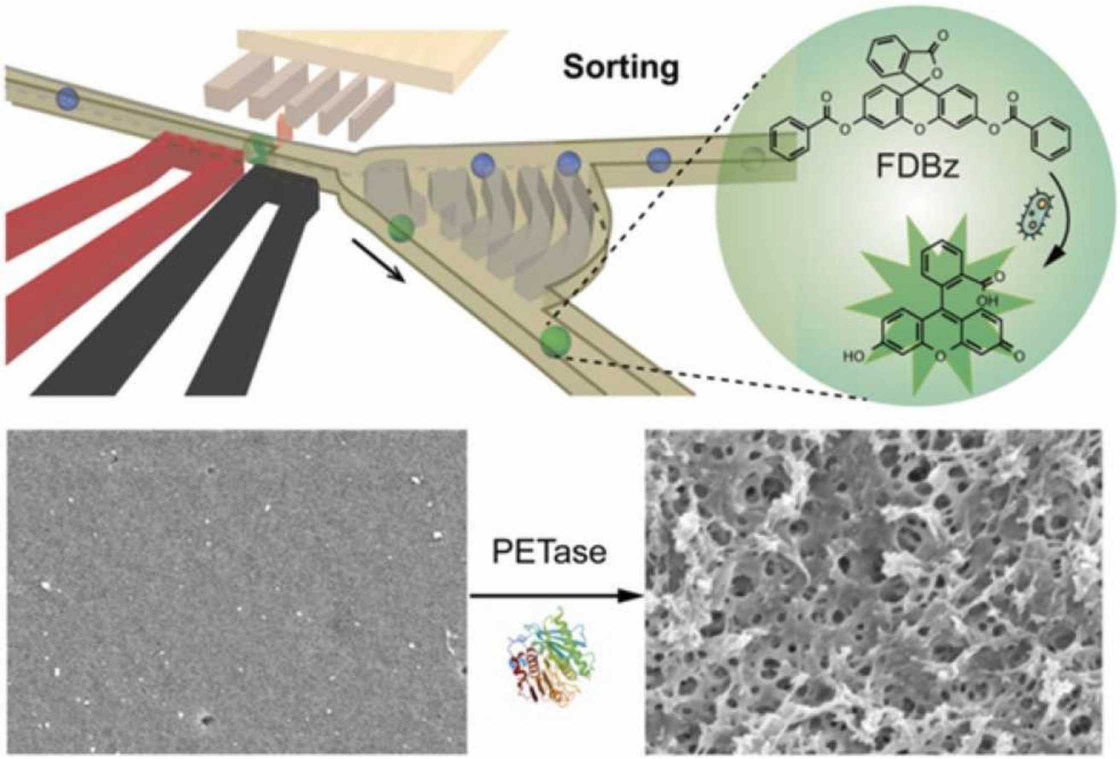
Single cell antibody screening



Directed evolution – new or improved enzyme activities

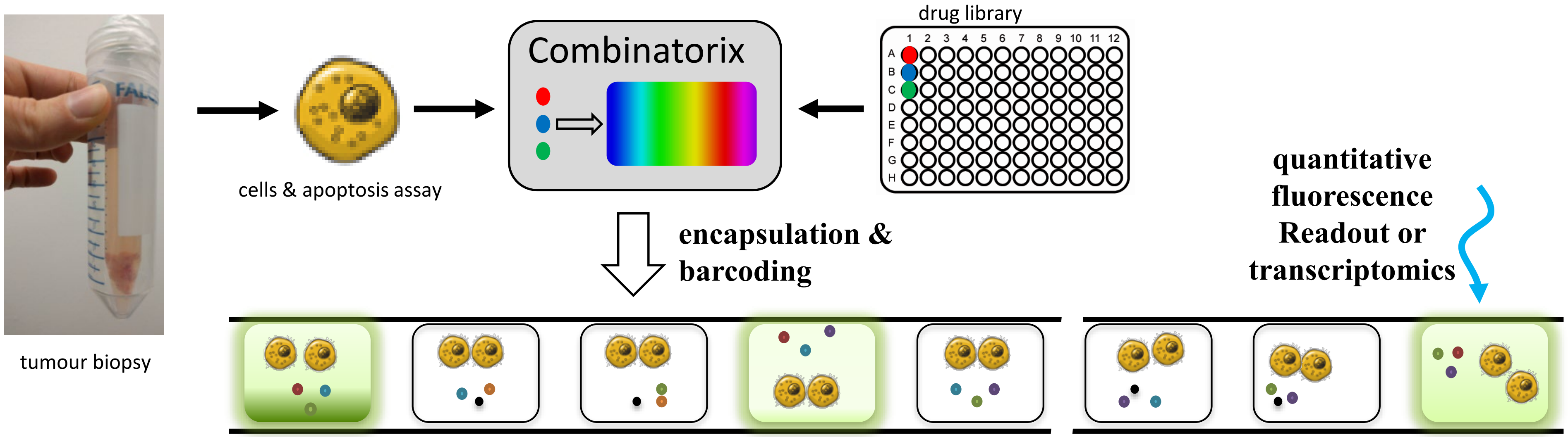


Fu et al., Front. Chem. 9:666867. doi: 10.3389/fchem.2021.666867



Qiao et al., 2022, https://doi.org/10.1016/j.jhazmat.2021.127417

Screening patient samples

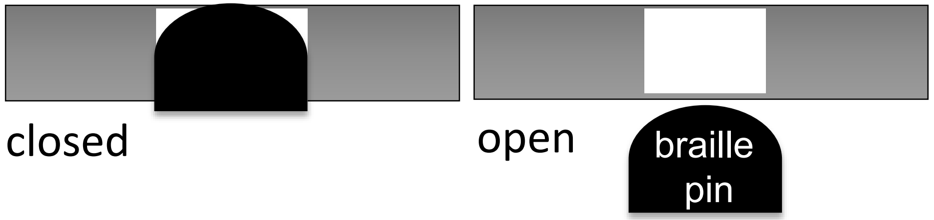


Based on the miniaturized assay volumes, **one can screen about 100 times more treatment conditions on very limited patient material** (e.g. cells from a biopsy) as compared to conventional formats

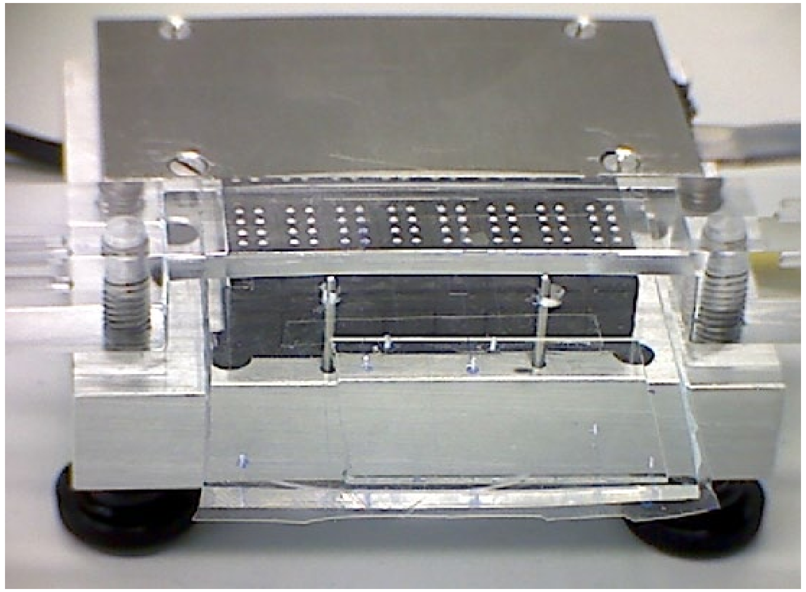
Underlying technology

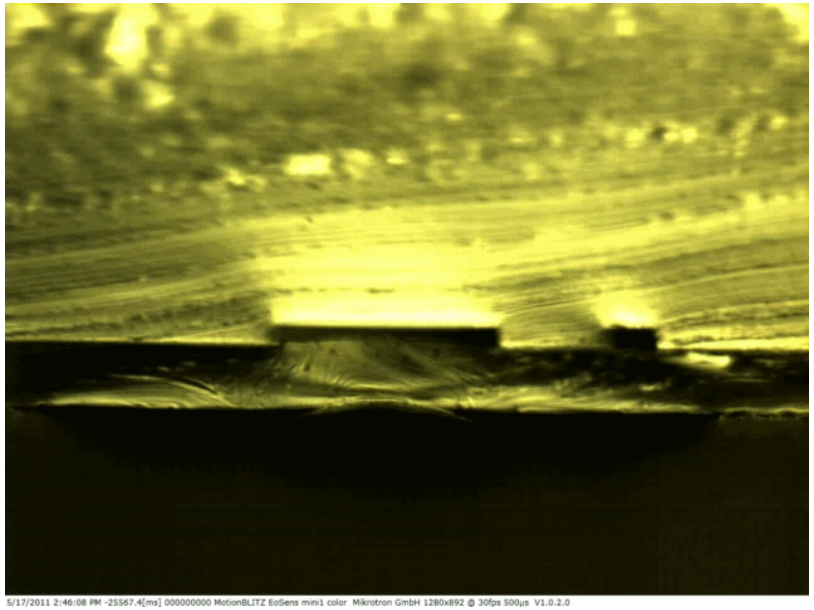
Picture by Hendrik Leber

Underlying technology



Ramesh
Utharala

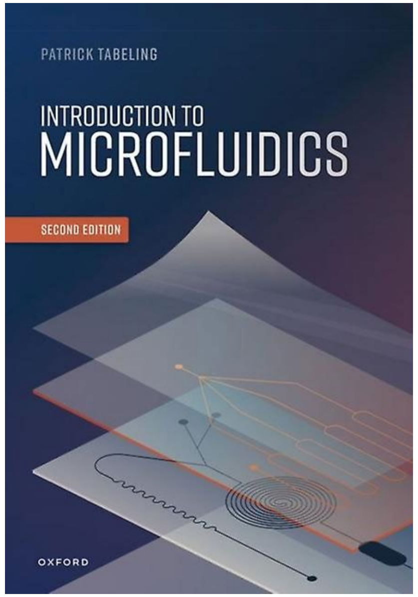


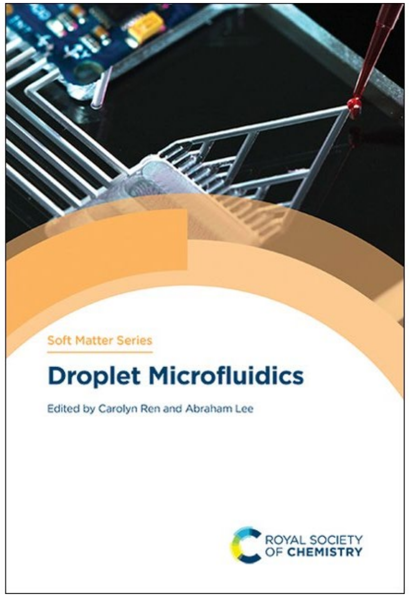


Initially conceived by Gu et al., https://www.pnas.org/doi/10.1073/pnas.0404353101

Microfluidics is a big playground at the interface of engineering and biology! 😉

Further reading





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Droplet-based microfluidics

Thomas Moragues, Diana Arguijo, Thomas Beneyton, Cyrus Modavi, Karolis Simutis, Adam R. Abate, Jean-Christophe Baret, Andrew J. deMello, Douglas Densmore & Andrew D. Griffiths

Nature Reviews Methods Primers **3**, Article number: 32 (2023) | Cite this article

Questions?