MICROFLUIDICS

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1. Fundamentals of microfluidics
2. Topics in microfluidics
Microfluidics is the area of science and technology that is focused on simple or complex, mono- or multiphasic flows that are circulating in natural or artificial micro systems with at least, one dimension of below 500 μm (sometimes below 1000 μm)
Microfluidic systems in nature

• A tree bringing water and nutrients to the leaves via a complex network of capillaries

A capillary network of hundreds of thousands of microchannels with diameters between 100 μm (in the trunk) and 10’s of nm (in the leaf).

The hydrodynamics of the system: the ability of the capillaries to deform under the effect of pressure), the significance of capillary effects and redundancy (if one capillary dies, another takes its place).

A spider web. The spider produces a long, exceptionally strong silken thread a few dozen μm in diameter. The silken thread is a protein that is synthesized in a gland

One of the silk glands of a *nephila clavipes*
### Blood circulation

<table>
<thead>
<tr>
<th>Vessel</th>
<th>$\bar{V}$ (m/s)</th>
<th>$d$ (mm)</th>
<th>$\dot{\gamma}$ (s$^{-1}$)</th>
<th>$Re$ (–)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aorta</td>
<td>0.4</td>
<td>25</td>
<td>130</td>
<td>2500</td>
</tr>
<tr>
<td>Arteries</td>
<td>0.45</td>
<td>4</td>
<td>900</td>
<td>450</td>
</tr>
<tr>
<td>Arterioles</td>
<td>0.05</td>
<td>0.05</td>
<td>8,000</td>
<td>0.5</td>
</tr>
<tr>
<td>Capillaries</td>
<td>0.001</td>
<td>0.008</td>
<td>1,000</td>
<td>0.002</td>
</tr>
<tr>
<td>Venules</td>
<td>0.002</td>
<td>0.02</td>
<td>800</td>
<td>0.01</td>
</tr>
<tr>
<td>Veins</td>
<td>0.1</td>
<td>5</td>
<td>160</td>
<td>125</td>
</tr>
<tr>
<td>Vena cava</td>
<td>0.38</td>
<td>30</td>
<td>100</td>
<td>2800</td>
</tr>
</tbody>
</table>

Man-made systems. Why Micro?

1. Unique physical and chemical effects, mass and heat transfer characteristics
2. Small volumes of expensive and/or dangerous reagents
3. Parallel operation
4. Portability, integration (reactions, separation, detection)
5. Implanting microfluidic devices in biological systems
6. Compatibility with other micro/nanoscale devices
Why Flow? Some of important features

- **Transform time to space.** Instead of absolving subsequent steps in one volume as a function of time, pass through distinct (fixed) sections of a microchannel at different times.

- Ability to integrate different subsequent steps

- Dynamically change conditions for each step

- Induce mechanically stimulated events (shear)

- Ability to transport (small/large molecules, nano and microparticles)

- Ability to conduct separations
Focus of Microfluidics

- Phenomena
- Components
- Systems
- Applications

  Chemical Analysis
  Biological Sensing
  Chemical sensing
  Drug Delivery
  Molecular Separation
  Amplification, sequencing, synthesis of nucleic acids
  Cell biology
Lab-on-a-Chip Systems

Applications

- Chemical, biological and environmental analysis
- Medical diagnosis
- Therapeutic devices

Advantages

- Small volumes of expensive reagents,
- Parallel operation,
- Shorter processing,
- Integration of flow, reactions, separation, and detection
- Integration with information management

Chow, AW, AIChE J 2002, 48, 1593

Burns, MA, Science 2002, 296
MEMS and microfluidics

Miniaturization → micrometer-size mechanical, fluidic, electromechanical, or thermal systems.

1980s

Wheel

Complex objects

1990s: birth of microfluidics

Dimensions: generally, below 500 μm; sometimes 1000 μm

This image was provided by the Karlsruhe group (Germany)
Microfluidics and high throughput screening/separation

Take a sample containing many different objects

A sensor with high throughput is a sensor that determines a substantial part of the elements present in your sample

Microfluidic screening
A few words about nanofluidics
A Surface Forces Balance Technique

Van der Waals forces between surfaces in the vacuum extends over nanometers

\[ F/R = -A/kD^2 \]
\[ A = 2.2 \times 10^{-20} \text{ J} \]

Forces in the presence of electrolyte (Debye layers)

Debye layers may have sizes comparable to Submicrometric channels.

DEBYE-HUCKEL layers - typically 100 nm up to 1 \( \mu \text{m} \) thick in pure water
(Expected) novel phenomena in nanochannels

- The presence of a slip over solid surfaces induces unusual flows.
- The dynamics of interfaces in nanochannels is not understood.
- Hydrodynamic instabilities have specific structures.
- Ordinary liquids may perhaps adopt unusual states.

Experimental studies of flow in nanofluidic devices are just beginning.

Difficulties in the fabrication of nanodevices and in the measurement techniques tend to slow down the investigation of the domain.
How to make liquid move through microfluidic channels?

*Pressure Driven Flow*

The fluid is pumped through the device *via* positive displacement pumps (syringe pumps) or using pressure gauges. One of the basic laws is the so-called *no-slip boundary condition*: the fluid velocity at the walls must be zero. This produces a parabolic velocity profile within the channel.

Velocity profile in a microchannel with aspect ratio 2:5 for pressure driven flow (calculation using Coventorware software.)

http://faculty.washington.edu/yagerp/microfluidictutorial/basicconcepts/basicconcepts.htm
Hydrodynamic resistance

\[ \Delta P = R_h Q \]

\( Q \) is the volumetric flow rate of the liquid, \( \Delta P \) is pressure drop, \( R_h \) is the hydrodynamic resistance (analogous to the electrokinetic law \( U=IR \))

Channel with a *circular* cross-section (total length \( L \), radius \( R \)): 

\[ R_h = \frac{8\mu L}{\pi R^4} \]

Channel with a *rectangular* cross-section (width \( w \) and height \( h \), \( h<w \))

\[ R_h \approx \frac{12\mu L}{wh^3(1-0.630h/w)} \]

\( R_h \) increases as the system size decreases

In a network of channels \( R_h \) can be computed as in electrokinetics:

- two channels *in series* have a resistance \( R_h = R_{h1} + R_{h2} \)
- two channels *in parallel* have a resistance \( \frac{1}{R_h} = \frac{1}{R_{h1}} + \frac{1}{R_{h2}} \).
**Electrokinetic Flow**

If the walls of a microchannel have a charge, an electric double layer of counter ions will form at the walls. When an electric field is applied across the length of the channel, the ions in the double layer move towards the electrode of opposite polarity. This creates motion of the fluid near the walls and transfers *via* viscous forces into convective motion of the bulk fluid.

- For the channel open at the electrodes, the velocity profile is uniform across the entire width of the channel.

- For a closed channel, a recirculation pattern forms, in which a fluid along the center of the channel moves in a direction opposite to that at the walls.
Fluid mechanics of microfluidics

Navier-Stokes equation is the central relationship of fluid dynamics

Basic assumptions
  continuous media
  continuum mechanics

For liquids:
  Assumptions made in macrofluidics, work for microfluidics
  (down to 10-100 nm)
How to describe the motion of a fluid?

- Velocity depends on space and time \( v = v(x(t), t) \)
- Consider Newton's law for a infinitesimal volume \( V \):

\[
\frac{F}{m} = f_a = m \cdot a = \rho \cdot \frac{dv}{dt}
\]

\( f_a \) and \( v \) are vectors!!
How to describe the motion of a fluid?

• Momentum equation (acceleration) in x-direction:

\[ f_{a,x} = \rho \cdot \frac{dv_x}{dt} = \rho \cdot \frac{d}{dt} v_x(x(t), y(t), z(t), t) = \]
\[ = \rho \cdot ( \frac{\partial v_x}{\partial x} \bigg|_{y,z,t} \cdot \frac{dx}{dt} + \frac{\partial v_x}{\partial y} \bigg|_{x,z,t} \cdot \frac{dy}{dt} + \frac{\partial v_x}{\partial z} \bigg|_{x,y,t} \cdot \frac{dz}{dt} + \frac{\partial v_x}{\partial t} \bigg|_{x,y,z} ) = \]
\[ = \rho \cdot ( \frac{\partial v_x}{\partial x} \bigg|_{y,z,t} \cdot v_x + \frac{\partial v_x}{\partial y} \bigg|_{x,z,t} \cdot v_y + \frac{\partial v_x}{\partial z} \bigg|_{x,y,t} \cdot v_z + \frac{\partial v_x}{\partial t} \bigg|_x ) \]

• Momentum equation in 3D (vector notation):

\[ \mathbf{f}_a = \rho \cdot \frac{d\mathbf{v}}{dt} = \rho \cdot ( \mathbf{v} \cdot \nabla ) \mathbf{v} + \frac{\partial \mathbf{v}}{\partial t} \]
\[ \nabla = \left( \frac{\partial}{\partial x}, \frac{\partial}{\partial y}, \frac{\partial}{\partial z} \right) = \bar{e}_1 \frac{\partial}{\partial x} + \bar{e}_2 \frac{\partial}{\partial y} + \bar{e}_3 \frac{\partial}{\partial z} \]

Nabla operator
Acceleration over time

\[ \frac{v(x, t_2)}{t = t_2} - \frac{v(x, t_1)}{t = t_1} \]

\[ f_a = \rho \cdot \frac{\partial v}{\partial t} \]
Acceleration along a streamline

- Increase of the fluid velocity due to mass conservation
- Fluid has to be accelerated along the streamline

\[ \mathbf{f}_a = \rho \cdot \mathbf{v} \cdot \nabla \mathbf{v} \]

\[ v(x(t),t) \]
The Navier-Stokes equation

• … for incompressible Newtonian fluids

\[ \mathbf{f}_a = \rho \left[ \frac{\partial}{\partial t} \mathbf{v} + (\mathbf{v} \cdot \nabla) \mathbf{v} \right] = \mathbf{f}_{\text{pressure}} + \mathbf{f}_{\text{friction}} + \mathbf{f}_{\text{volume}} \]

• left hand side
  – Change in momentum (Newton)
    • due to change of velocity over time at a given location
    • due to acceleration of fluid e.g. when moving into smaller flow channel cross-sections (also in stationary cases)

• right hand side
  – Forces acting on fluid
    • pressure gradient
    • friction forces
    • volume forces
Pressure gradient

\[ \mathbf{f}_a = \mathbf{f}_{\text{pressure}} + \mathbf{f}_{\text{friction}} + \mathbf{f}_{\text{volume}} \]

\[ \mathbf{f}_{\text{pressure}} = \frac{dF_{\text{pressure}}}{dV} = -\nabla p \]
Body force (= volume force)

\[ f_a = f_{\text{pressure}} + f_{\text{friction}} + f_{\text{volume}} \]

- Actuation of fluid by the body force:
  force \( f_{\text{volume}} \) acts in the volume itself

**Body forces**: centrifugal forces, gravity forces, electrostatic forces
Example 1: static pressure under gravity

– Only gravity is considered in NS-equation

\[ \mathbf{f}_{\text{volume}} = \mathbf{f}_g = \rho \mathbf{g} \]

– Stationary flow (v=const.):
  
  • friction is zero (no motion)
  • acceleration is zero (dv/dt=0)

\[ \mathbf{f}_a = \mathbf{f}_{\text{pressure}} + \mathbf{f}_{\text{friction}} + \mathbf{f}_{\text{volume}} \quad \Rightarrow \quad -\mathbf{f}_{\text{pressure}} = \mathbf{f}_{\text{volume}} \]

– Result:

\[ \nabla p = \rho_\infty g \]

\[ \frac{dp}{dy} = \rho_\infty g \quad \Rightarrow \quad p_{\text{max}} = \rho_\infty gh + p_0 \]
• **Example 1:** for water in a microchannel:

\[\rho = 1000 \text{ kg/m}^3\]
\[g = 9.81 \text{ m/s}^2\]
\[h = 100 \text{ \mu m}\]

\[p_{\text{max}} = 0.981 \text{ Pa} = 9.81 \cdot 10^{-6} \text{ bar}\]

Gravitational effects are negligible in microfluidics.
Friction

- Friction affects the motion (velocity) of the fluid

\[ f_{\text{friction}, z} = \frac{dF_\eta}{dV} = \eta \frac{\partial^2 v_z}{\partial x^2} \]

\[ f_{\text{friction}} = \eta \nabla^2 v \]

The motion is damped by the friction force
Reynolds number \((Re)\)

- Approximate friction energy
  \[ E_{\text{friction}} \propto \|F_{\text{friction}}\| \cdot l = \eta \frac{V}{l} A \cdot l = \eta \frac{V}{l} V \]

- Approximate kinetic energy
  \[ E_{\text{kin}} \propto m v^2 \]

\[
\frac{E_{\text{kin}}}{E_{\text{friction}}} = \frac{mv^2 l}{\eta v V} = \frac{\rho lv}{\eta} = \boxed{Re} \quad \boxed{Re = \frac{\rho lv}{\eta}}
\]

Reynolds number is the ratio of work spent on acceleration to energy dissipated by friction (A more general definition: \(Re\) a dimensionless number that gives the ratio of inertial forces (characterizing how much a particular fluid resists to motion) to viscous forces.

- the Re-number is the most important dimensionless number in microfluidics
- low \(Re\)-numbers, i.e. **viscous forces dominate, are typical for microfluidics**
- \(l\) is a characteristic length scale
Simplifications in microfluidics

\[ \rho_{\infty} \left[ \frac{\partial}{\partial t} \mathbf{v} + \left( \mathbf{v} \cdot \nabla \right) \mathbf{v} \right] = -\nabla p + \eta \nabla^2 \mathbf{v} + \mathbf{f}_{\text{volume, g}} \]

- Gravity is neglected
- Influence of convection is small, \( \rho (v \cdot \nabla)v \to 0 \), i.e. we assume that there is no convective momentum transport
- If additionally a stationary flow is considered ...

• Poisson equation (driving pressure and friction are balanced in a stationary laminar flow):

\[ \rho_{\infty} \left[ \frac{\partial}{\partial t} \mathbf{v} + \left( \mathbf{v} \cdot \nabla \right) \mathbf{v} \right] = -\nabla p + \eta \nabla^2 \mathbf{v} + \rho_{\infty} g \]

\[ \nabla p = \eta \nabla^2 \mathbf{v} \implies \mathbf{f}_{\text{pressure}} = -\mathbf{f}_{\text{friction}} \]
Flow Regimes

- **Re < 1 (Stokes regime)**
  - No lateral convection
  - Adjacent layers (lamellae) do not “interfere” (lamellae do not mix)
  - Inertial terms are neglected

- **1 < Re < Re* (Intermediate)**
  - Lateral convection becomes increasingly important

- **Re > Re* (Turbulent)**
  - Perturbations are amplified
  - Curling of field lines
  - “Unpredictable” development of field of velocity vectors over time

\[ Re* \approx 2300 \]
Critical Reynolds number

- Critical $Re^*$ corresponds to a critical velocity $v^*$

$$v^* = Re^* \frac{\eta}{\rho l}$$

- Typically $Re^*$ is in the range of 2300

- For a microdevice $v^*$ is hardly reached
  ($l = 100 \ \mu m \rightarrow v^* \approx 25 \ \text{m/s}$)

- As $Re$ increases further, the turbulent character of flow increases
Examples of laminar flow

Laminar flow means that diffusion is the only mechanism to achieve mixing between parallel fluid streams. This is a slow process.
Other dimensionless numbers

Capillary phenomena are extremely important in microsystems

Laplace law

\[ \delta E = -p \delta V + \gamma \delta S \]

\[ V = \frac{4}{3} \pi R^3 \Rightarrow \delta V = 4 \pi R^3 \delta R \]

\[ S = 4 \pi R^2 \Rightarrow \delta S = 8 \pi R \delta R \]

At mechanical equilibrium: \( \delta E = 0 \)

\[ p = \frac{2 \gamma}{R} \]
Pressure drops caused by capillarity are $\sim l^{-1}$ while those due to viscosity scale as $l^0$

The capillary number, $Ca$, represents the relative effect of **viscous forces** versus **surface tension** acting across an interface between a liquid and a gas, or between two immiscible liquids

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

$\mu$ is the viscosity of the liquid, $U$ is a characteristic velocity and $\gamma$ is the surface or interfacial tension between the two fluid phases

*Microfluidics: $Ca \approx 10^{-1} - 10^{-3}$*
Summary

– NS-equation is derived by a momentum balance for a continuum element
– For **Newtonian, incompressible** fluids the NS equation is

\[
\rho_\infty \left[ \frac{\partial}{\partial t} \mathbf{v} + (\mathbf{v} \cdot \nabla) \mathbf{v} \right] = -\nabla p + \eta \nabla^2 \mathbf{v} + \rho_\infty \mathbf{g}
\]

• Momentum change (right-hand side) can be caused by:
  – Pressure gradients
  – Viscous losses
  – Body forces
• **Re** number characterizes the damping of disturbances \( \rightarrow \) laminar and turbulent flow

\[
Re = \frac{\rho l v}{\eta}
\]
Summary (cont)

- For $Re < Re^*$ the flow is laminar
- For $Re > Re^*$ the flow is turbulent

• In microfluidics we (usually) assume
  - No gravity
  - Incompressibility
  - Dominance of viscous forces

• Capillarity plays an important role in microfluidics, as represented nu small Capillary numbers
How to make a microfluidic device?

- Thermoembossing
- Wet and dry etching
- Moulding
- Laser ablation
- Photolithography
- Soft lithography
- .... other clever methods
EFFECT OF SURFACE ENERGY OF MICROCHANNELS

design ~ 1 hour
print out ~ 1 day
fab master ~ 3 hours ~ 2 days
make copies of the device ~ 2 hours each

Speed matters!
Fabrication of Microfluidic Device: Soft Lithography

1) Spincoat photoresist
2) UV Exposure
3) Develop photoresist
4) Prepare mold
5) Seal to substrate

MIXING IN MICROFLUIDICS
Mixing in microfluidics

- No turbulence in microfluidics. Mixing occurs by diffusion. Little or no mixing.

- A dimensionless number, analogous to the Reynolds number, is **Peclet number**

  \[ Pe = \frac{Ul}{D} \sim \frac{\text{advection}}{\text{diffusion}} \]

*Diffusion time for a 100 μm wide channel (for a molecule such as fluorescein):*

\[ \tau = \frac{l^2}{D} \sim 100 \text{s} \]

This time may be too long, especially if one develops several chemical reactions on the same chip
Mixing by diffusion

Residence time: $t_R = \frac{L}{U} = \frac{Lw}{Q}$
Mixing time: $t_D = \frac{w^2}{D}$

In order to achieve mixing, one must have $t_R >> t_D$; this implies:

$L >> \frac{Qw}{bD}$

In practice, one reaches lengths on the order of centimeters
Clever solutions

The distributive micromixer

Hydrodynamic focusing

From A. Manz (2004)

Mixing in tens of microseconds
Austin et al, PRL (2002)
Chaotic mixer for microchannels

To generate transverse flows in microchannels, ridges were placed on the floor of the channel at an oblique angle, with respect to the long axis (y) of the channel.

Cross-channel mixer


Time periodic transverse flow

Main Flow

400 μm

Perturbation is applied
Line is stretched

Perturbation is stopped
Line is folded
Droplet microfluidics
Generation of droplets

A and B are two immiscible liquids

Narrow size distribution
High frequency of droplet generation
Control of droplet morphology (double emulsions)
Flow-focusing

<table>
<thead>
<tr>
<th>$Q_0$ [mL/sec]</th>
<th>$Q_i / Q_0$</th>
<th>1/4</th>
<th>1/40</th>
<th>1/400</th>
</tr>
</thead>
<tbody>
<tr>
<td>5.6 x 10^{-5}</td>
<td>(a)</td>
<td>(g)</td>
<td>(m)</td>
<td></td>
</tr>
<tr>
<td>1.4 x 10^{-4}</td>
<td>(b)</td>
<td>(h)</td>
<td>(n)</td>
<td></td>
</tr>
<tr>
<td>4.2 x 10^{-4}</td>
<td>(c)</td>
<td>(i)</td>
<td>(o)</td>
<td></td>
</tr>
<tr>
<td>1.4 x 10^{-3}</td>
<td>(d)</td>
<td>(j)</td>
<td>(p)</td>
<td></td>
</tr>
<tr>
<td>4.2 x 10^{-3}</td>
<td>(e)</td>
<td>(k)</td>
<td>(q)</td>
<td></td>
</tr>
<tr>
<td>8.3 x 10^{-3}</td>
<td>(f)</td>
<td>(l)</td>
<td>(r)</td>
<td></td>
</tr>
</tbody>
</table>

Stone, APL, 2002
Flow-focusing

A 100-fold reduced speed

T-junction

100 µm
\[ d = \left[ \frac{4}{\pi} \frac{Q_{\text{drop}}}{v_{x, \text{cont}}} \right]^{1/2} \]

\textit{d} \textit{ is the average diameter of the coaxial jet}

\[ d_0 = (1.5 \lambda_{\text{breakup}} d^2)^{1/3} \]

\textit{d}_0 \textit{ is the average diameter of droplets}

Water

Oil 1

Oil 2

CORES

CORE-SHELL
Multicore Capsules: Hypothesis

Interfacial wavelength

\[ \lambda \approx 9.02d_{jet} \]

\[ \lambda \sim \left( \frac{Q_{disp}}{Q_{cont}} \right)^{1/2} \]

\[ \lambda_m / \lambda_o \cong n \]

\[ n - 1 < \lambda_m / \lambda_o < n \]

\( \lambda_o \) is the breakup wavelength of oil jet; \( \lambda_m \) is the breakup wavelength of monomer jet.

\( n \) is the number of monodisperse oil cores per capsule.

\( n \) is the number of polydisperse oil cores per capsule.
Stable breakup of coaxial jet

$n - 1 < \frac{\lambda_m}{\lambda_o} < n$
Synthesis of Polymer Capsules
Continuous Microfluidic Reactors

Microfluidic Flow-Focusing Device

Poly(tripropylene glycole diacrylate)

Polyacrylates
Polystyrene
Polyurethane
Biopolymer gels

0.5 % < CV < 3 %

Polymer microbeads
Nanoparticle-loaded beads
Liquid Crystal-polymer particles
Porous microbeads
FITC-BSA-conjugated beads

Shape and morphology control
Synthesis of Janus particles

A: MAOP-DMS + dye
B: PETA-3/AA

\[ V \sim \frac{(Q_{m1} + Q_{m2})}{Q_w} \]

\[
\frac{Q_{m1}}{Q_{m2}} = \frac{h_1^2 (3R - h_1)}{h_2^2 (3R - h_2)}
\]
Exploratory droplet microfluidics

- Optimization of chemical reactions
- High-throughput generation of cellular microenvironments

Ismagilov, 2005
Microenvironments for cell co-culture

Direct and indirect cell-cell interactions:
proliferation, self-renewal, death or differentiation

Applications:
• wound healing
• tissue engineering
• inhibition of cancer spreading

\[ Q_{tor} = Q_G + Q_R = \text{const} \]
Change the ratio \( Q_G/Q_R \)
Microenvironments for cell co-culture

Murine embryonic stem (mES) cells labelled with Vybrant Cell Tracer ("green") and CellTracker Orange ("red")

Embryoid bodies (YC5 Mouse Embryonic Stem cells)

Cell co-culture

Scale bar is 100 μm
Microfluidics and single molecule studies

Experiment by S. Chu et al (1994)
Books

P. Tabeling. Introduction to Microfluidics.

Microfluidics for Biotechnology - J.Berthier P.Silberzan

Micro and NanoFlows (2-nd edition of Karnadiakis’s book)

Reviews:


- Analytical Chemistry, Lab on a Chip
