



Effect of wall shear rate on wall flux of bacteria

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Industrial
partners



COST FP1005 "Fibre Suspension Flow Modelling"- ERCOFTAC SIG43 "Fibre suspension flows".
Coimbra – March 2013

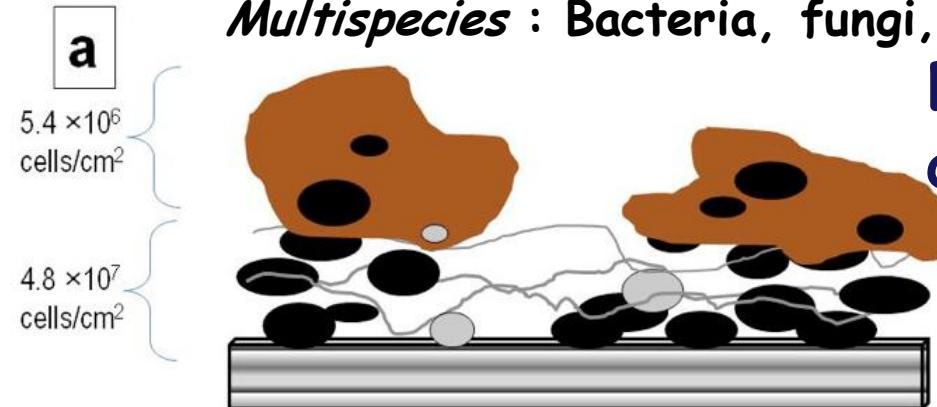
Introduction

- Bacteria in drinking water : migration to the wall -> biofim ?
- Formation mechanisms of biofilm ? How to investigate?
- Test sections
- Transport equations
- Results
- Bacteria adhesion at the wall (cohesion of biofilm)
- Cleaning the surface colonized by biofilm
- Conclusions

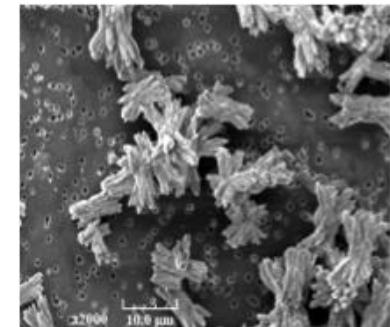
Bacteria and deposits = Biofilm

Drinking water bacteria and deposits

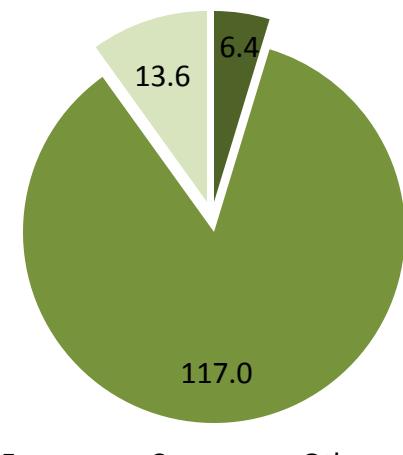
Multispecies : Bacteria, fungi, ...



Carbonates



Metals ($\mu\text{g}/\text{cm}^2$)



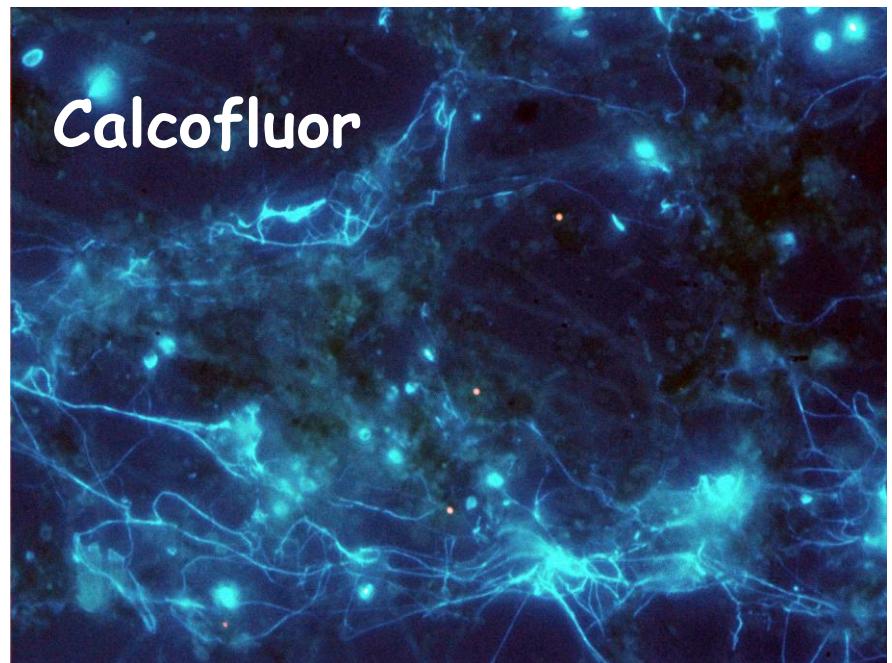
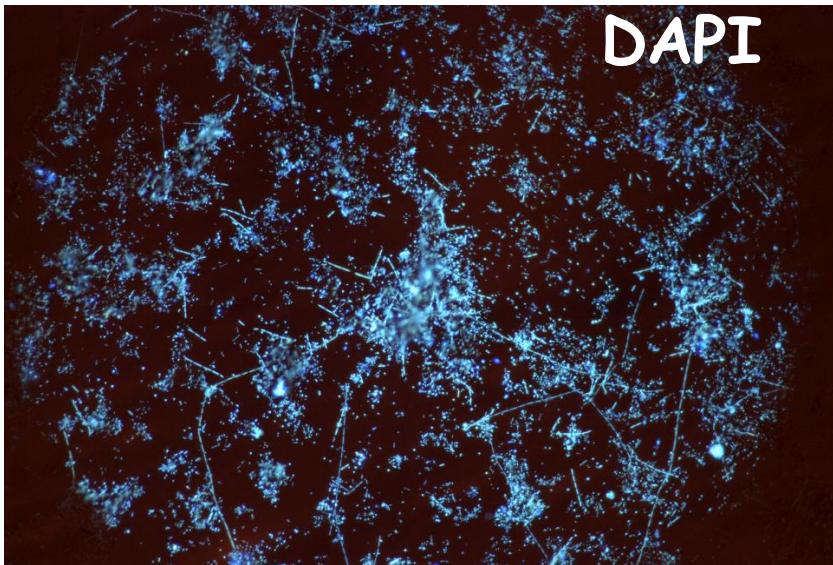
Brown deposits

Bacteria

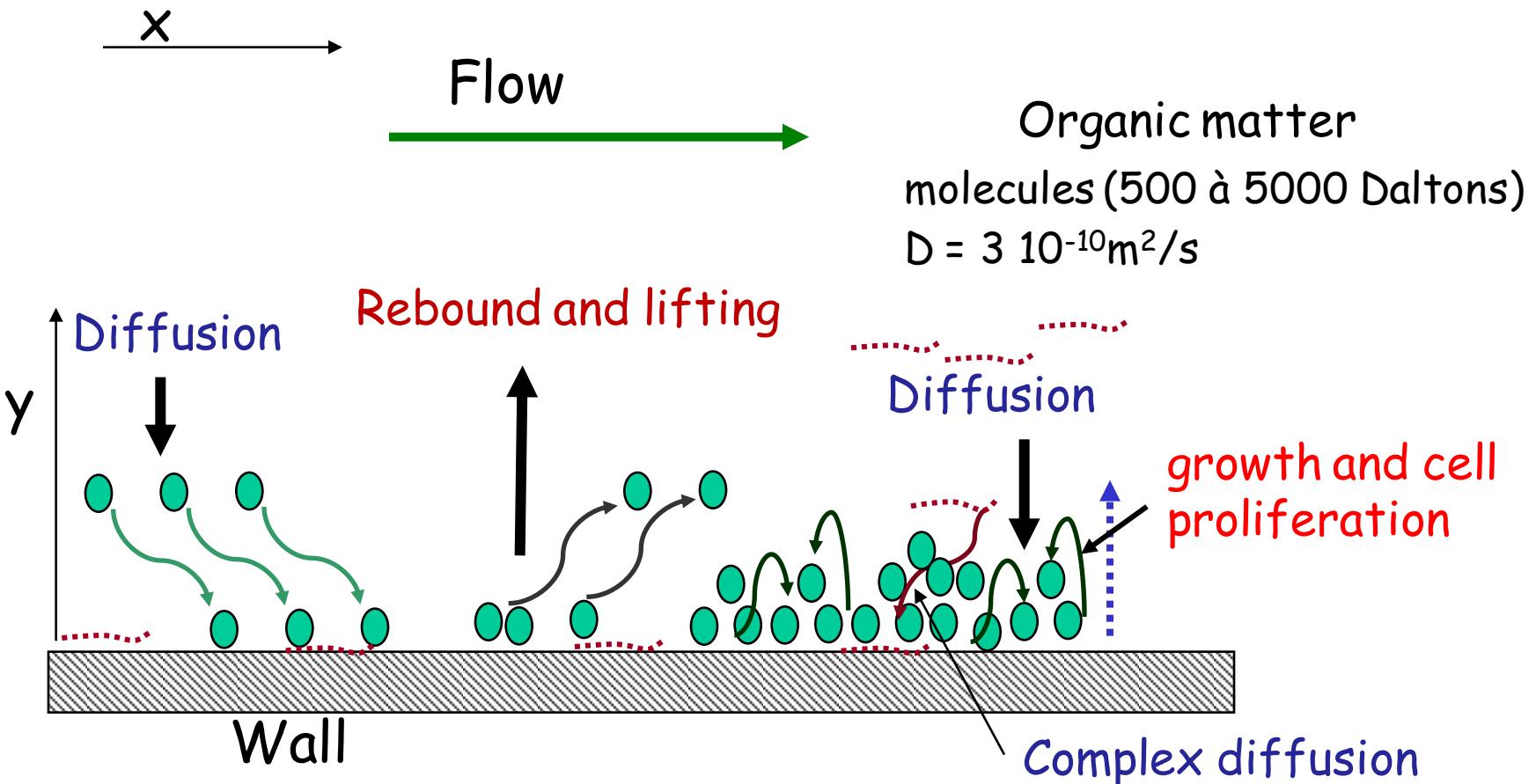
Organic matter :

20 to 100 µg TOC/cm²

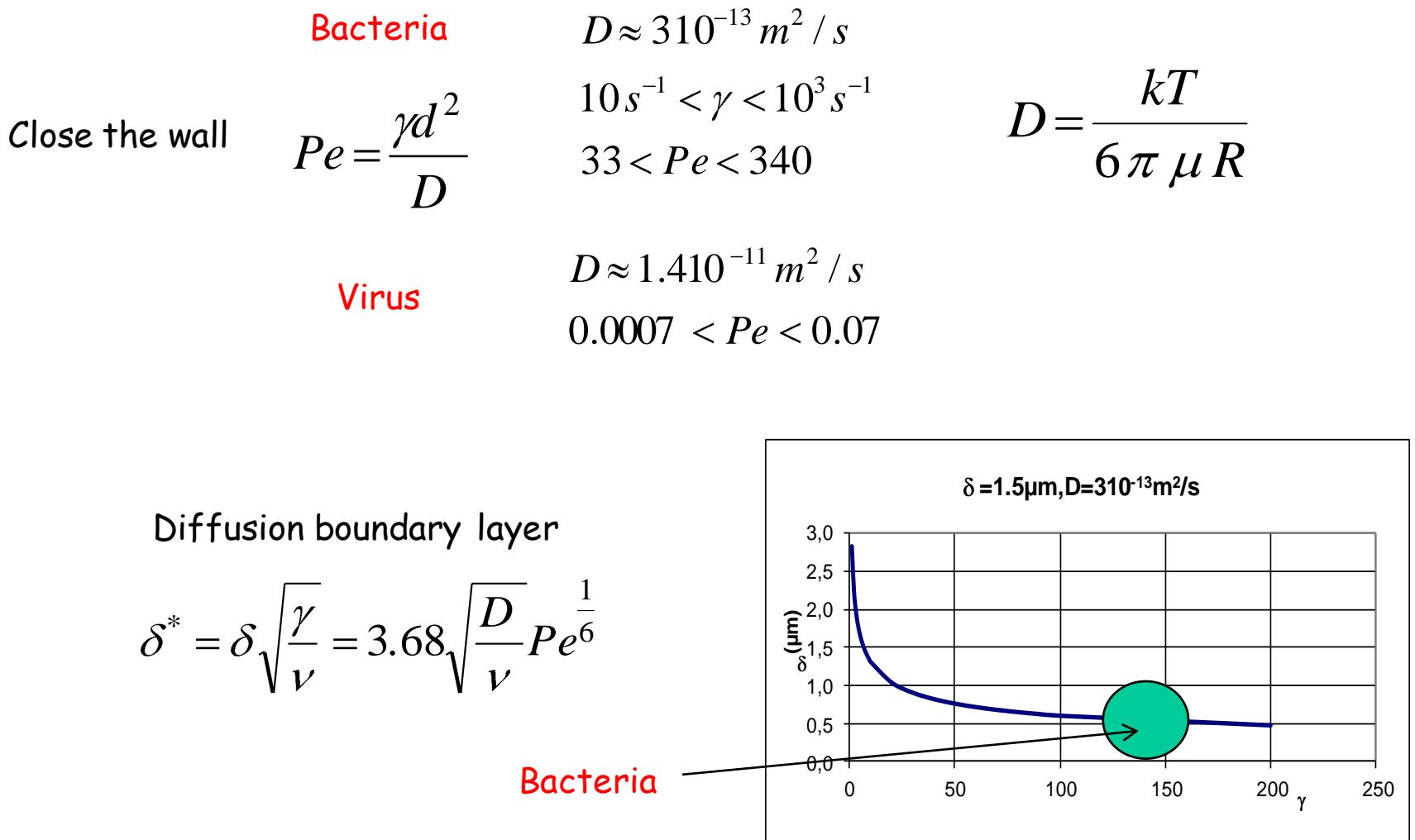
Drinking water biofilms (4 months old)



Wall activities



Phenomenological variables - mass transfer



Transport equation

$$\vec{J} = C \vec{V} - D \vec{\nabla} C$$

$$\frac{\partial C}{\partial t} + \operatorname{div} \vec{J} = 0 \Rightarrow \frac{\partial C}{\partial t} + \vec{V} \cdot \vec{\nabla} C = D \Delta C$$

$$\frac{\partial C}{\partial t} + \vec{V} \cdot \vec{\nabla} C = D \Delta C \begin{cases} C = 0 & \text{for } y = 0 \text{ (active wall)} \\ C = C_0 & \text{for } y \rightarrow \infty \text{ (bulk)} \\ \left. \frac{dC}{dy} \right|_{y=0} & \text{for } y = 0 \text{ (Inert wall)} \end{cases}$$

Transport equation

In permanent regime

$$y\gamma \frac{\partial C}{\partial x} - \frac{y^2}{2} \frac{d\gamma}{dx} \frac{dC}{dy} = D \frac{d^2 C}{dy^2}$$

$$\frac{C}{C_0} = \frac{1}{\Gamma(4/3)} \int_0^\eta \exp(-\xi^3) d\xi \quad \text{avec} \quad \eta = y \sqrt{\gamma} \left[9D \int_0^x \sqrt{\gamma} dx \right]^{\frac{1}{3}}$$

$$\Phi = \int D \frac{dC}{dy} dy \Big|_{y=0} = \frac{3/2}{\Gamma(4/3)} \frac{C_0 D L}{9^{1/3}} \left[\frac{\gamma d_o^2}{D} \right]^{1/3} = 0.807 C_0 A_0 \left(\frac{\gamma D^2}{d_o} \right)^{1/3}$$

Transport equation

$$\Phi = 0.807 C_0 A \left(\frac{\gamma D^2}{d_0} \right)^{1/3}$$

Coefficient
characterizing
wall surface

Coefficient
characterizing
colonized surface

Probability of
effective shocks

$$\beta = \frac{A_f}{A_o}$$

$$A = \alpha_1 A_0 - \alpha_2 A_f$$

Wall flux

$$\Phi = \frac{d \left(N_f v_0 \right)}{dt} = 0.807 C_0 \frac{A_f}{A_0} A_0 \left(\alpha_1 - \alpha_2 \frac{A_f}{A_0} \right) \left(\frac{\gamma D^2}{d_0} \right)^{1/3}$$

Transport equation

The number of bacteria/unit surface (N^*) deposited is related to an adimensional time t^*

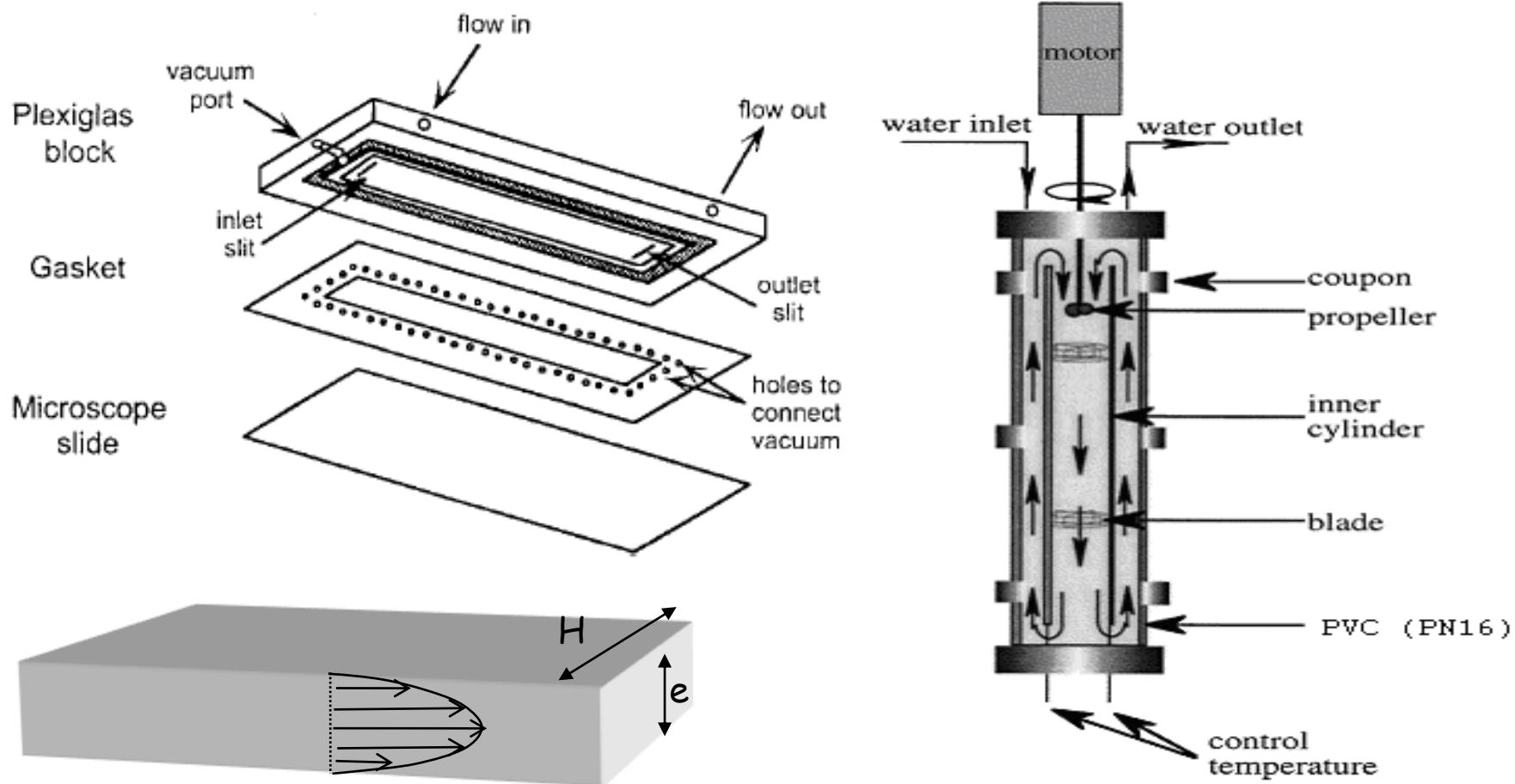
$$N^* = \frac{\alpha_2 s_0}{\alpha_1} + S \exp \left[-0.807 \frac{\alpha_1 s_0 C_o}{v_0} t^* \right]$$

Characterize the saturation

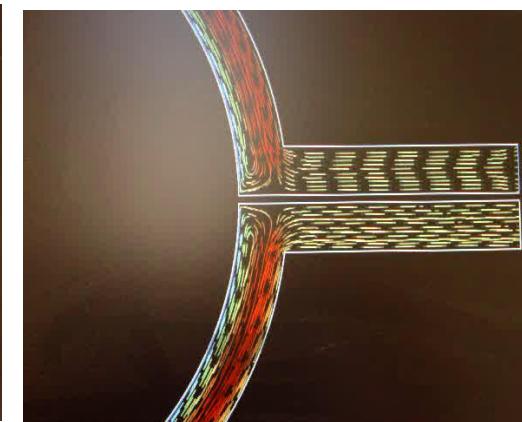
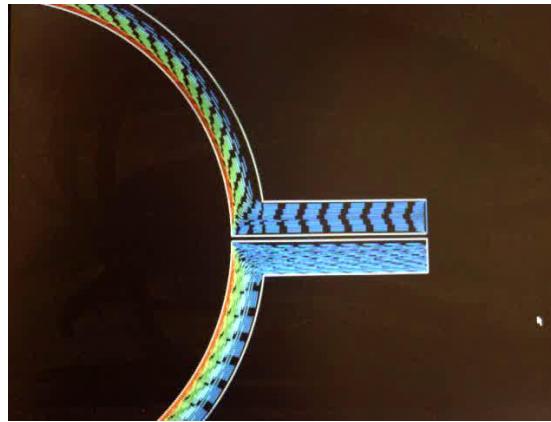
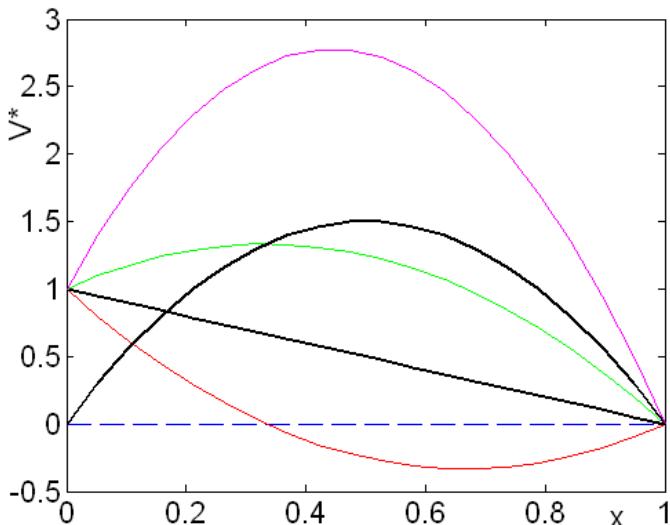
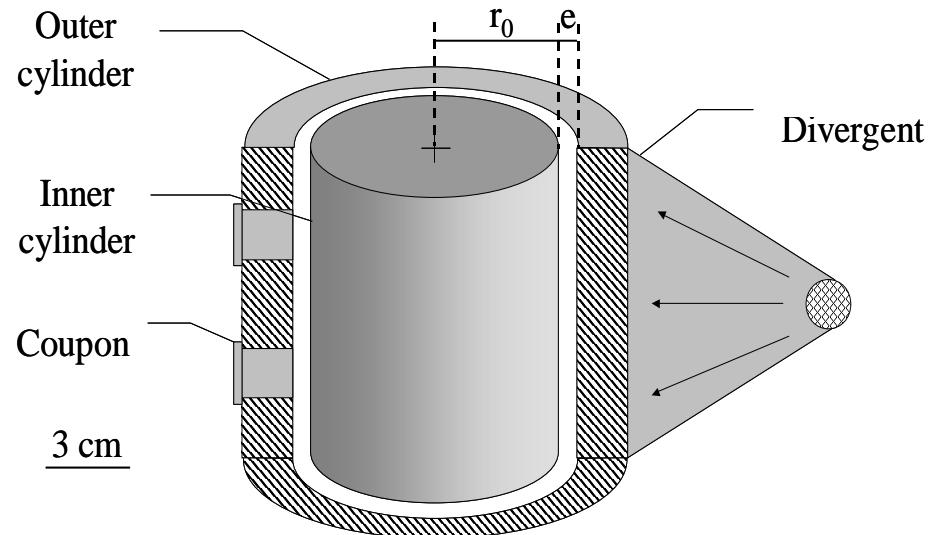
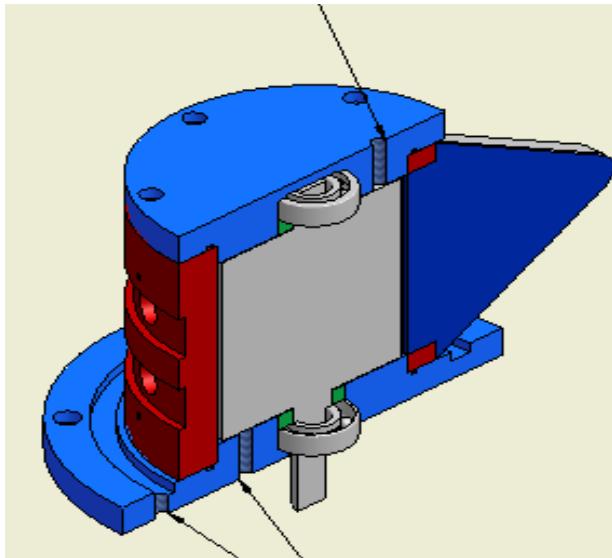
Characterize the initial state of the wall

Characterize the accumulation kinetic

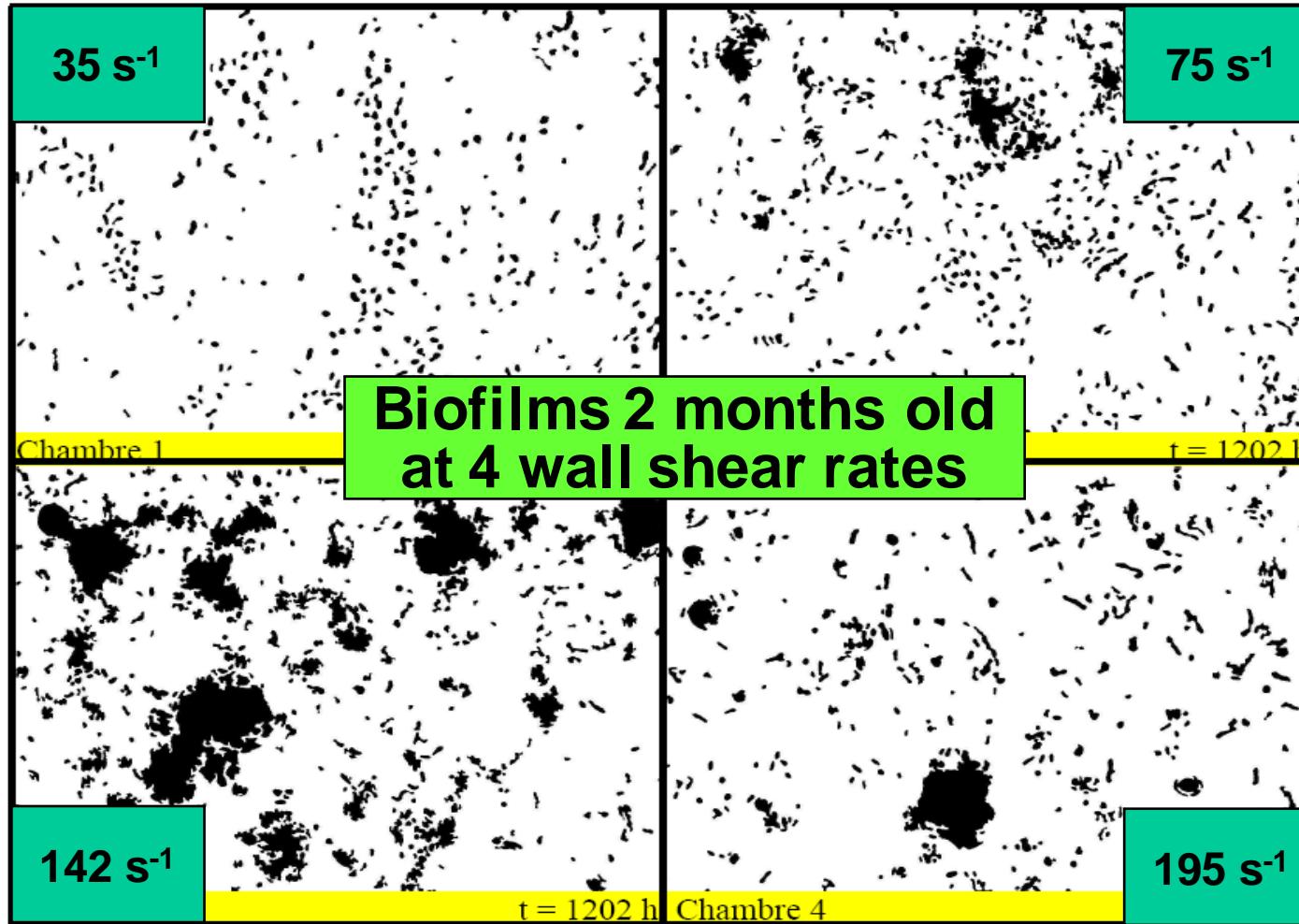
Test sections



Test sections

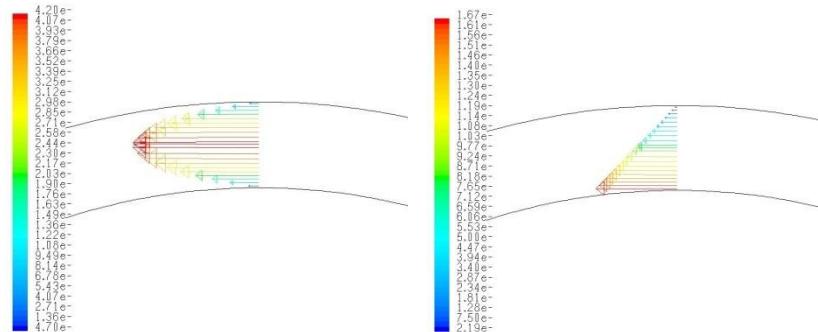


Test sections

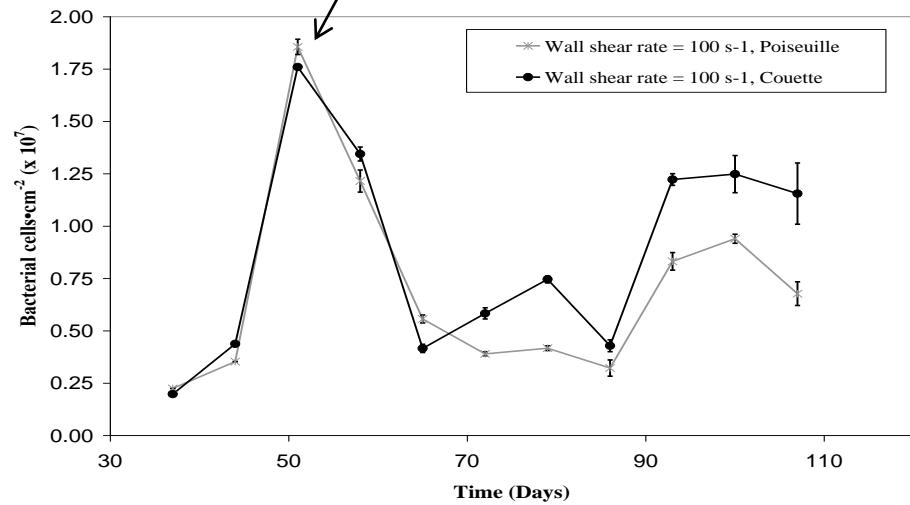
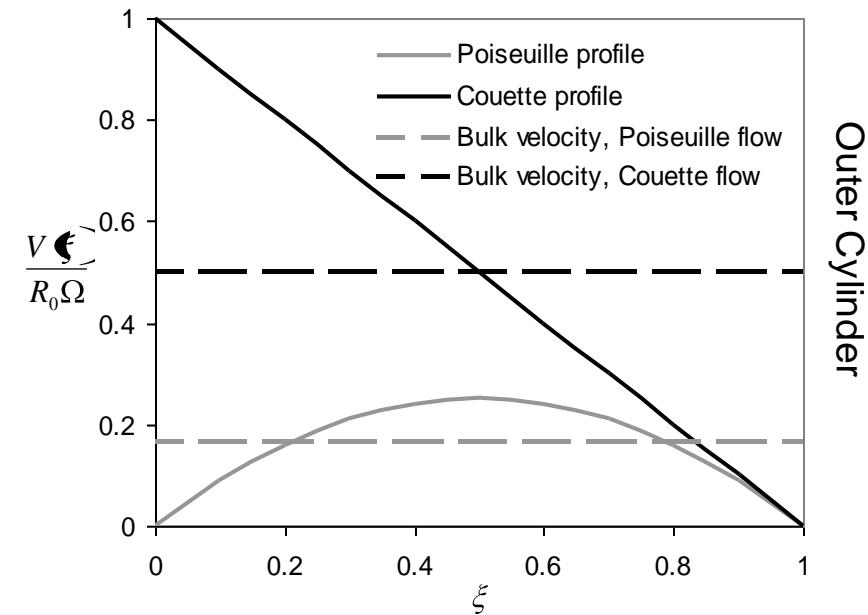


Increasing the wall shear rate led to a space organization of bacteria deposition

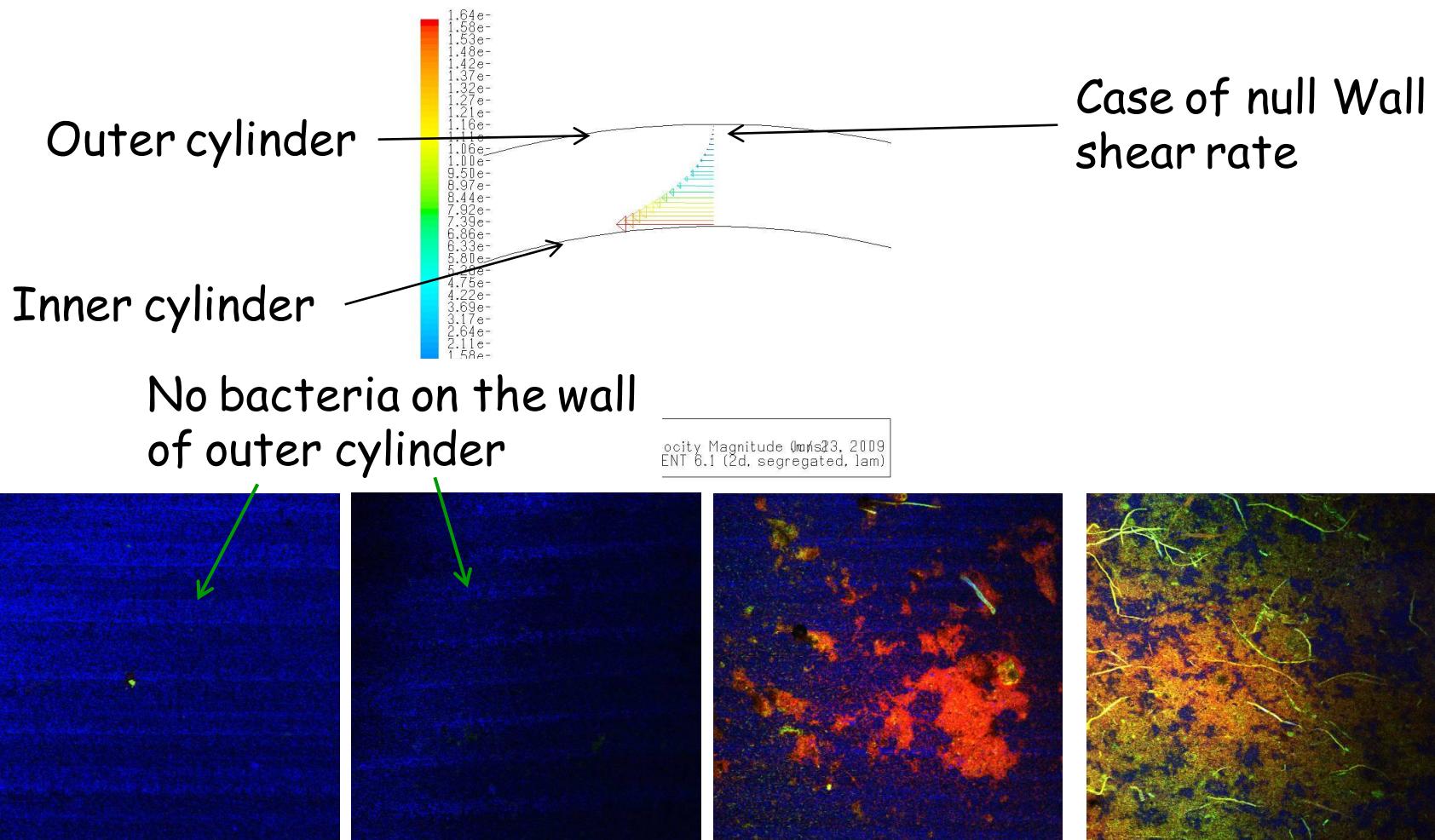
Test sections



Grazing by Amoeba



Test sections



Conclusions on wall flux

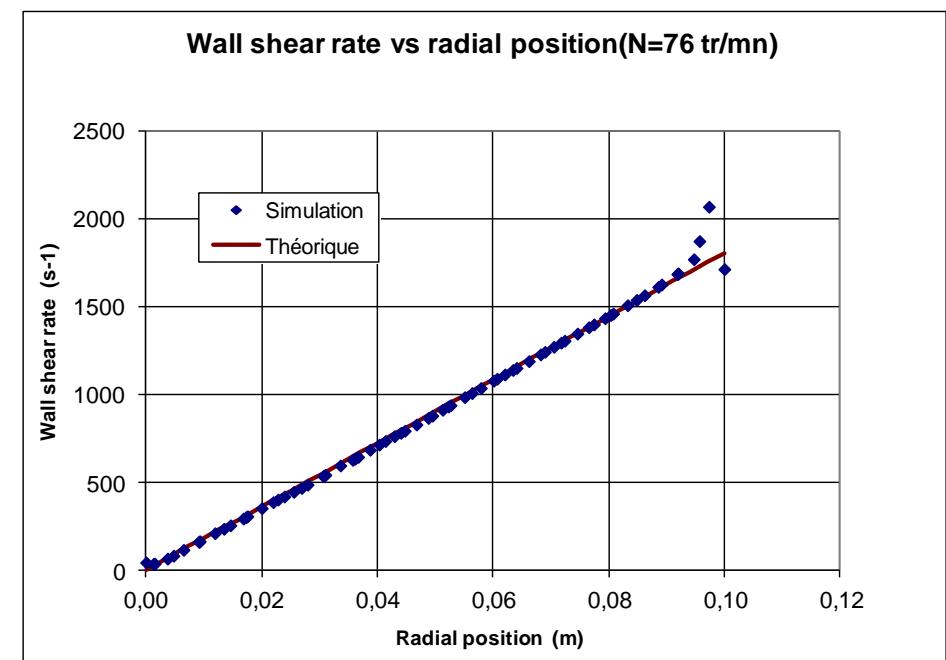
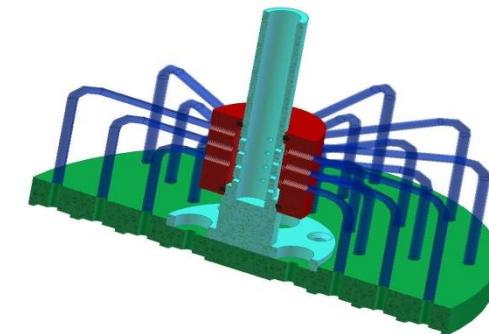
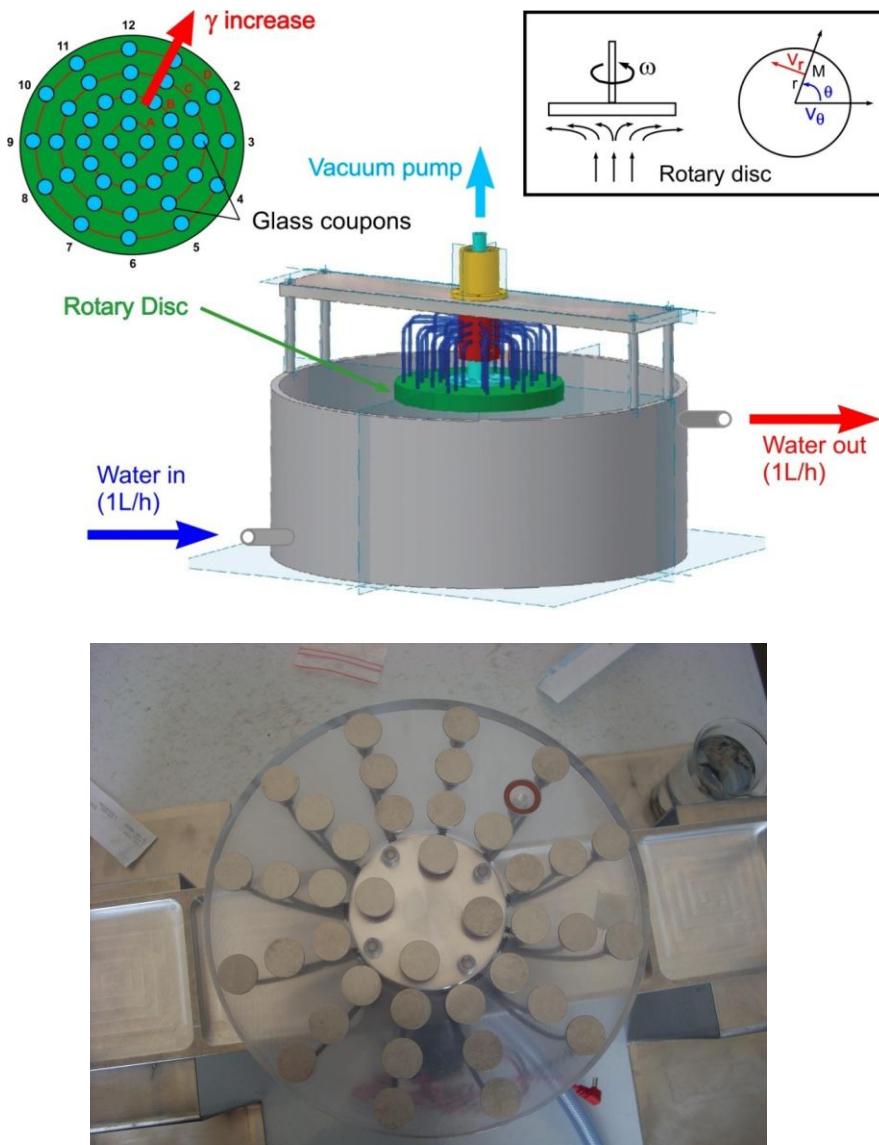
Bacteria in drinking water : migration to the wall -> biofilm ?

No wall shear rate -> no bacteria on the wall

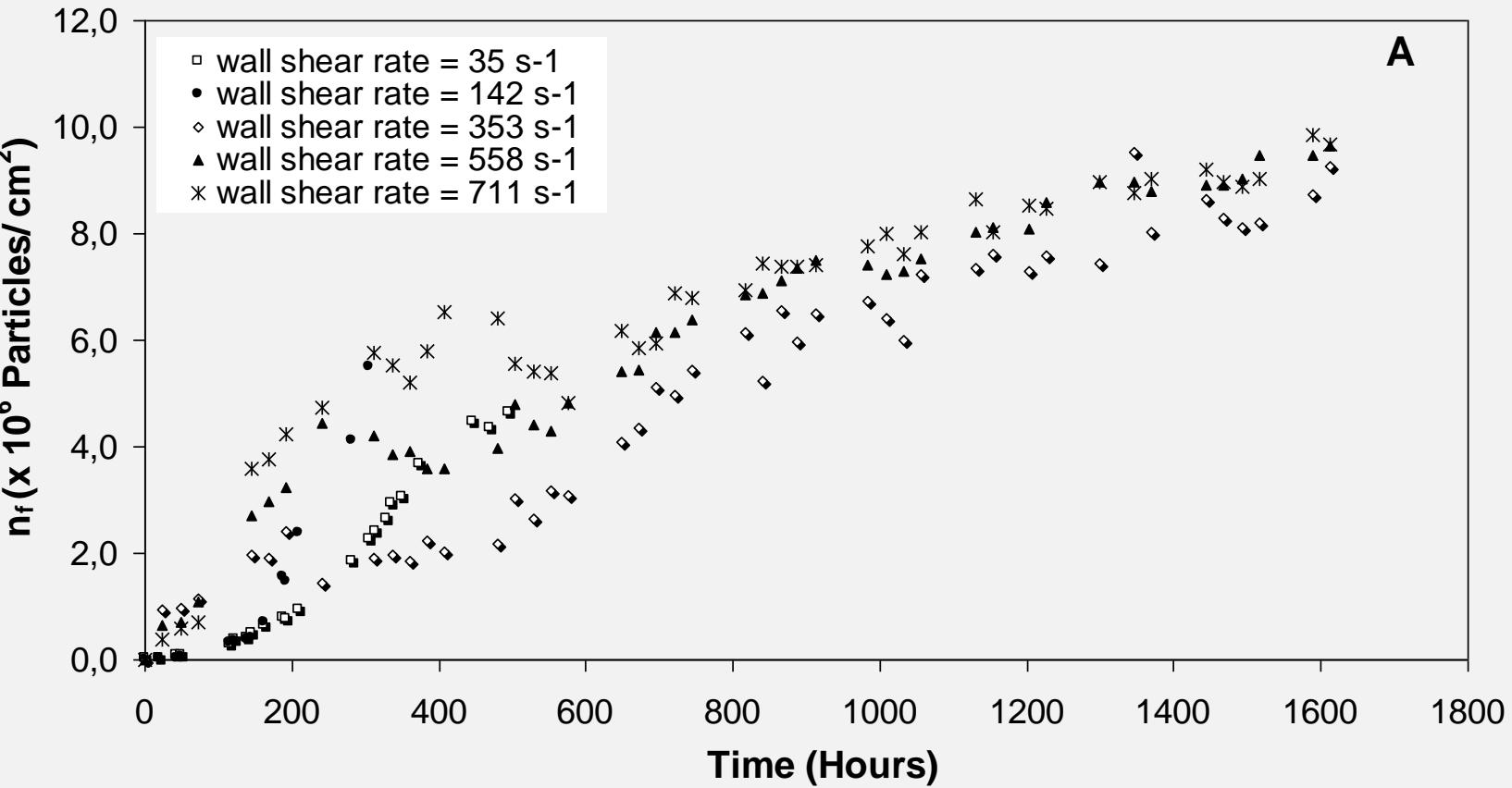
Even if the Peclet number is high -> Convection –diffusion works

$$N^* = \frac{1}{\frac{\alpha_2 s_0}{\alpha_1} + s \exp \left[-0.807 \frac{\alpha_1 s_0 C_o}{v_0} t^* \right]}$$

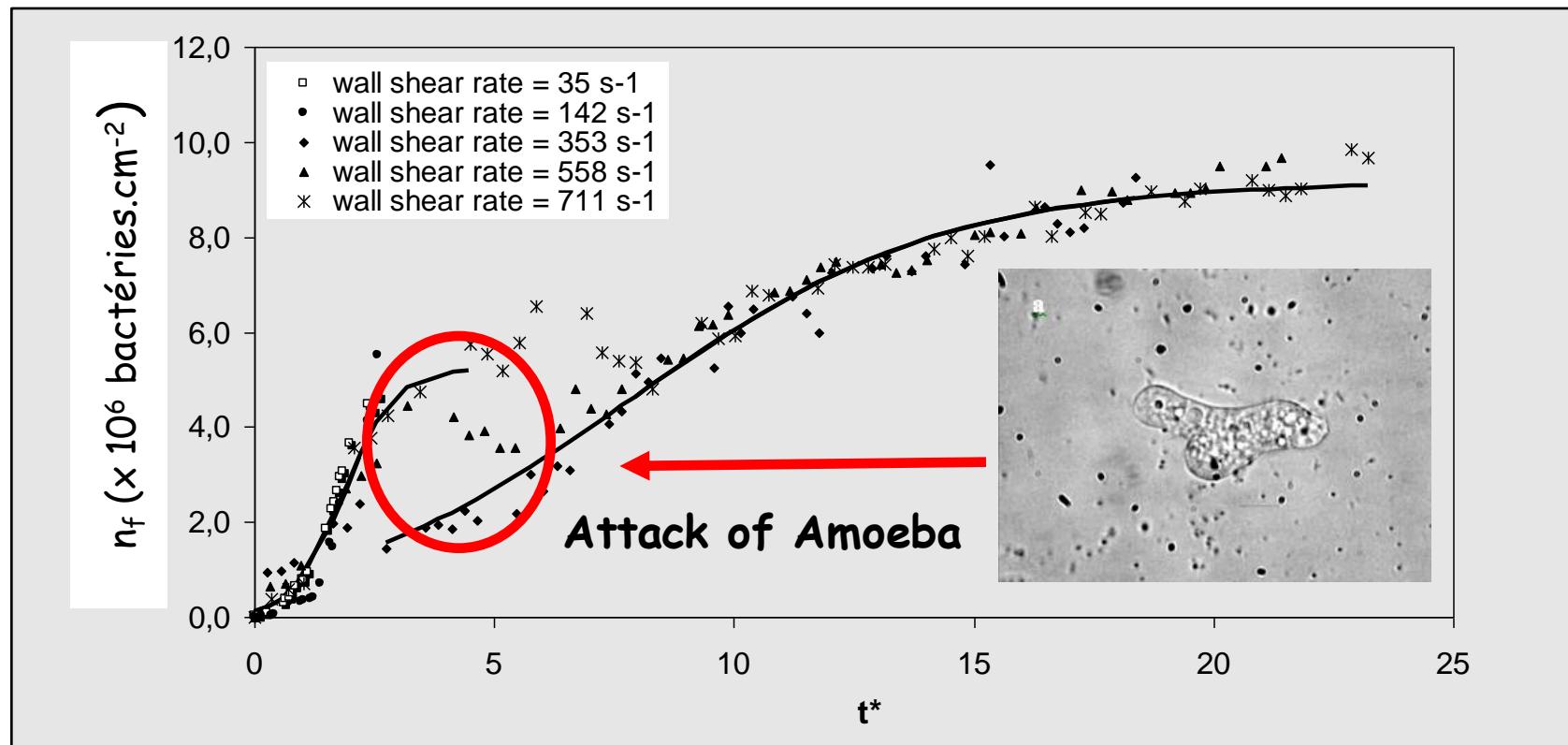
Test sections (Rotating disc)



Results

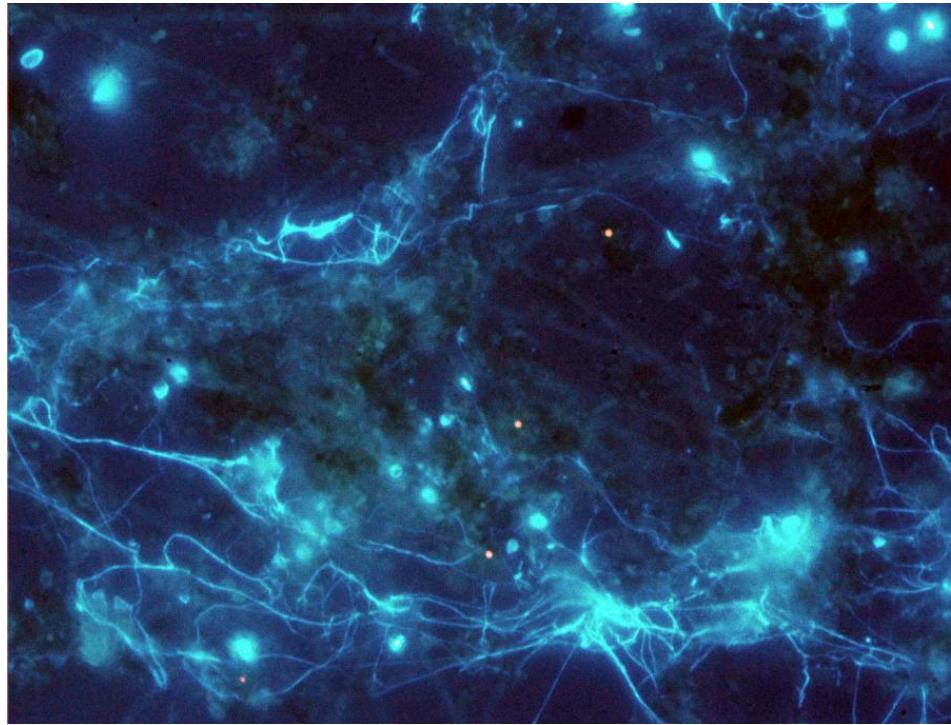


Results



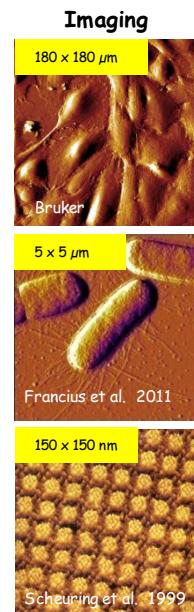
| | Phase 1 | Phase 2 |
|---------|-------------------|-------------------|
| alpha 1 | 1 | 0,3 |
| alpha 2 | 30 | 5 |
| S_0 | $8 \cdot 10^{-6}$ | $1 \cdot 10^{-6}$ |

Bacteria adhesion at the wall (cohesion of biofilm)



Measurement technique

AFM \Rightarrow an application of the scanning tunneling microscope (STM) :
➤ Imaging samples surface in various environments



Various Scales of Observation

Microscopic

Micro

Nano

Molecular

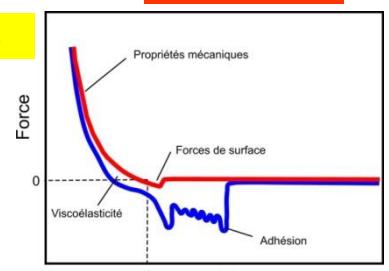
$100 \mu\text{m}$

$1 \mu\text{m}$

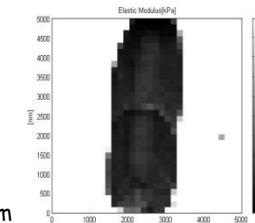
0.1 nm

Spectroscopy

Interfacial Properties



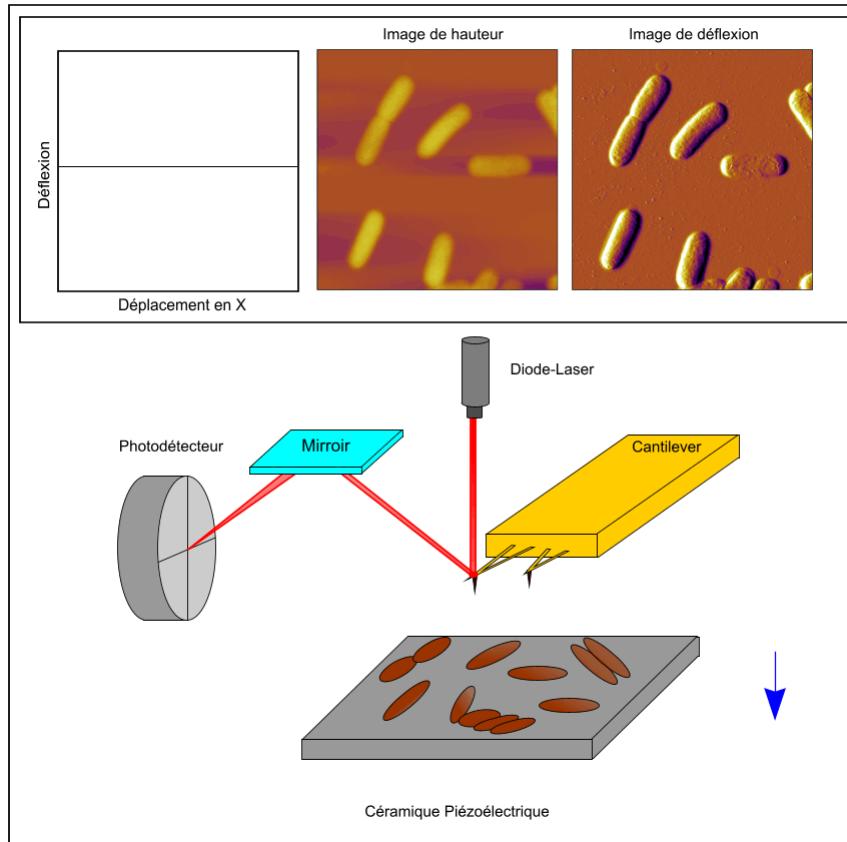
Cartography



In the last decade \Rightarrow experiencing boom of AFM in nanosciences and life sciences :

AFM technique

Panorama of measurable forces in AFM



| Approach | Retraction |
|---|---|
| a van der Waals $F(D) = \frac{AR}{12D^2}$ | e Adhesion $F = -3\pi R\gamma$ |
| b Electrostatic $F(D) = \frac{4\pi R\lambda\sigma_R\sigma_S}{\epsilon} e^{-D\lambda}$ | f Capillary force $F = 4\pi R\gamma_L \cos\theta$ |
| c Brush $F(D) \approx \frac{50LkT}{s^3} e^{-2\pi DL} \quad 0.2 \leq D \leq L \leq 0.9$ | g Polymer extension $F(x) = \frac{kT}{a} L^* \left(\frac{x}{Na} \right)$ |
| d Elastic $F(\delta) = \frac{4E\sqrt{R}}{3(1-\nu^2)} \delta^{3/2}$ | h Binding $F = \frac{U - kT \ln(\tau/\tau_0)}{\Delta}$ |
| Definitions | |
| A Hamaker constant a Monomer length D Probe-sample separation distance E Elastic modulus k Boltzmann's constant L Brush thickness in a good solvent L^* Inverse Langevin function N Number of units in polymer R Radius of probe sphere s Mean distance between polymers | |
| T Absolute temperature U Bond energy x Elongation of polymer δ Indentation depth ϵ Dielectric of the medium γ Surface energy between tip and sample γ_L Surface energy of the liquid ν Poisson ratio | |
| Δ Characteristic length of bond λ Debye length of the medium θ Angle related to the geometry of the tip-sample contact σ_R Surface-charge density of sphere σ_S Surface-charge density of sample τ Period over which the bond will rupture τ_0 Reciprocal of the natural bond frequency | |

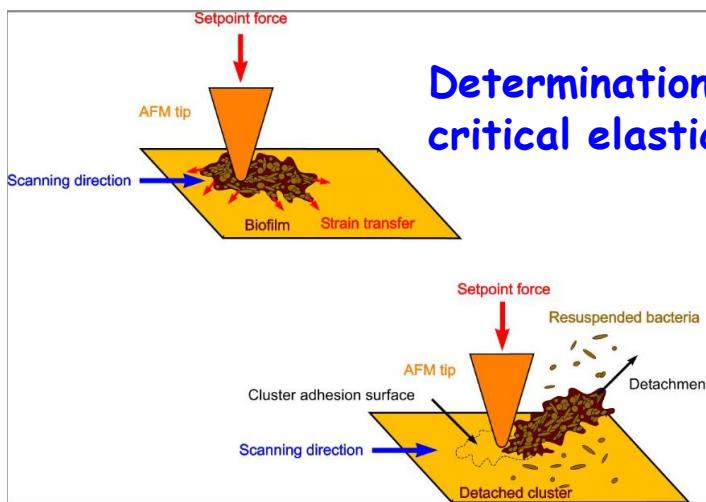
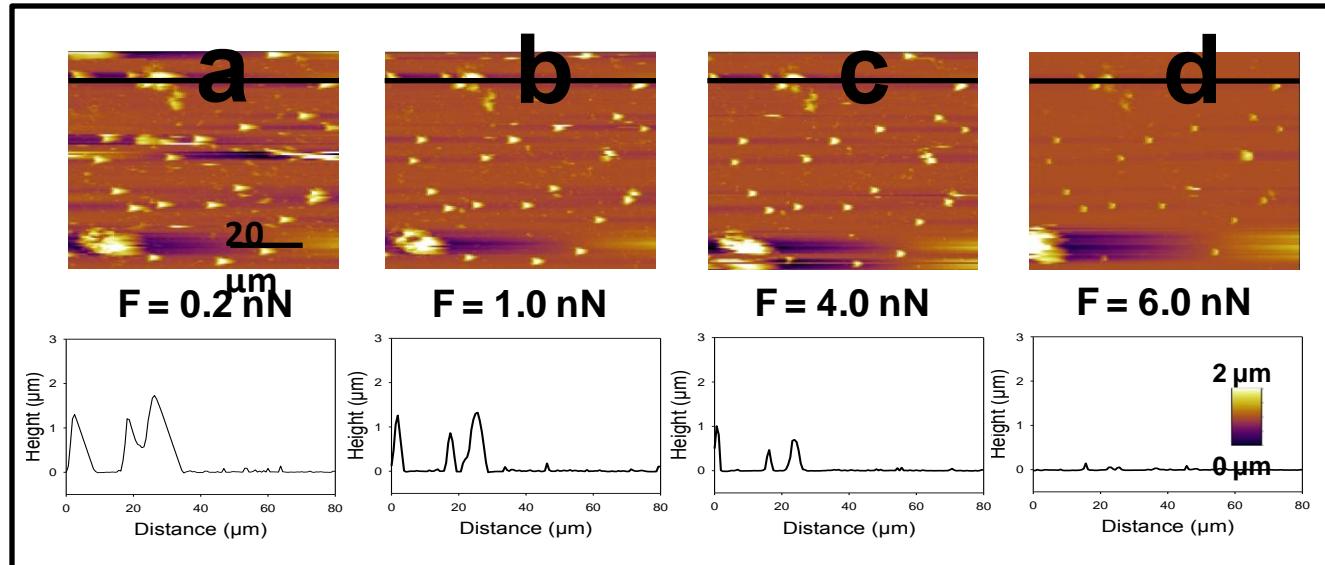
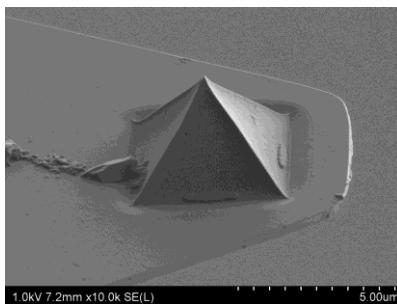
Transfert en Biotechnologie

Atomic force spectroscopy :

- Combination of imaging and force measurements
- Mapping reconstruction with spatially resolved physical parameter value for each pixel
- Which physico-chemical parameters can be extracted from the AFM force-curves ?

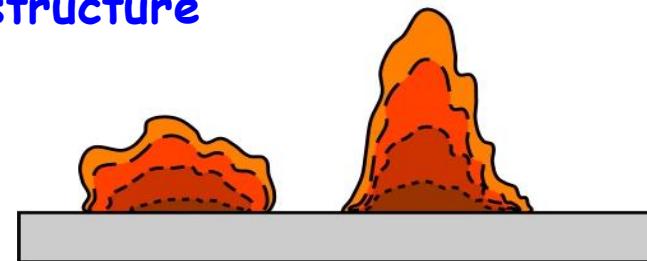
AFM : Cohesion of drinking water biofilm clusters

Scanning surface
by AFM ($F \uparrow$)



Young modulus
0 MPa  10 MPa

Stratification of the cluster structure



Water drinking biofilm

The rate of entanglement ξ is a factor of cohesion linked to the volume and elasticity of the clusters

A large microscopy image on the left shows a dense network of fibers and clusters. A red dashed box highlights a specific region, which is magnified in an inset at the top right. Below the main image, a red dashed line extends from the highlighted area to a schematic diagram. The schematic shows a 3D representation of a cluster of fibers within a blue-shaded volume. A horizontal arrow labeled "3D" indicates the third dimension. A smaller diagram above it shows a single fiber segment with nodes, where the distance between two nodes is labeled ε and the total length of the segment is labeled L . A box contains the equation $\xi = 1 - \varepsilon / L$.

G. De Genné

ε : coherence length

$\xi = 1 - \varepsilon / L$

3D

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AFM : Cohesion of clusters in biofilm drinking water

$$W_{elas} \approx G L^3 \approx G (\xi d)^3 \approx G \xi^3 V_{cluster} \quad \text{avec} \quad \begin{cases} G: \text{Shear modulus elasticity} \\ \xi: \text{rate of entanglement in cluster} \\ V_{cluster}: \text{volume of cluster} \end{cases}$$

$$W_{elas} = k_B T \quad \text{avec} \quad \begin{cases} k_B = 1.38 \cdot 10^{-23} \text{ m}^2 \text{kg/s}^2/\text{K} \text{ Bolzmann constant} \\ T: \text{absolute temperature} \end{cases}$$

At the limit of cohesion, cluster we can put:

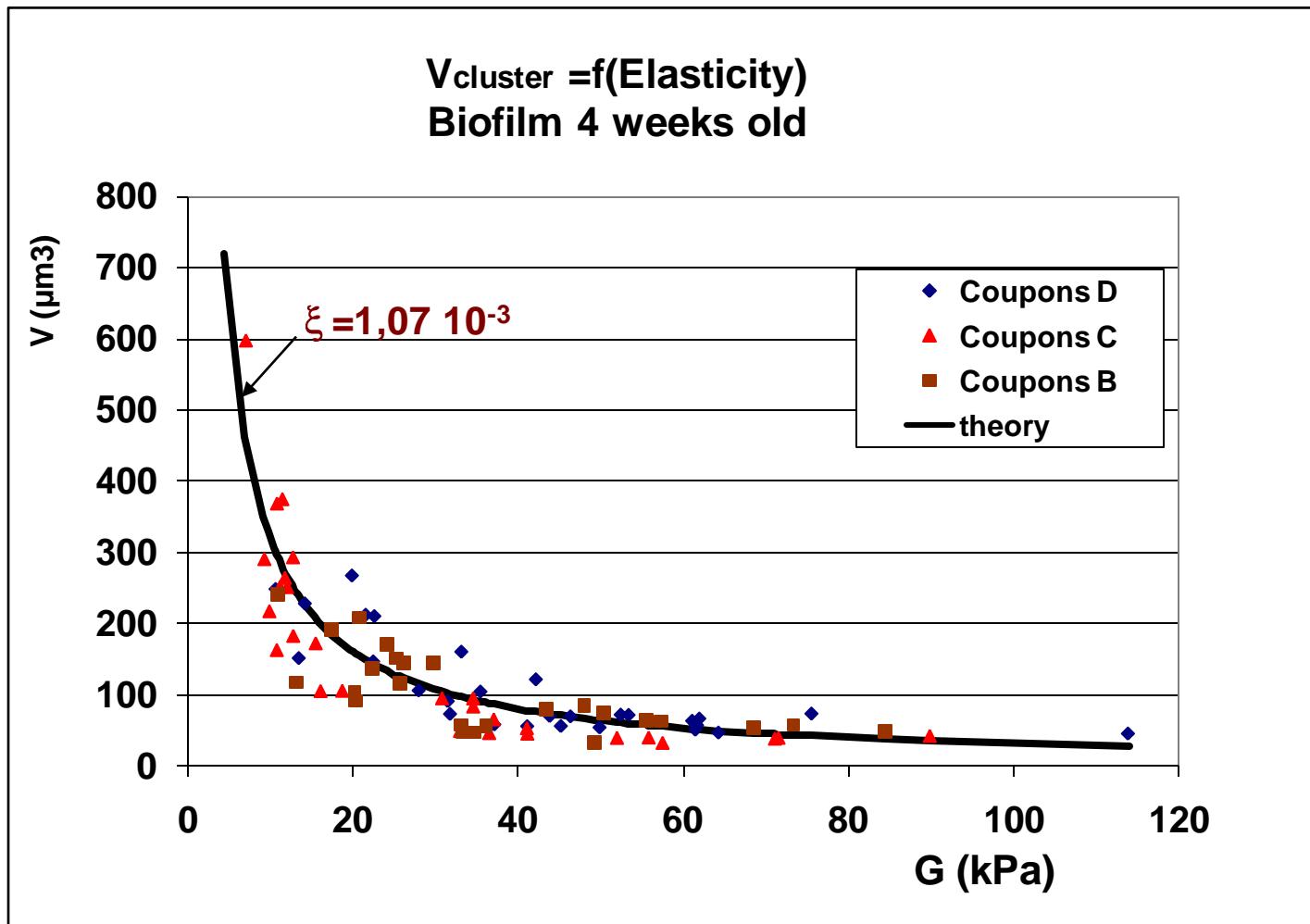
$$G \xi^3 V_{cluster} = k_B T \Rightarrow$$

$$V_{cluster} = \frac{k_B T}{\xi^3 G}$$

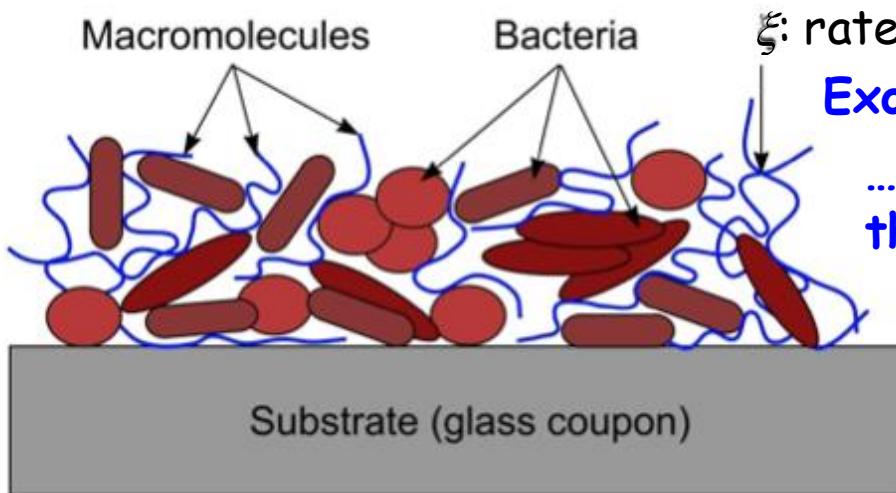
$$k_B T = 4 \cdot 10^{-21} \text{m}^2 \text{Kg/s}^2/\text{K}$$

$V_{cluster}$ and G given by AFM

AFM : Cohesion of clusters in biofilm drinking water

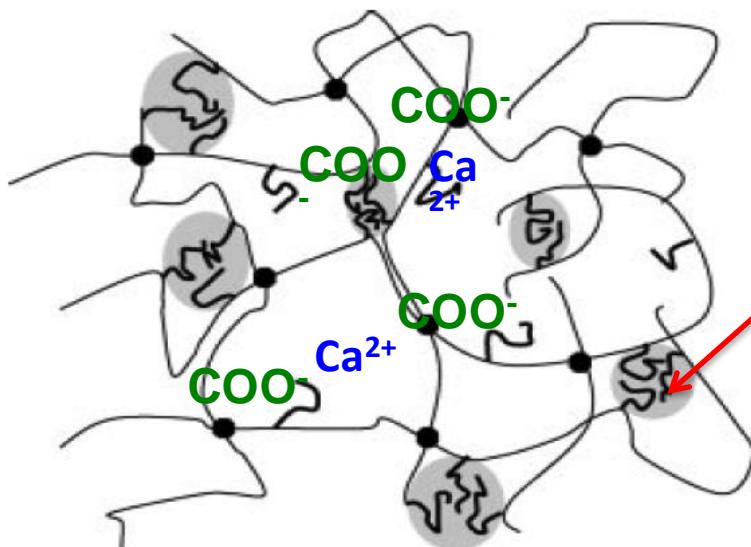


Cohesion of clusters in biofilm drinking water

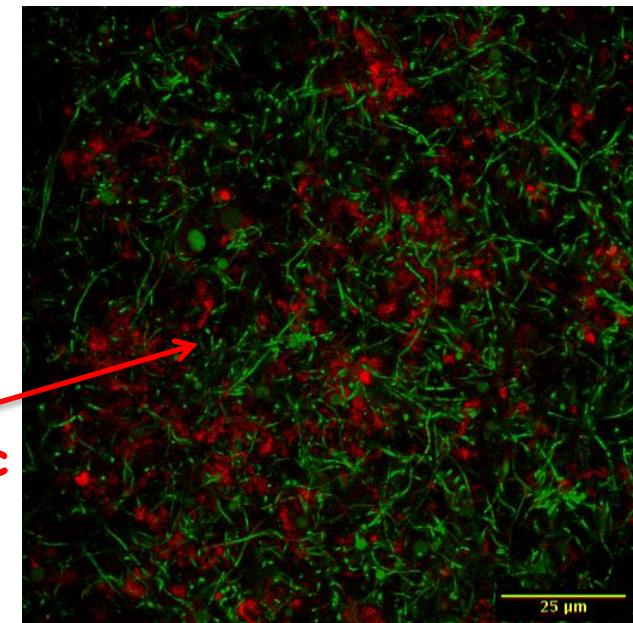


ξ : rate of entanglement

Exopolymers in drinking water biofilms ...
... govern the elastic deformation of
the clusters



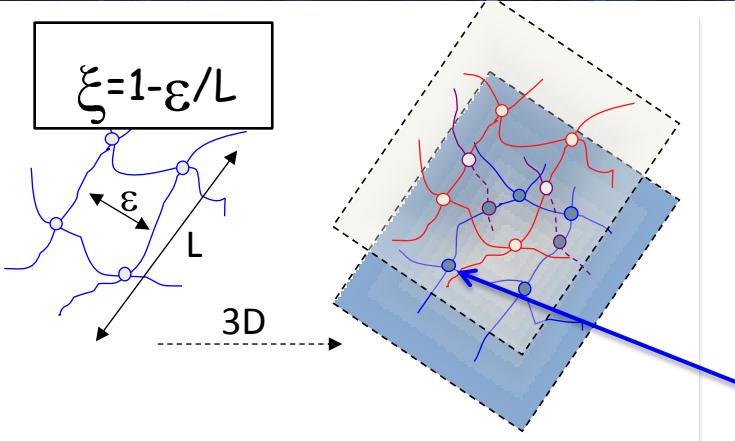
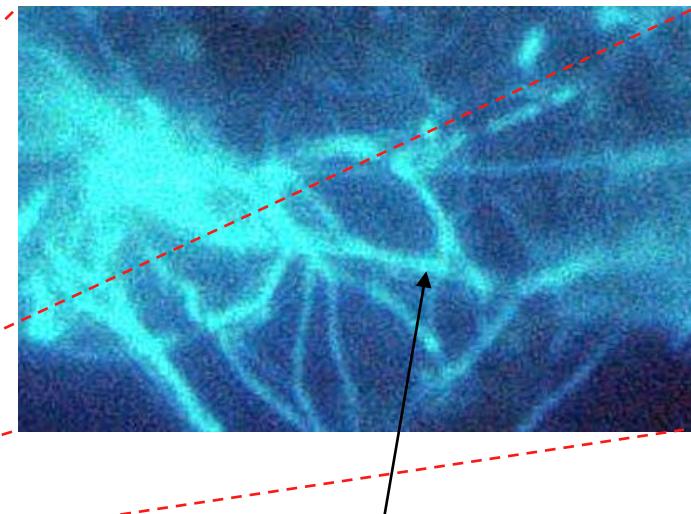
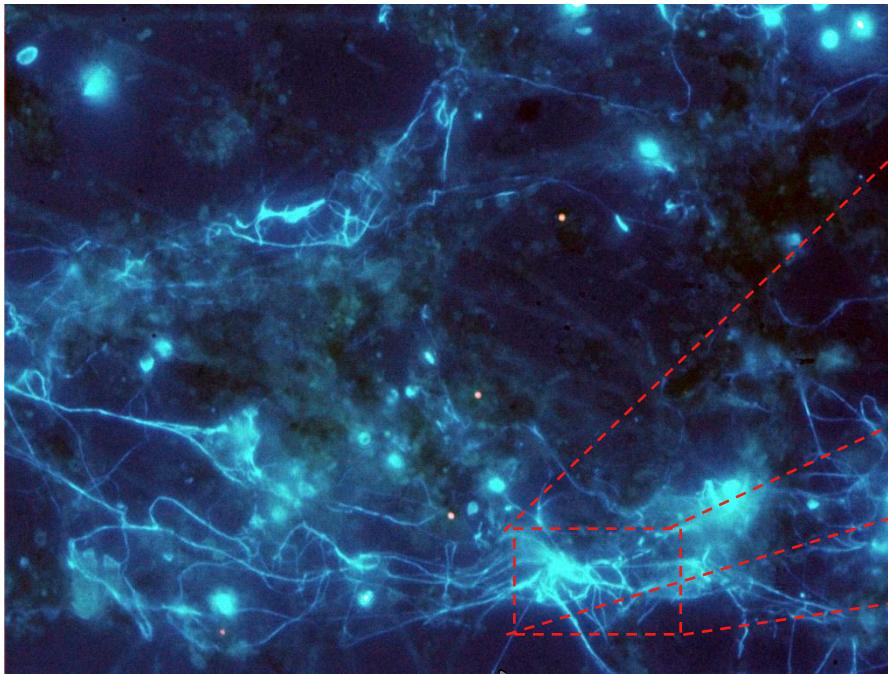
(Miquelard-Garnier et al., 2007)



Aldeek et al., AEM, 2013
(Biofilm of *Shewanella oneidensis*)

Number of entanglement points $10^9/\text{mm}^3$

The rate of entanglement ξ is a factor of cohesion linked to the volume and elasticity of the clusters



$$V_{\text{cluster}} = \frac{k_B T}{x^3 G}$$

Drinking water biofilms behave like a viscoelastic solid

$\xi = 10^{-4} \text{ to } 10^{-3}$

Number of entanglement points $10^8 \text{ to } 10^9/\text{mm}^3$

Cleaning the surface colonized by biofilm hydrodynamic shear stress vs cluster volume

Mean shear stress in volume of cluster V: $\sigma_{ij} = \frac{1}{V} \sum_1^n \left(\int_{A_0 - A_1} \sigma_{ik} x_j n_k dA + \int_{A_1} \sigma_{ik}^b x_j n_k dA \right)$

$$\sigma_{ij} = \sigma_{ij}^f C (1 - \xi) + \sigma_{ij}^b C \xi$$

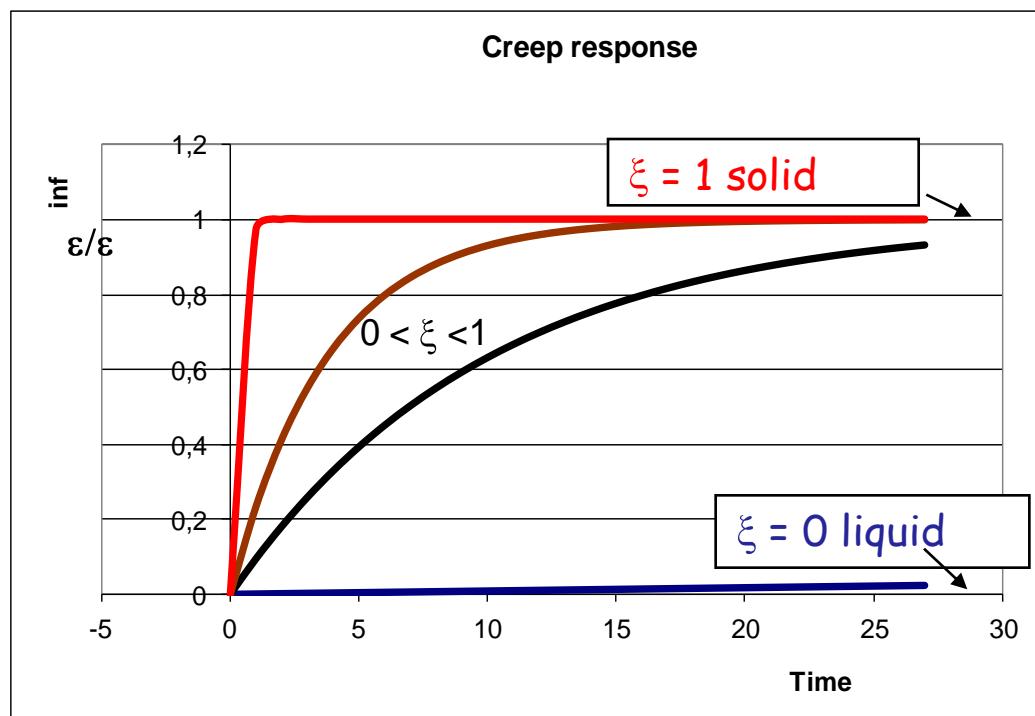
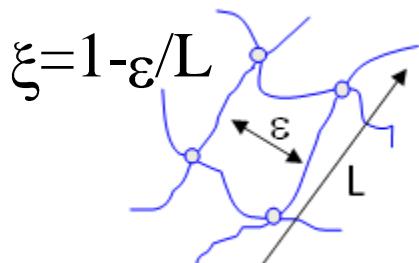
$$\sigma_{ij}^f = \mu \frac{d\alpha}{dt} = \mu \gamma \quad \text{et} \quad \sigma_{ij}^b = G \alpha$$

$$\tau = \mu C (1 - \xi) \frac{d\alpha}{dt} + G C \xi \alpha$$

Creep function

$$f(t) = \frac{1}{G \xi} \left(1 - e^{-\frac{t}{\theta}} \right)$$

$$\text{with } \theta = \frac{\mu (-\xi)}{G \xi}$$



Cleaning the surface colonized by biofilm hydrodynamic shear stress vs cluster volume

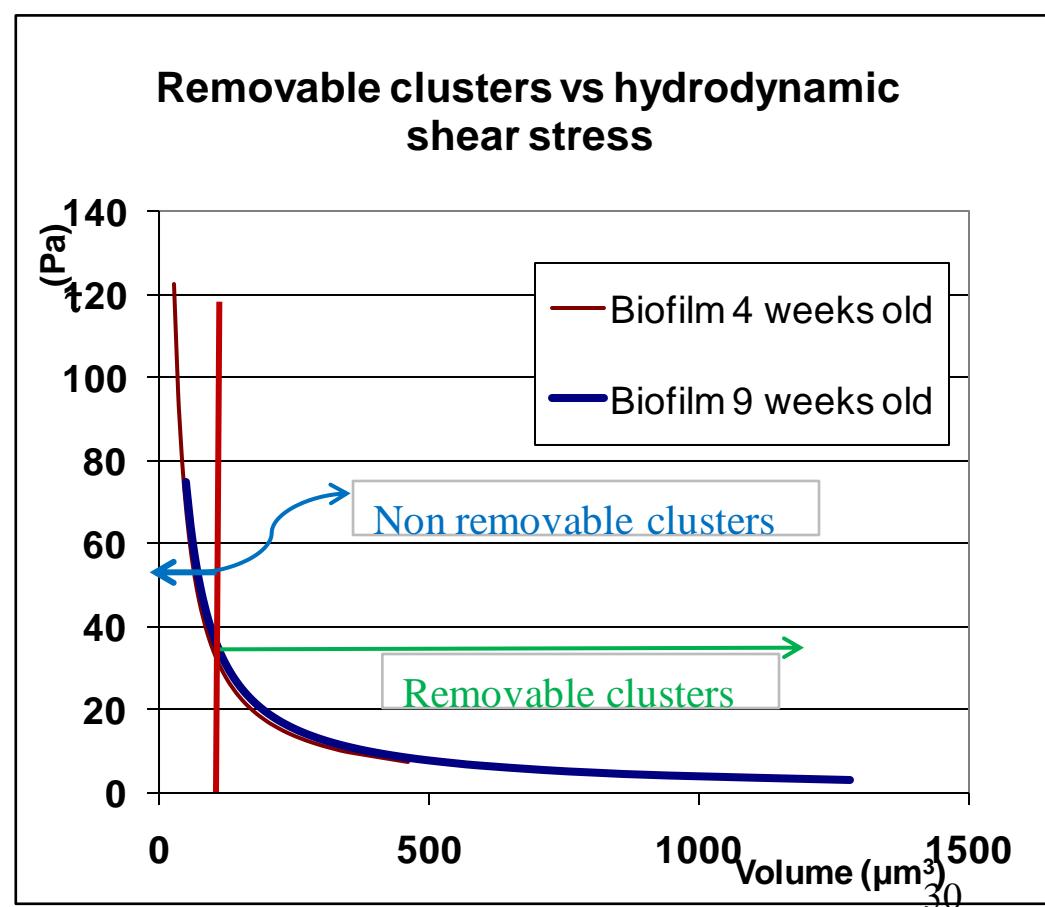
Elastic limit before removing clusters:

$$\mu \frac{d\alpha}{dt} C(1-\xi) = \mu \gamma C(1-\xi) = G C \xi \alpha_{\max}$$

Strain rate $\alpha_{\max} = 100\%:$

$$\tau_{hyd} = \mu \gamma = \frac{G \xi}{1 - \xi}$$

City network drinking water distribution $\tau < 30 \text{ Pa}$



Conclusions

- Biofilm formation –convective diffusion
- Strong adhesion at the wall (AFM measurements)
- Cleaning the surface colonized by biofilm with only hydrodynamic shear stress -> volume clusters $<100\mu\text{m}^3$ non removable
- Perspective : diffusion of nutrients on biofilm - population balance (growth, mortality and partial detachment)

THANK YOU FOR YOUR ATTENTION

- Tony Paris, Salaheddine Skali-Lami, Jean-Claude Block (2007) : *Effect of Wall Shear Rate on Biofilm Deposition and Grazing in Drinking Water Flow Chambers*. Biotechnology and Bioengineering, V. 97, issue. 6: 1550_1561.
- Paris T., Skali-Lami S., and Block J.C. (2009) : *Probing young drinking water biofilms with hard and soft particles*. *Water Research*, v. 43, issue 1: 117 – 126
- El Khatib R., Lartiges B., Mustin C., Skali-Lami S. (2011) : *Hydrodynamic control of wastewater biofilm organization*. *La Houille Blanche*, Issue 5: 29 – 33.
- Abe Y., Polyakov P., Skali-Lami S., and Francius G. (2011) : *Elasticity and physico-chemical properties during drinking water biofilms formation*. *Biofouling*, v. 27(7): 739 – 750.
- Abe Y., Skali-Lami S., Block J.C., and Francius G. (2012) : *Cohesiveness and hydrodynamic properties of young drinking water biofilms*. *Water Research*, 46: 1155 – 1166.

