Transport phenomena in a suspension of swimming micro-organisms

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Introduction

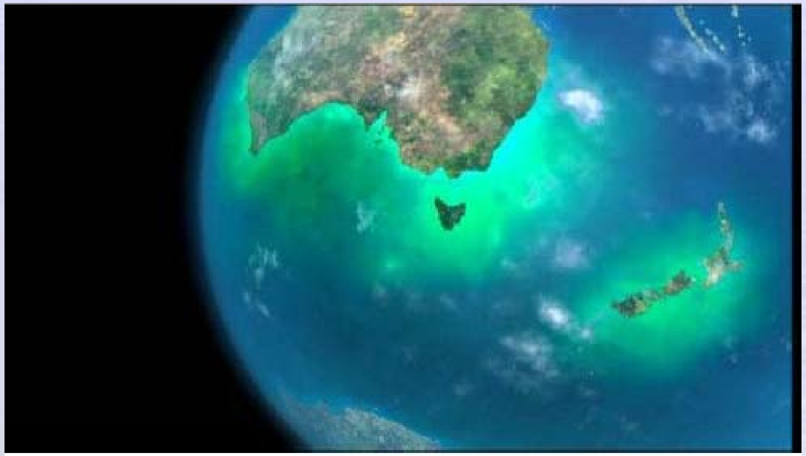
The role of the infinitely small in nature is infinitely large.

Louis Pasteur (1822-1895)

Micro-organisms in the ocean

50% of biomass
Bottom of food chain
⇨Oceanic ecosystem

Absorption of CO_2
Nitrogen cycle
⇨Global environment



Plankton blooms around Australia and New Zealand in October.Picture from Byatt et al. (2001)

Introduction

Micro-organisms in bioreactors

Making food

Yeast, Lactic acid bacterium

⇒Food industry

Making medicine and cosmetics

⇒Chemical industry

Sewage treatment

⇒Plant industry

Algae fuel

an alternative to fossil fuel

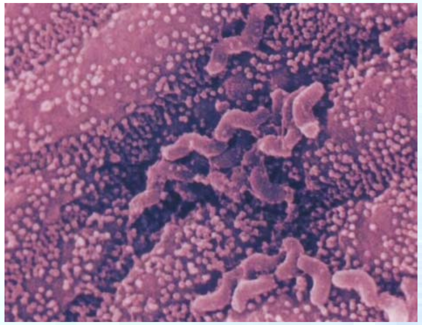
⇒Energy revolution?

Introduction

Micro-organisms in human body

hundreds of species
about 10^{14} cells

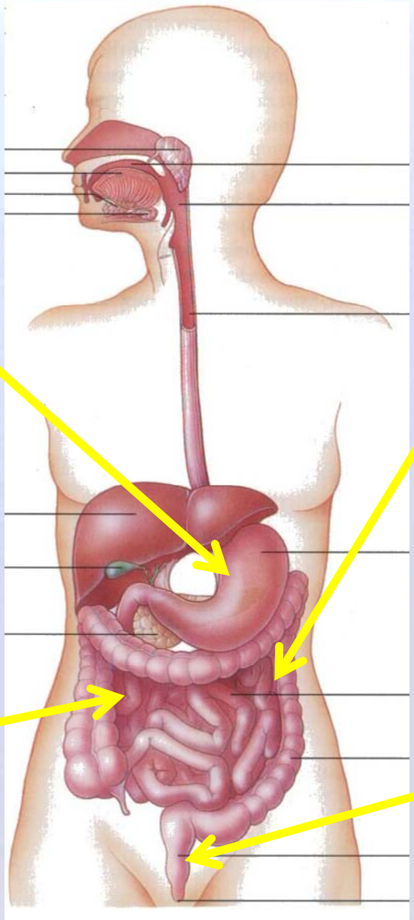
Helicobacter pylori
in the stomach



from Introduction to Microbiology

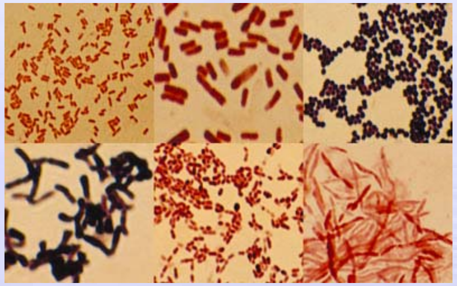
Infection by *salmonella*

from Introduction to Microbiology



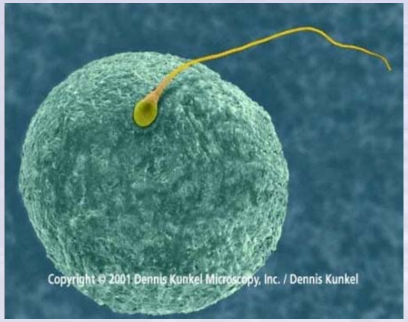
from Introduction to Microbiology

Digestion helped by
enterobacteria
(vitamin K)



http://www.riken.go.jp/

Reproduction owed
to sperm swimming



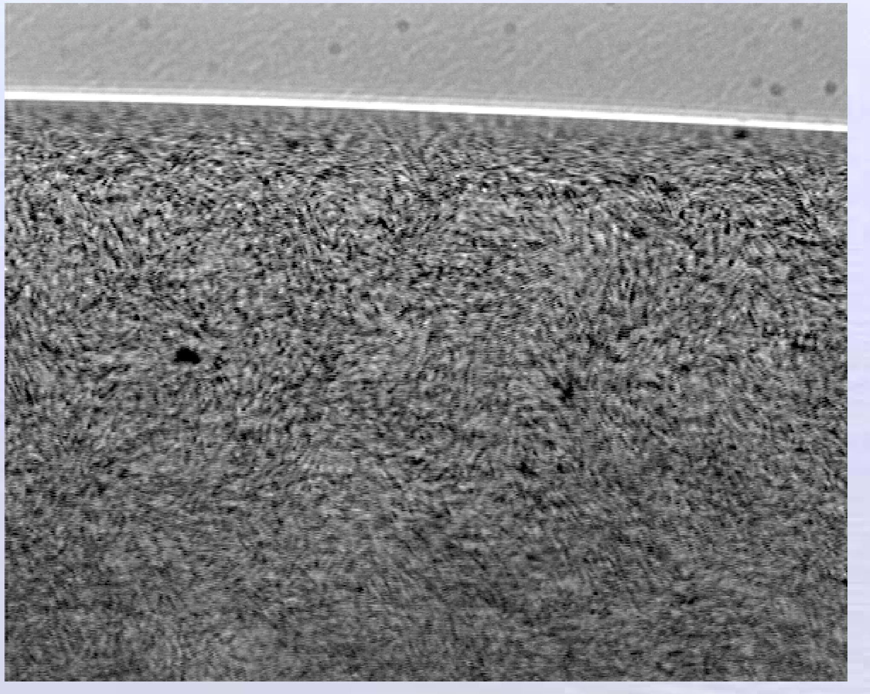
Introduction

In order to understand variety of micro-organisms' phenomena Ecology, Biology & Chemistry have been used.

One example of bacterial phenomena

In this suspension, chemical substances spread 10^3 times more than the Brownian diffusion.

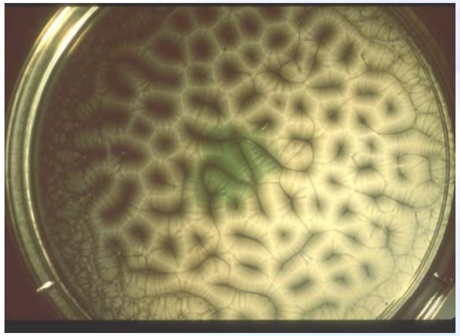
Can we explain this by ecology, biology or chemistry?



Cell motions of *Bacillus subtilis*.
Movie from R. E. Goldstein Lab

Biophysics & biomechanics can contribute more in this field

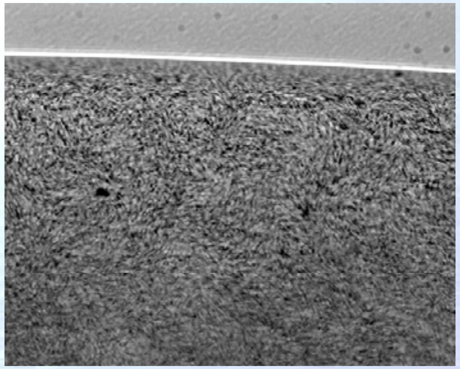
Bottom-up Strategy



Macroscopic level

Rheological and Diffusion properties

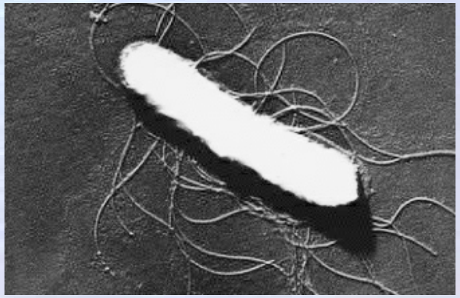
Strong influence



Mesoscale level

Collective motions, Coherent structures

Strong influence



Cellular level

Cell-cell interactions

Bottom-up Strategy

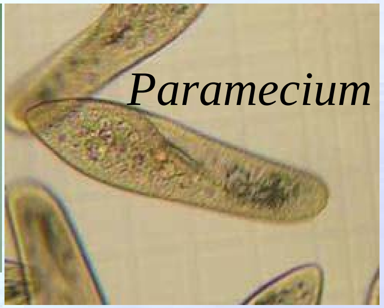
Outline of the talk

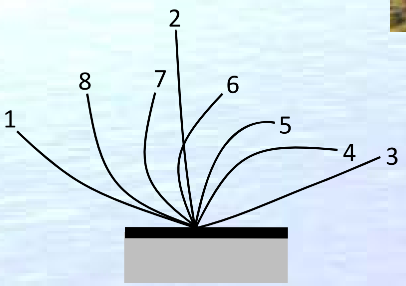
- Introduction
- Biomechanics of an **individual** and a **pair** of micro-organisms
- **Collective swimming** in meso-scale
- **Macroscopic properties** of a suspension of micro-organisms
- Conclusions

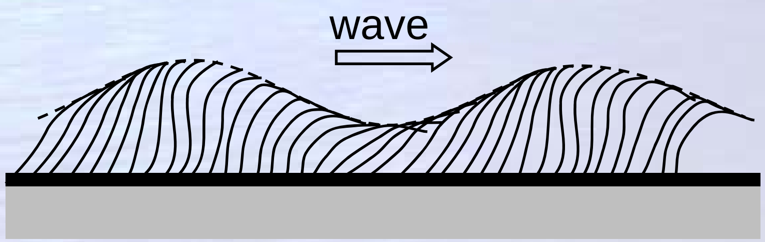
Classification of micro-organisms

In terms of swimming motion

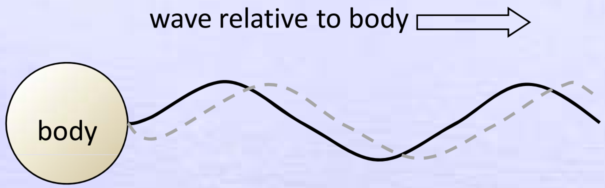
Ciliate



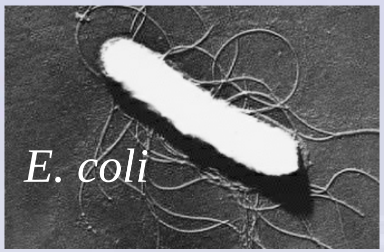


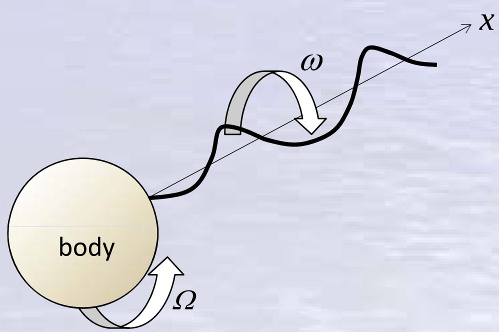


Flagellate (eukaryote)



Bacteria





Hydrodynamics of a single cell

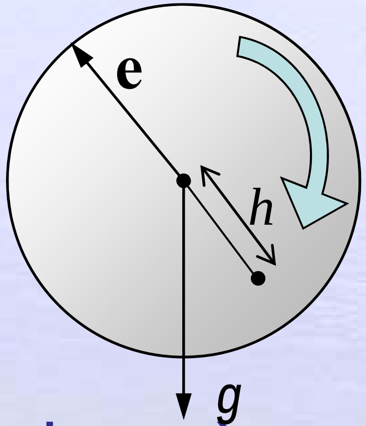
Flow field

Size of a single cell: 1-100μm

Swimming speed: 1-10 body length / sec

⇨ Re = 10^{-6} – 10^{-3}

Stokes flow (Inertia-free)



Force-Torque condition of a cell

Force is almost free

Torque may not be free (bottom-heaviness)

Review paper

Brennen & Winet, *Ann. Rev. Fluid Mech.* (1977)

Lauga & Powers, *Rep. Prog. Phys.* (2009)

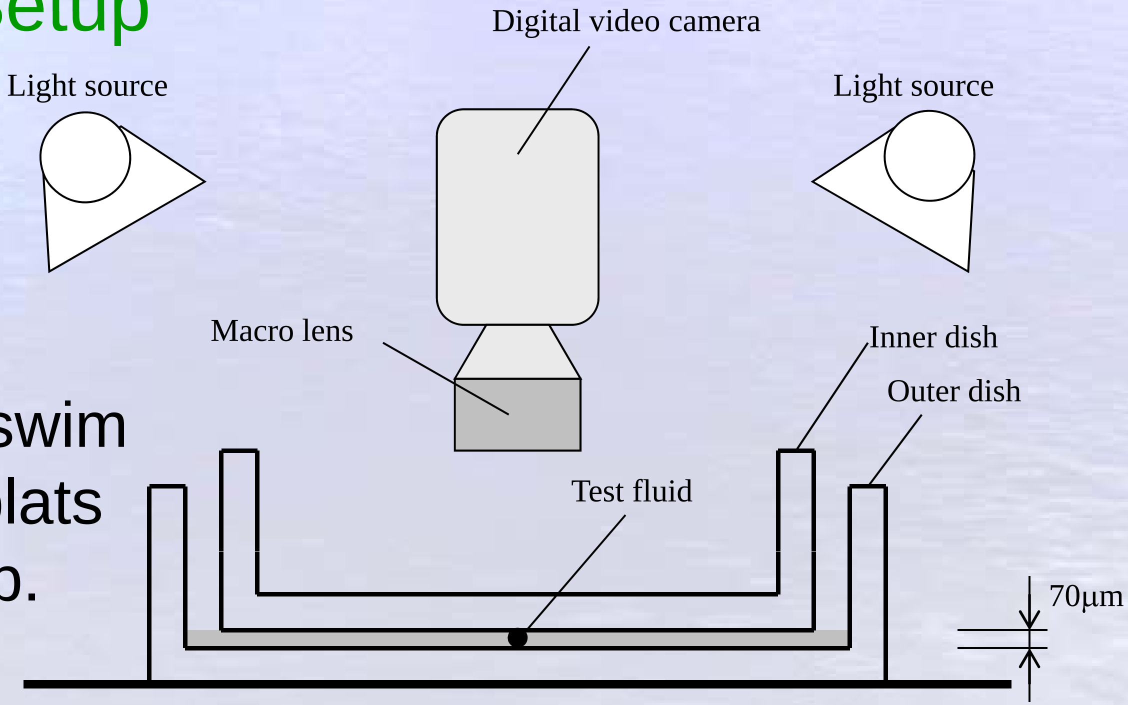
<u>When two cells come close, what happens?</u>

Experiment of *Paramecia*

Paramecium caudatum

Approximately
250μm in length
50μm in width

Experimental Setup

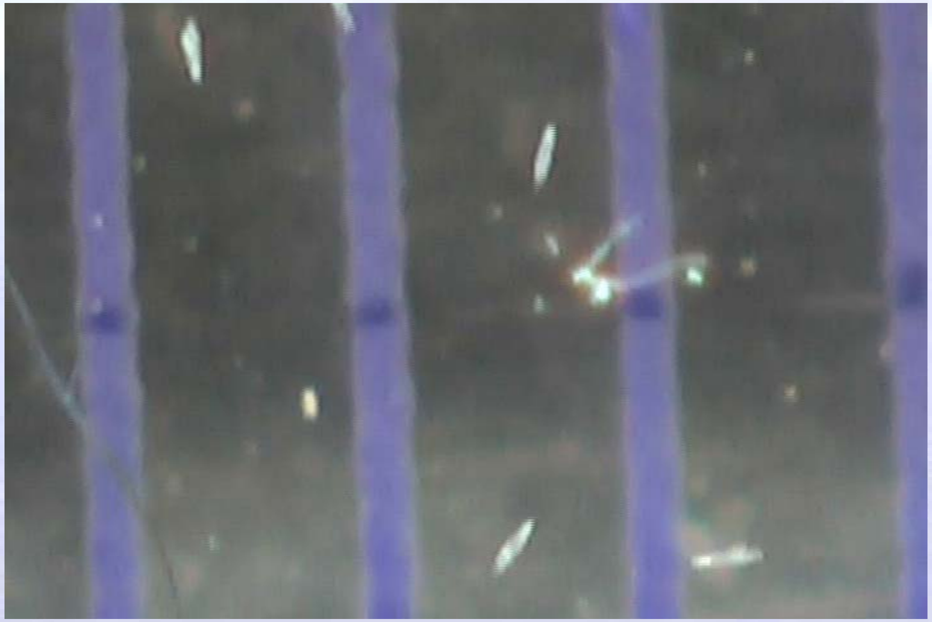


Paramesium swim between flat plats with 70μm gap.

Biological reaction

Avoiding Reaction (AR)

Anterior end: Ca^{2+} channel



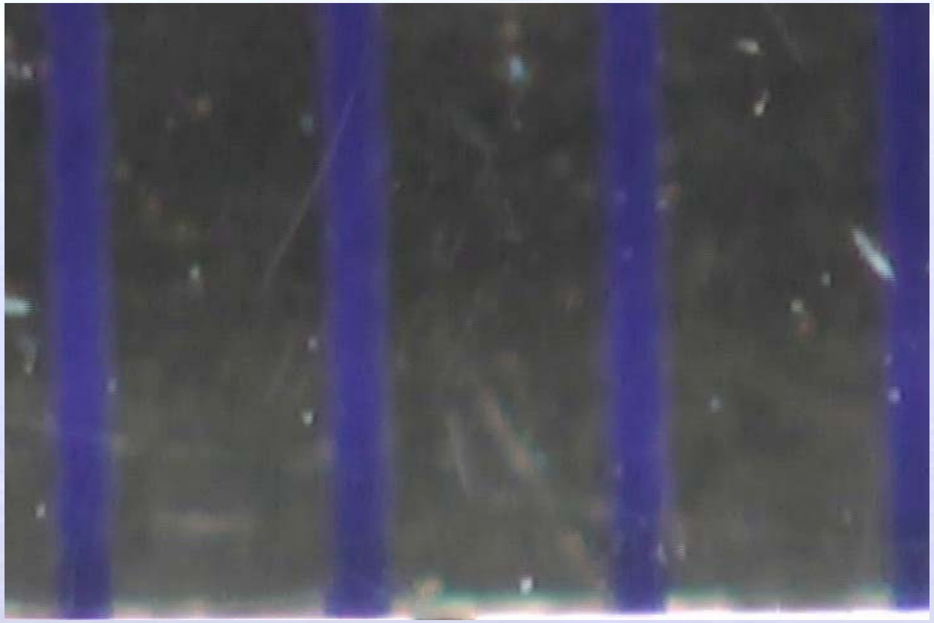
Escape Reaction (ER)

Posterior end: K^{+} channel

Ishikawa and Hota, *J. Exp. Biol.* (2006)

Hydrodynamic interaction

Initially facing each other



Two orientation vectors initially have a large angle

Ishikawa and Hota, *J. Exp. Biol.* (2006)

Experimental results

Ratio of three kinds of interaction

The total number of experimental cases recorded in this study is 301, and the total number of cells is 602.

Kinds of interaction	Number of cells	Percent [%]
Hydrodynamic Interaction (HI)	510	84.7
Avoiding Reaction (AR)	29	4.8
Escape Reaction (ER)	63	10.5

Ishikawa and Hota, *J. Exp. Biol.* (2006)

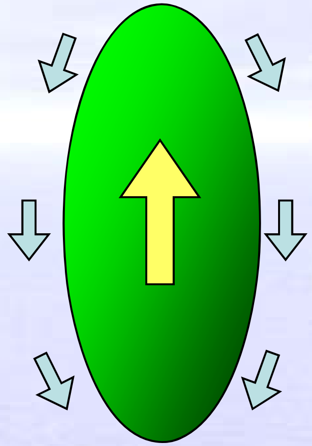
Mainly hydrodynamic interaction

How to model *Paramecium*?

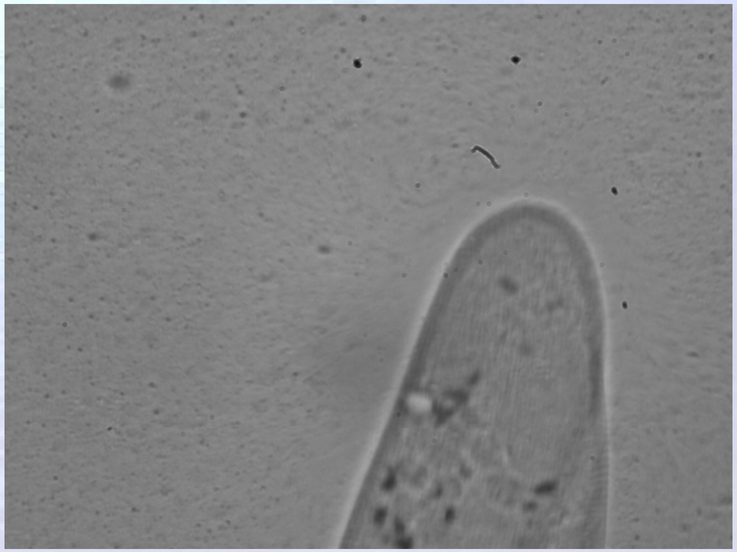
Squirmer model

assumed to propel itself by generating tangential velocities on its surface.

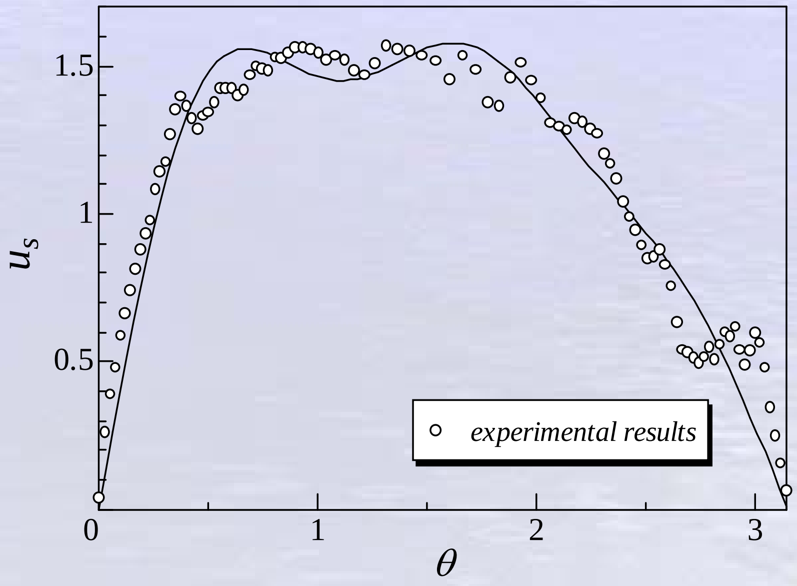
Surface velocity is given as a B.C.



Velocity field around *Paramecium*



PIV



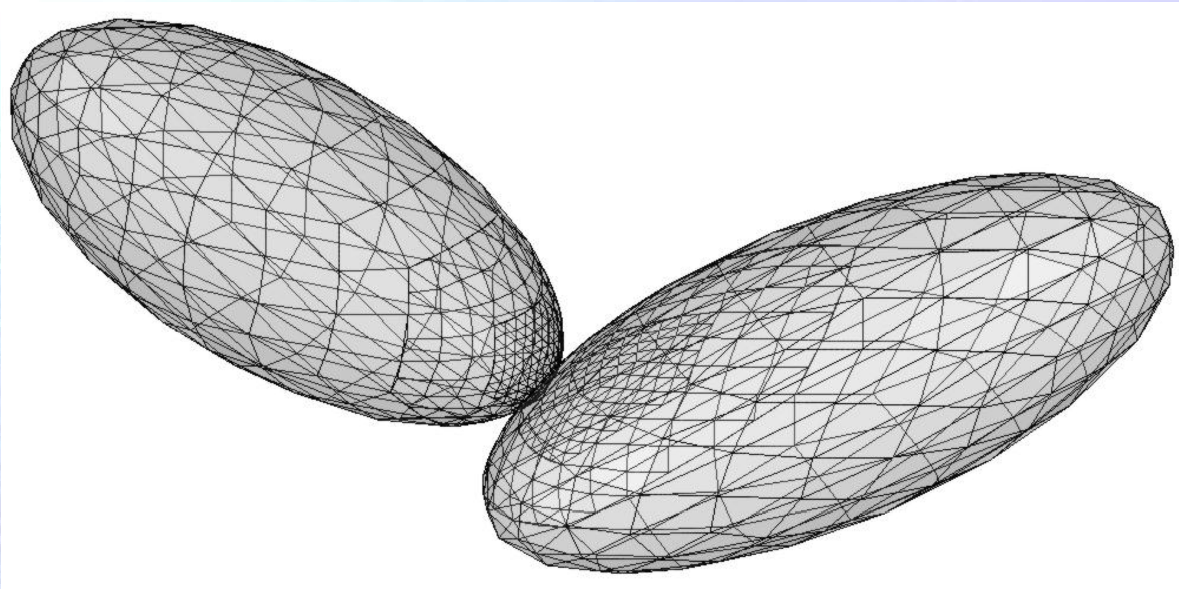
Numerical Methods

Paramecium: Force-free, Torque-free

Flow Field: Boundary Element Method

Ishikawa *et al.*, *J. Fluid Mech.* (2006)

$$u_i(\mathbf{x}) - \langle u_i(\mathbf{x}) \rangle = -\frac{1}{8\pi\mu} \sum_{\alpha=1}^{N} \int_{A_\alpha} J_{ij}(\mathbf{x}-\mathbf{y}) q_j(\mathbf{y}) dA_y$$



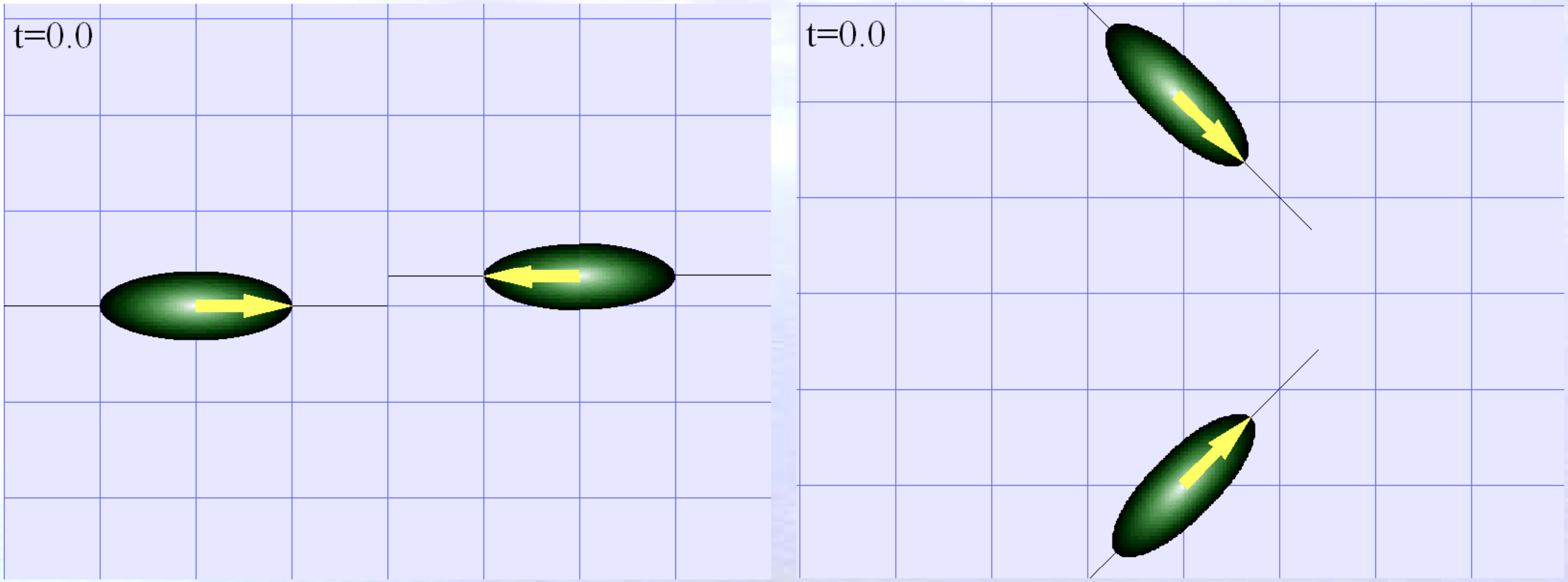
q : single-layer potential

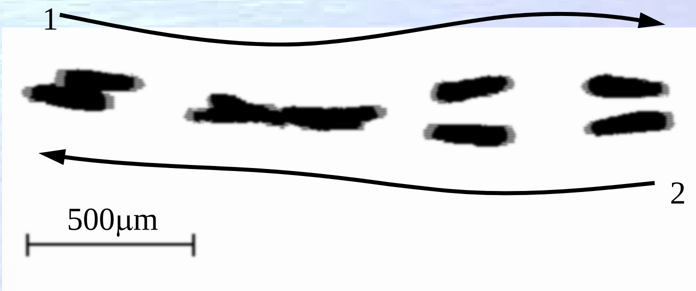
A : surface of a particle

u : velocity

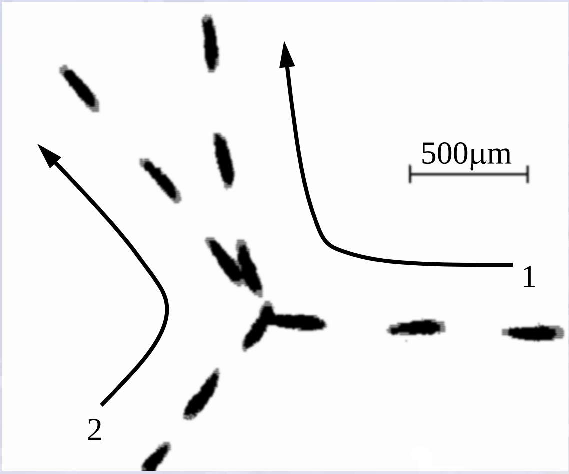
J : Green function

Numerical Results





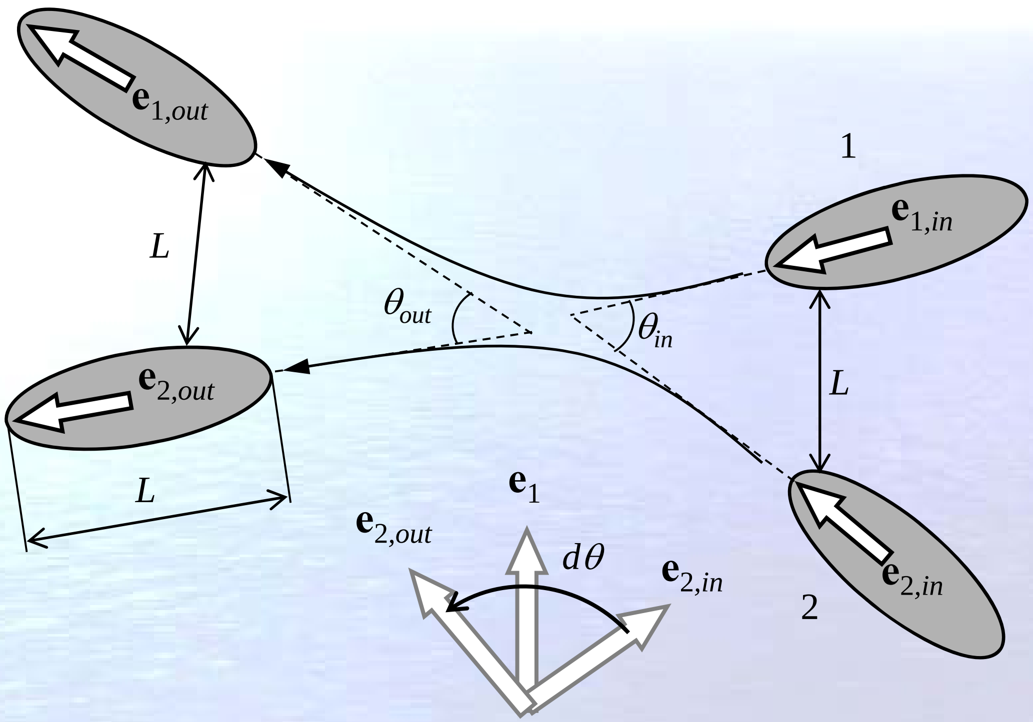
Agree with the experiment



Ishikawa and Hota, *J. Exp. Biol.* (2006)

Definition of Collisions

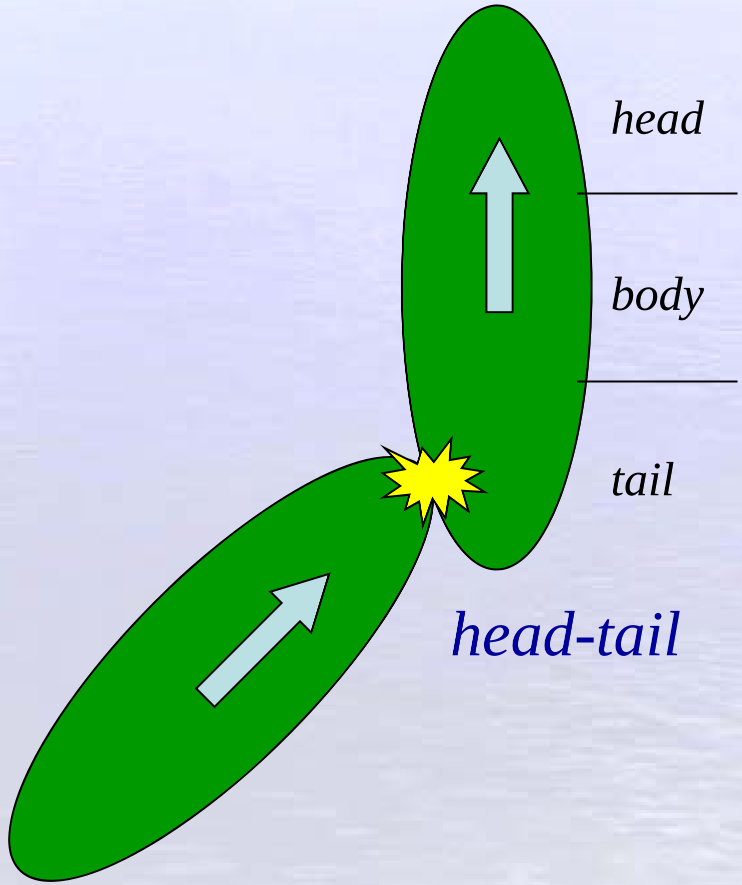
Definition of angles



θ_{in} : angle between $\mathbf{e}_{1,in}$ and $\mathbf{e}_{2,in}$

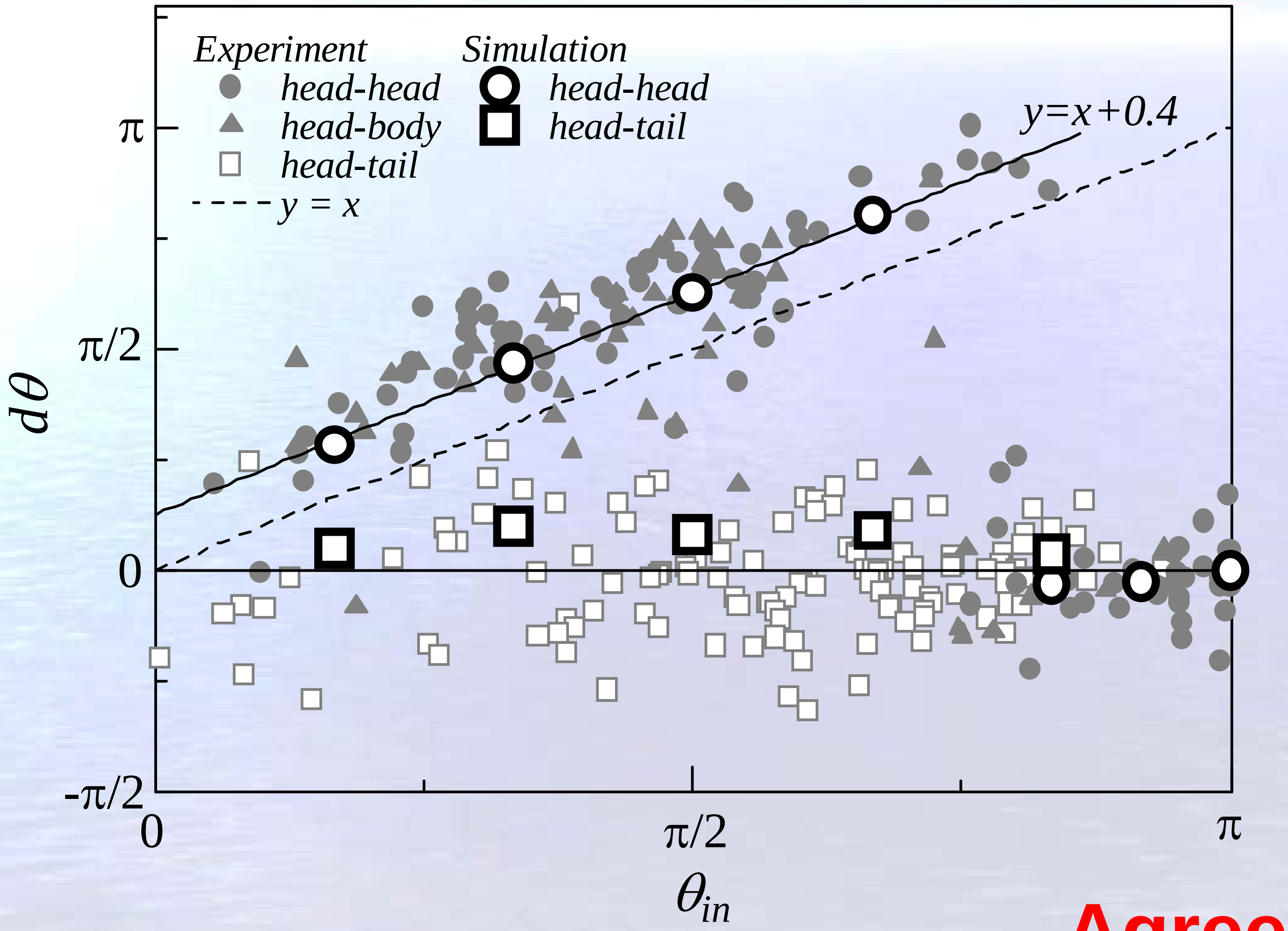
$d\theta$: change in the orientation vector of cell 2 relative to cell 1

Contact position



Comparison : Exp. & Sim.

Correlation between θ_{in} and $d\theta$



Ishikawa and Hota, *J. Exp. Biol.* (2006)

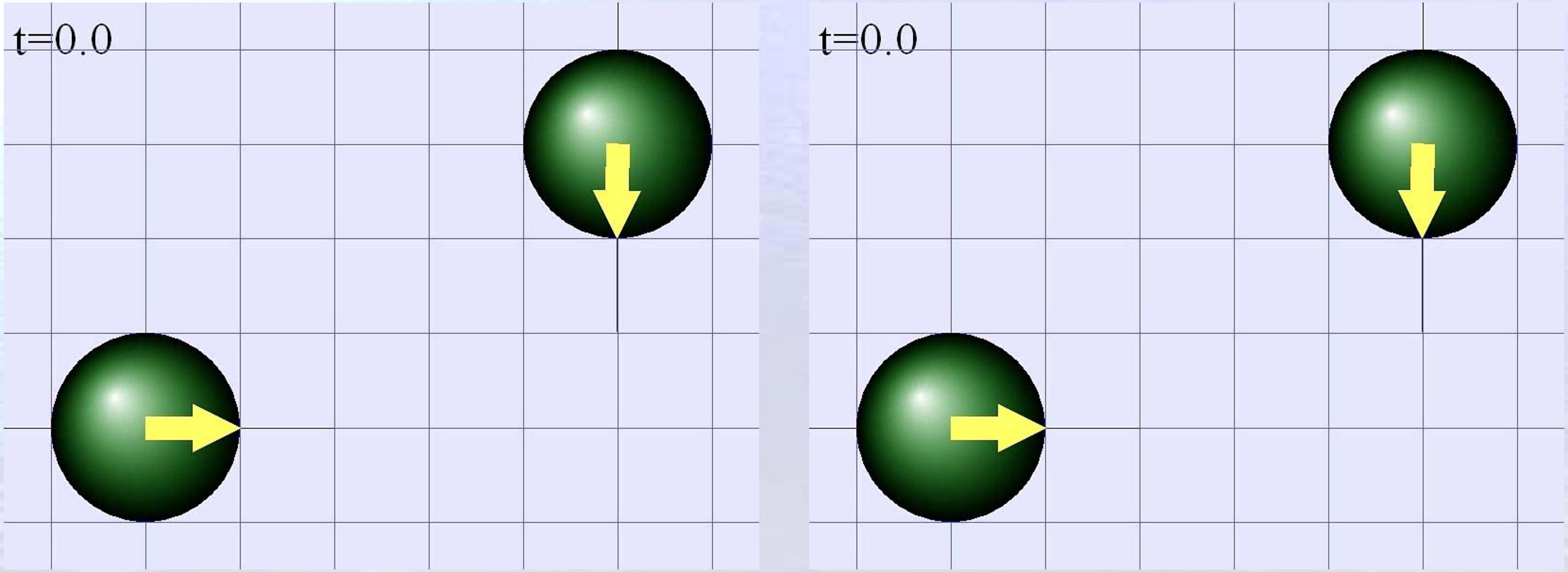
Agree well

Unsteady Squirmers

Paramecium is not a steady squirmer.

Spherical unsteady squirmer model: Blake (1971), Magar & Pedley (2005)

Surface tangential displacements: $$\theta = \theta_0 + \varepsilon \sum_{n=1}^{\infty} \beta_n(t) V_n(\cos\theta_0)$$



Giacche & Ishikawa, *J. Theor. Biol.* (2010)

Unsteady Squirmers

Shear stress on the surface of a squirmer

Intensity of the stimulus

⇩

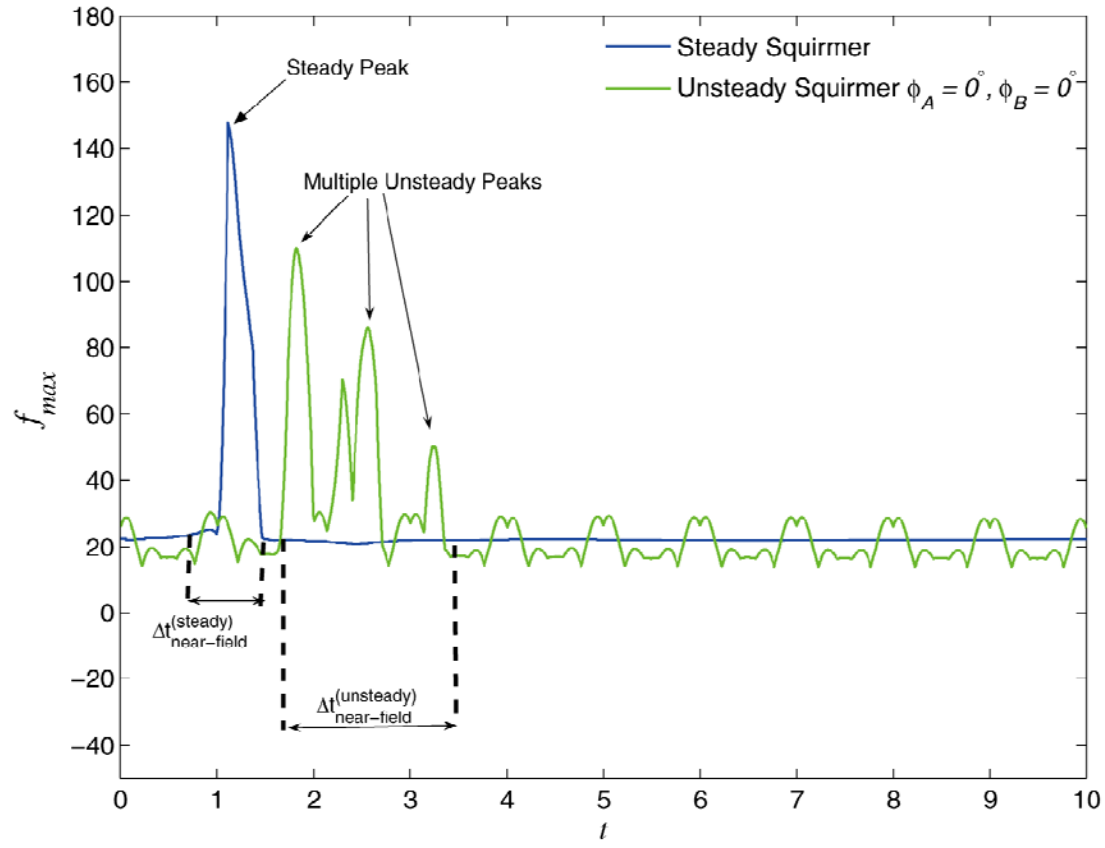
Intensity of the calcium influx

⇩

Determines the membrane depolarization

⇩

Consequently response of the ciliary apparatus

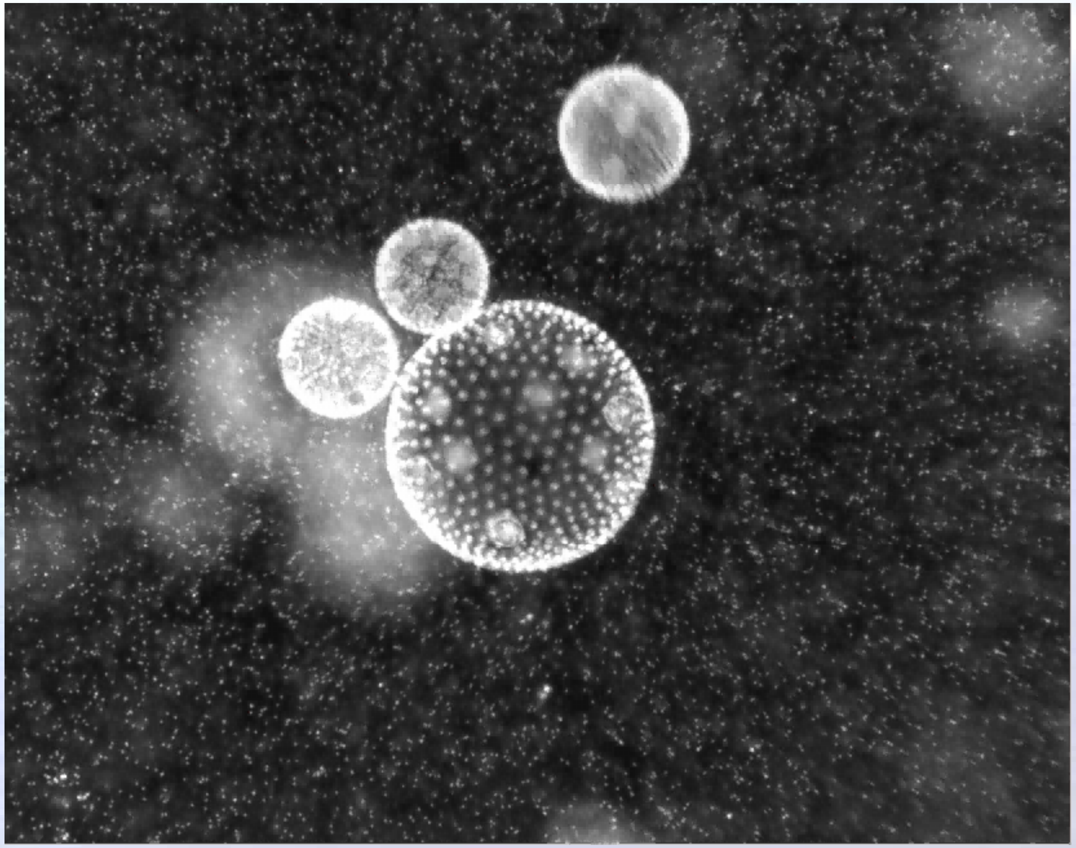


Unsteadiness adds hydrodynamic nature of randomness in biological reactions

Giacche & Ishikawa, *J. Theor. Biol.* (2010)

Waltzing motion of *Volvox*

A waltzing motion was found by R.E.Goldstein's group.



for fertilization?

http://www.damtp.cam.ac.uk/user/gold/

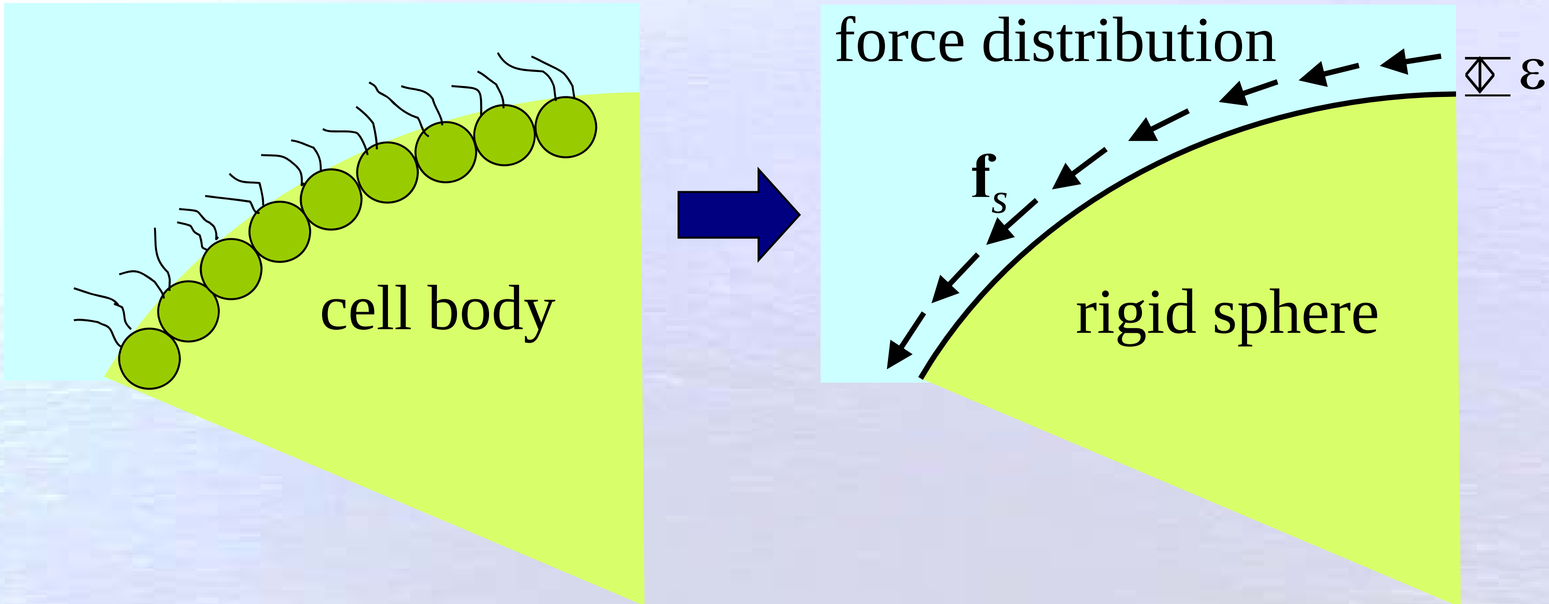
Mechanism: Biological? Hydrodynamical?

How to model Volvox?

Two important boundary conditions

- Force distribution generated by flagella
- No-slip on the cell body

ε is set as $0.1a$



Volvox is assumed as a rigid sphere, and force distribution $\mathbf{f}_s$ is generated ε above the spherical surface

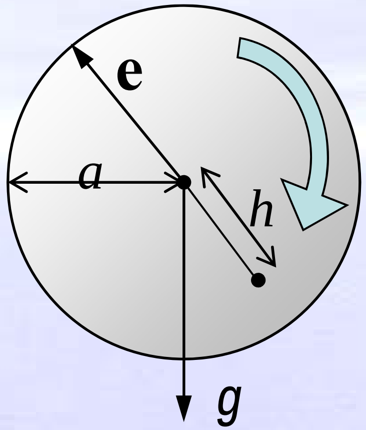
$\mathbf{f}_s$ is the force vector per unit area due to the flagella motion

Governing equations

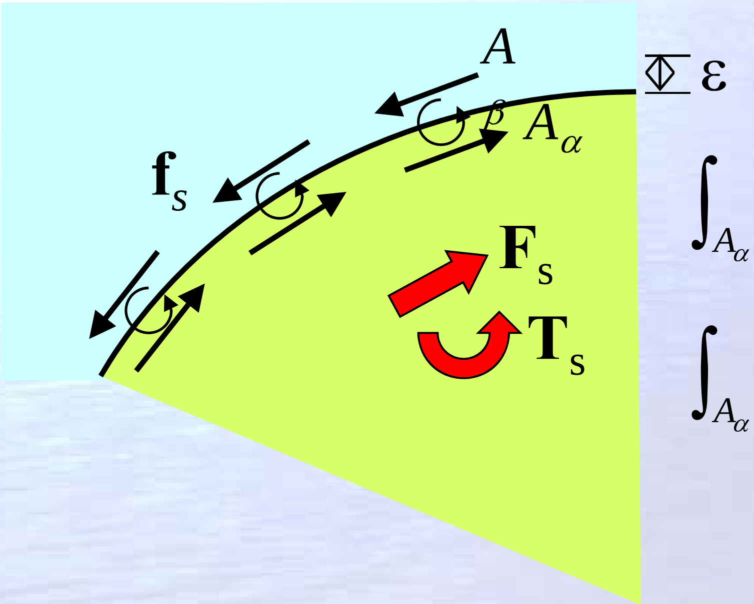
Flow Field : Stokes Flow

Cell Body Motion : Force *not free*

Torque *not free*



bottom-heaviness



$$\int_{A_\alpha} \boldsymbol{\sigma}\cdot\mathbf{n}\,dA = -\mathbf{F}_s \left(= \int_{A_\beta} \mathbf{f}_s dA \right)$$

$$\int_{A_\alpha} (\boldsymbol{\sigma}\cdot\mathbf{n})\wedge\mathbf{r}\,dA = -\mathbf{T}_s \left(= \int_{A_\beta} \mathbf{f}_s \wedge (1+\varepsilon)\mathbf{n}\,dA \right)$$

$\boldsymbol{\sigma}$: stress tensor, $\mathbf{n}$: normal vector

A_α : surface of the cell body

A_β : surface where the force is generated

Reaction force ⇨ Thrust force & torque

Force distribution

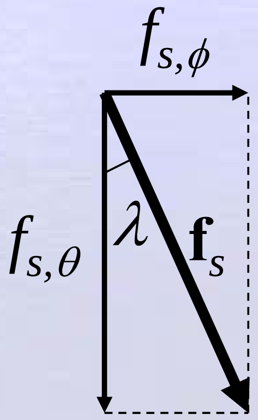
Force distribution : time-invariant, with swirl

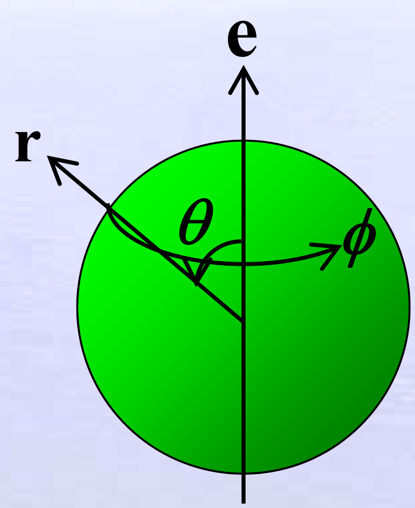
$$f_{s,r} = 0$$

$$f_{s,\theta} = const$$

$$f_{s,\phi} = f_{s,\theta} \tan \lambda$$

$(\mathbf{F}_s = \mathrm{const},\ \mathbf{T}_s = \mathrm{const})$





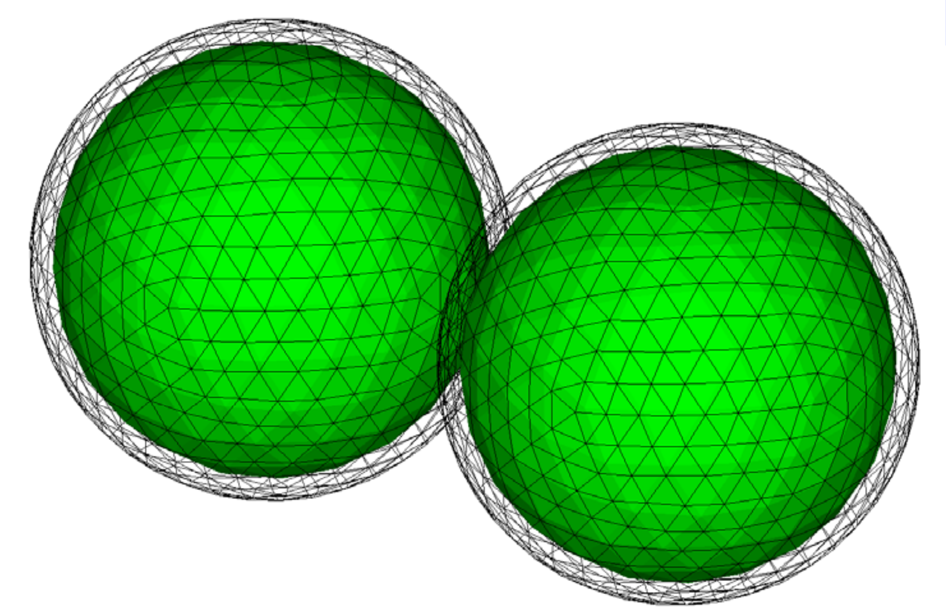
λ : tilt angle

When λ = 5deg, U = 1 and Ω =-1.8

Numerical Methods

Boundary Element Method

$$u_i(\mathbf{x}) - \langle u_i(\mathbf{x}) \rangle = -\frac{1}{8\pi\mu}\left[\sum_{\alpha=1}^{N}\int_{A_\alpha} J_{ij}(\mathbf{x}-\mathbf{y})t_j(\mathbf{y})dA_y - \sum_{\beta=1}^{N}\int_{A_\beta} J_{ij}(\mathbf{x}-\mathbf{y})f_j(\mathbf{y})dA_y\right]$$



Computational mesh

u : velocity
<**u**> : background velocity
t : traction force on the cell body
f : force due to the flagella motion
A_α : surface of the cell body
A_β : surface where the force is generated

J : Half-space Green function

When there is a wall, the kernel function need to be modified

Blake (1971)

Numerical set-up

Initial condition

6 meridians are drawn for reference

Volvox parameters

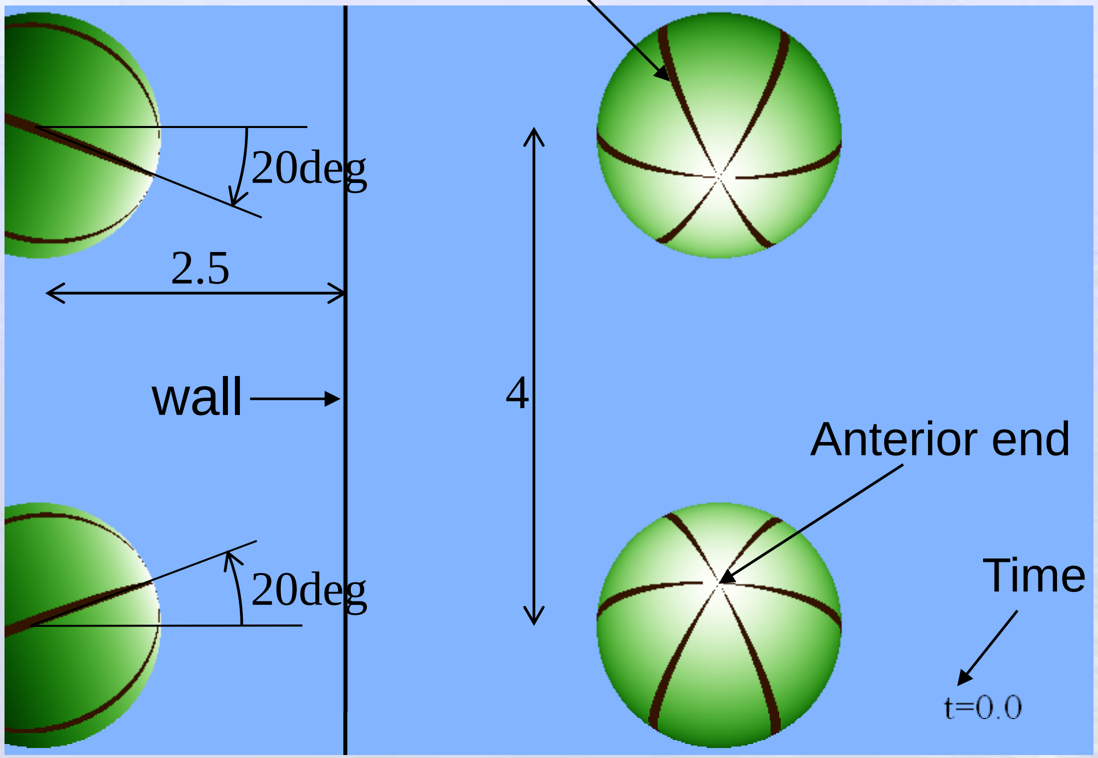
cilia length : $\varepsilon = 0.1$

tilt angle :

$\lambda = 0, 5, 10\text{deg}$

Bottom-heaviness :

$$G_{bh} = \frac{4\pi\rho g a^2}{3\mu U} = 0, 10, 50$$

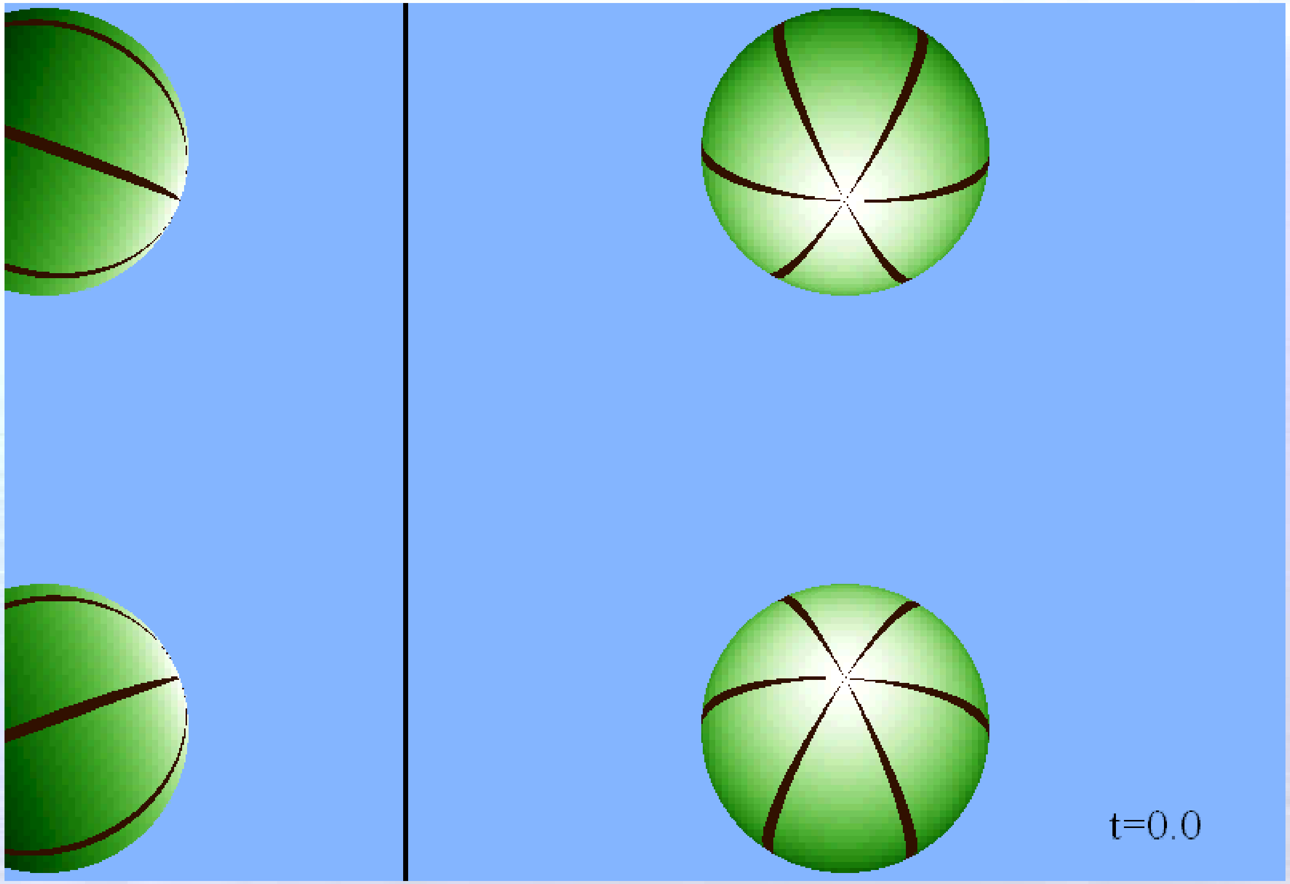


Side view

Top view

Resluts : effect of Bottom-heaviness

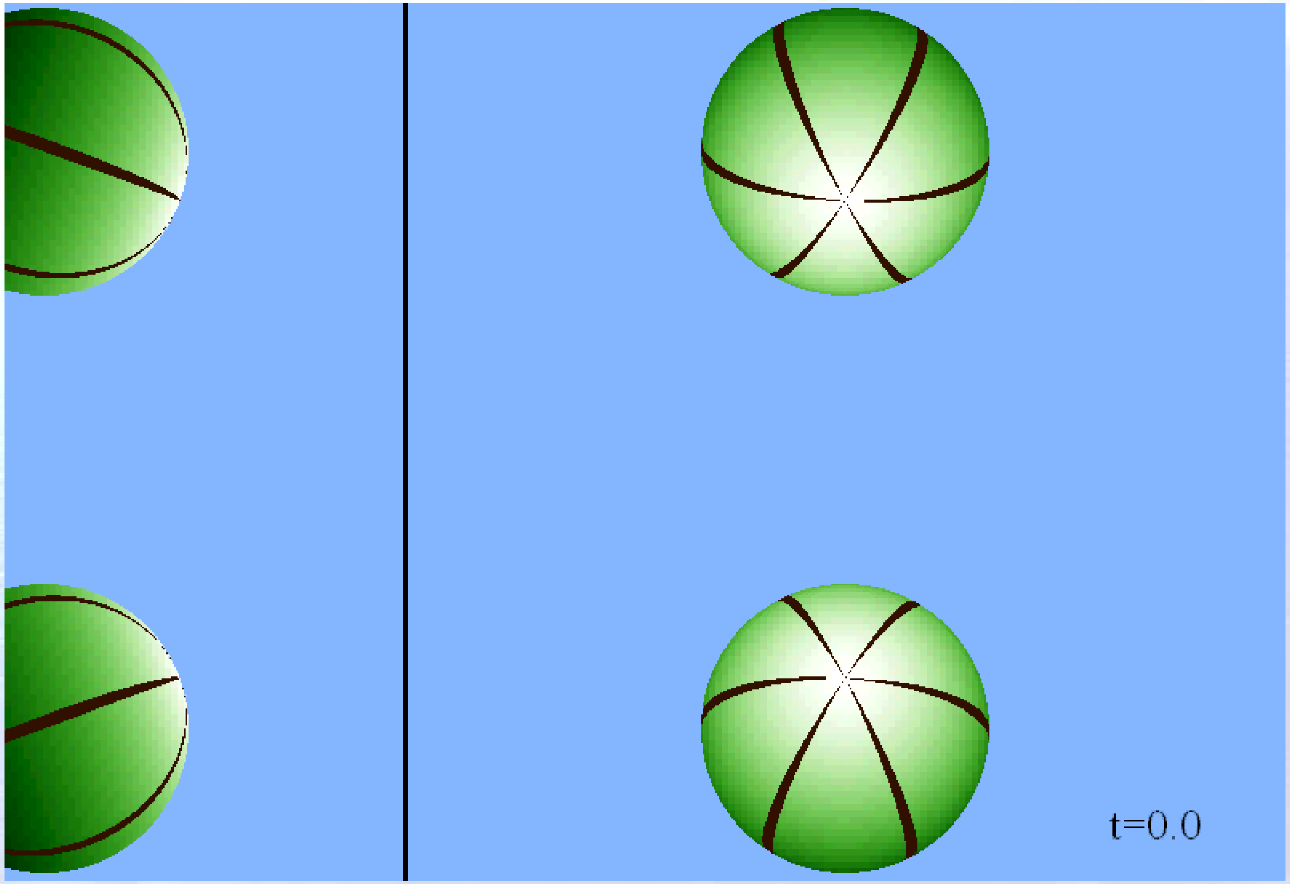
G_{bh}=0 : non-bottom-heavy (λ = 5deg)



Waltzing motion does not appear

Resluts : effect of Bottom-heaviness

G_{bh}=50 : bottom-heavy $(\lambda = 5\text{deg})$



Waltzing motion does appear

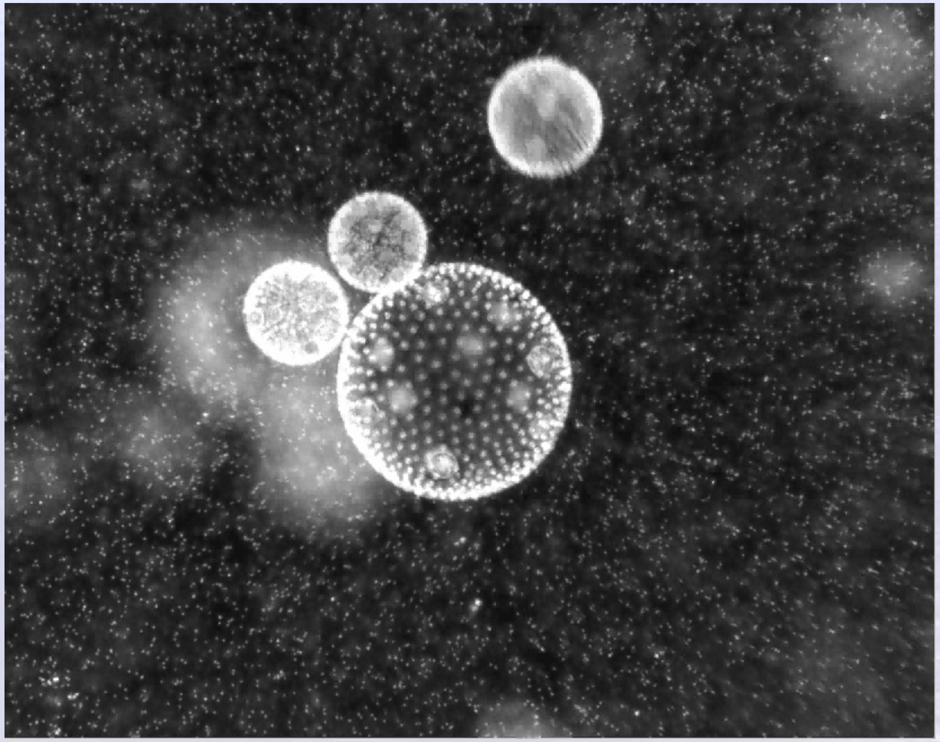
Waltzing motion of *Volvox*

The waltzing motion can be reproduced by introducing:

(a) A wall boundary

(b) Bottom-heaviness

(c) Swirl velocity

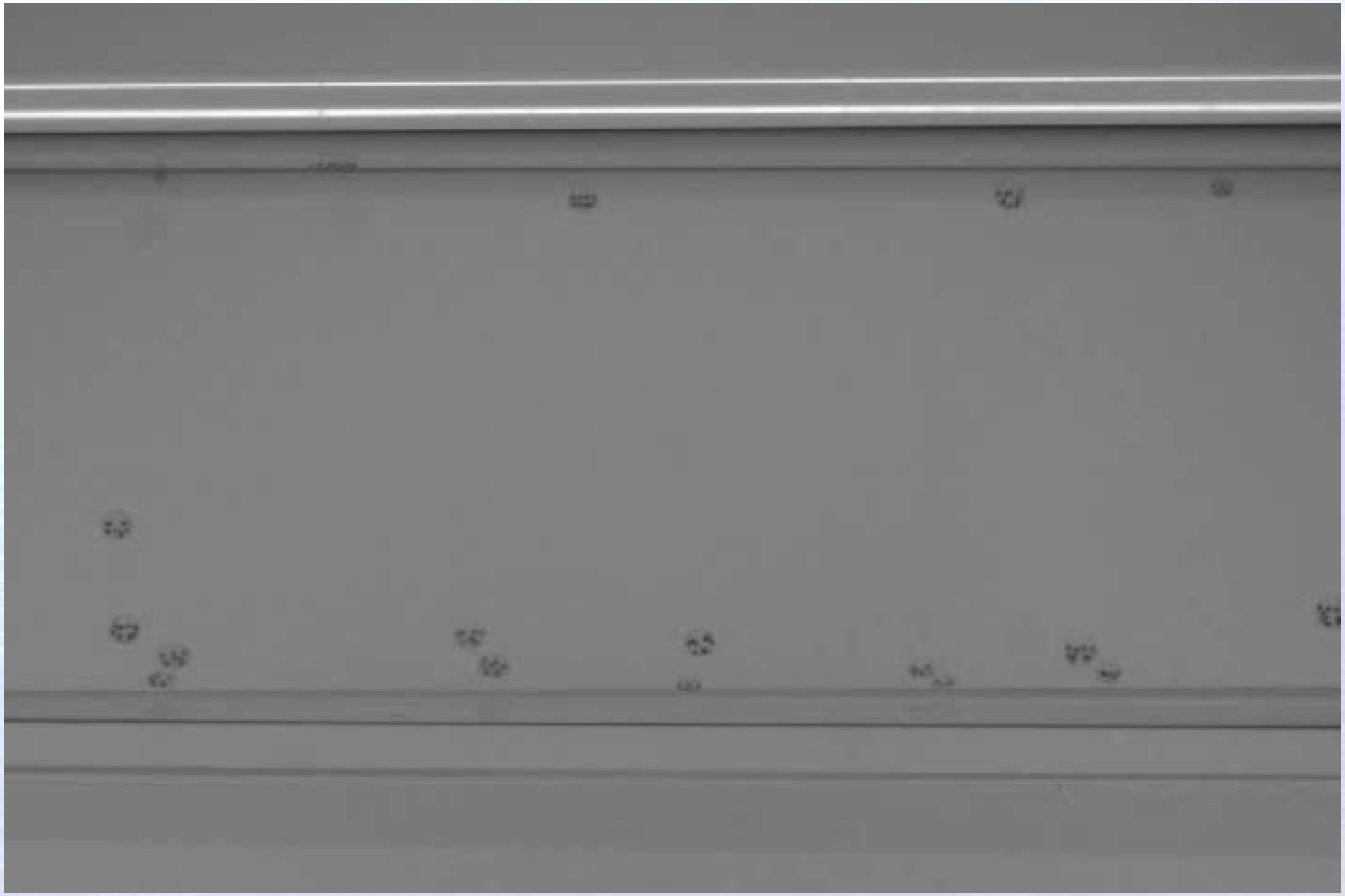


Mechanism
= Hydrodynamics

Drescher *et al*., *Phys. Rev. Lett.* (2009)

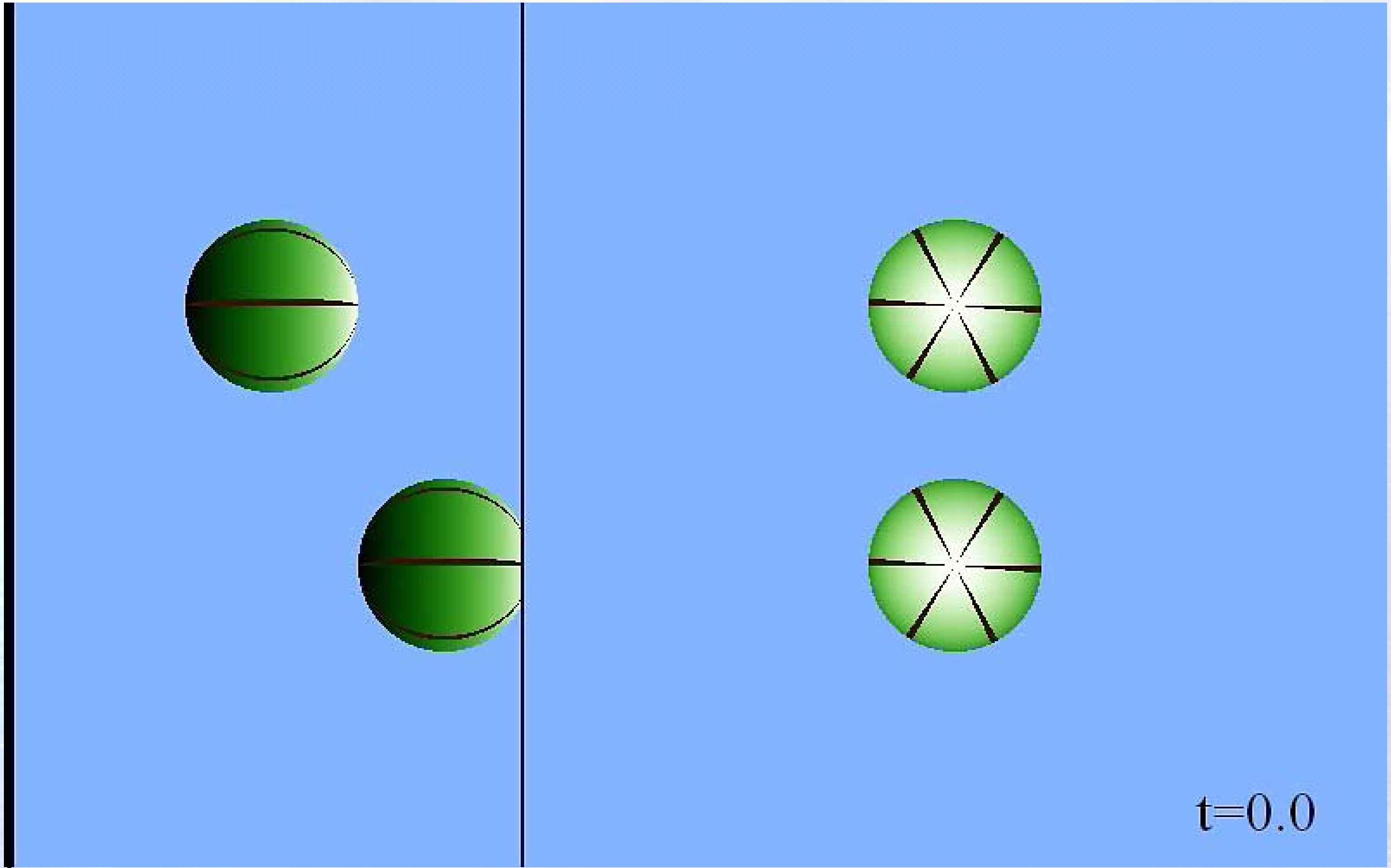
Another interaction of *Volvox*

Another interaction, referred to as minuet, was found.



How about this?

Numerical Results : another bound state



Bottom wall

Side view

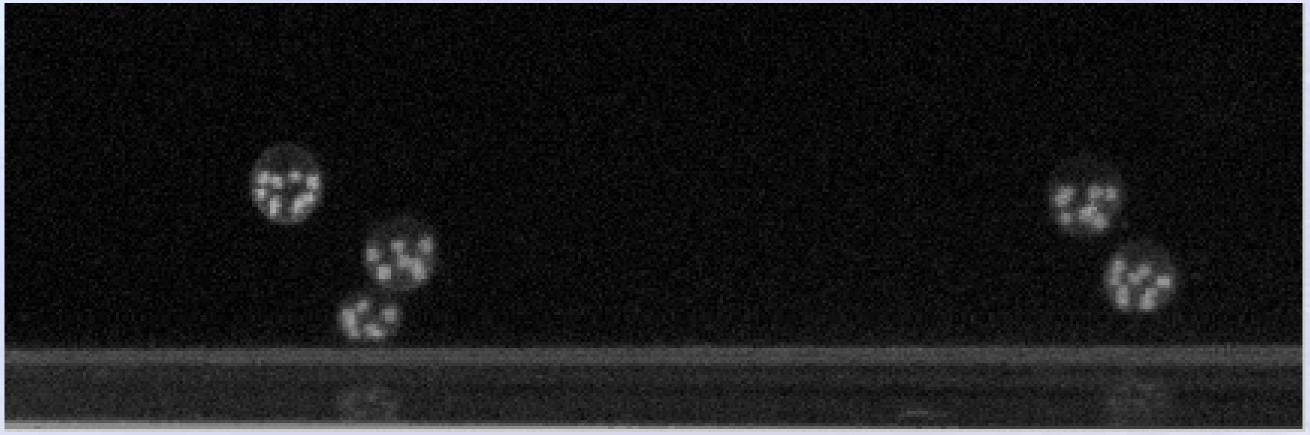
Top view

Another bound state does appear (G_{bh} = 3)

Another dancing of *Volvox*

Minuet bound state requires:

(a) Sedimentation
(b) Bottom wall
(c) Bottom-heaviness
(d) Swirl

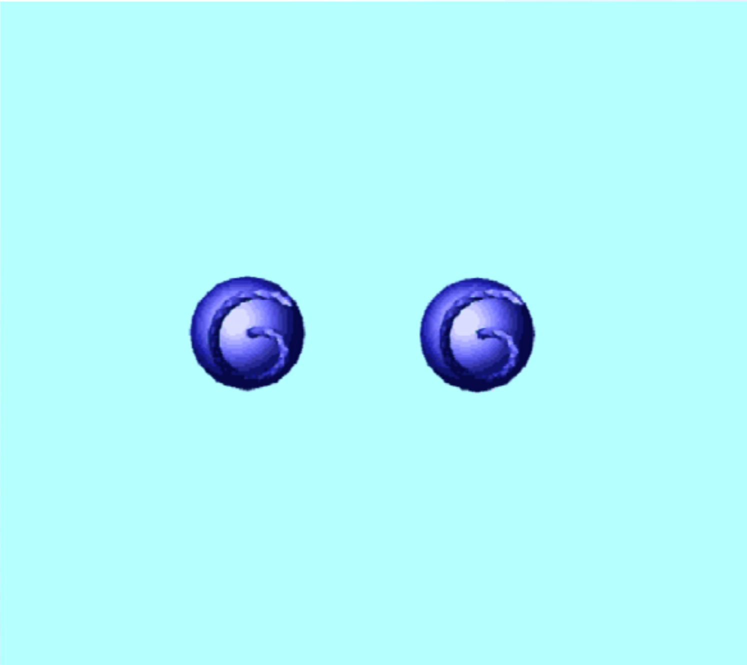


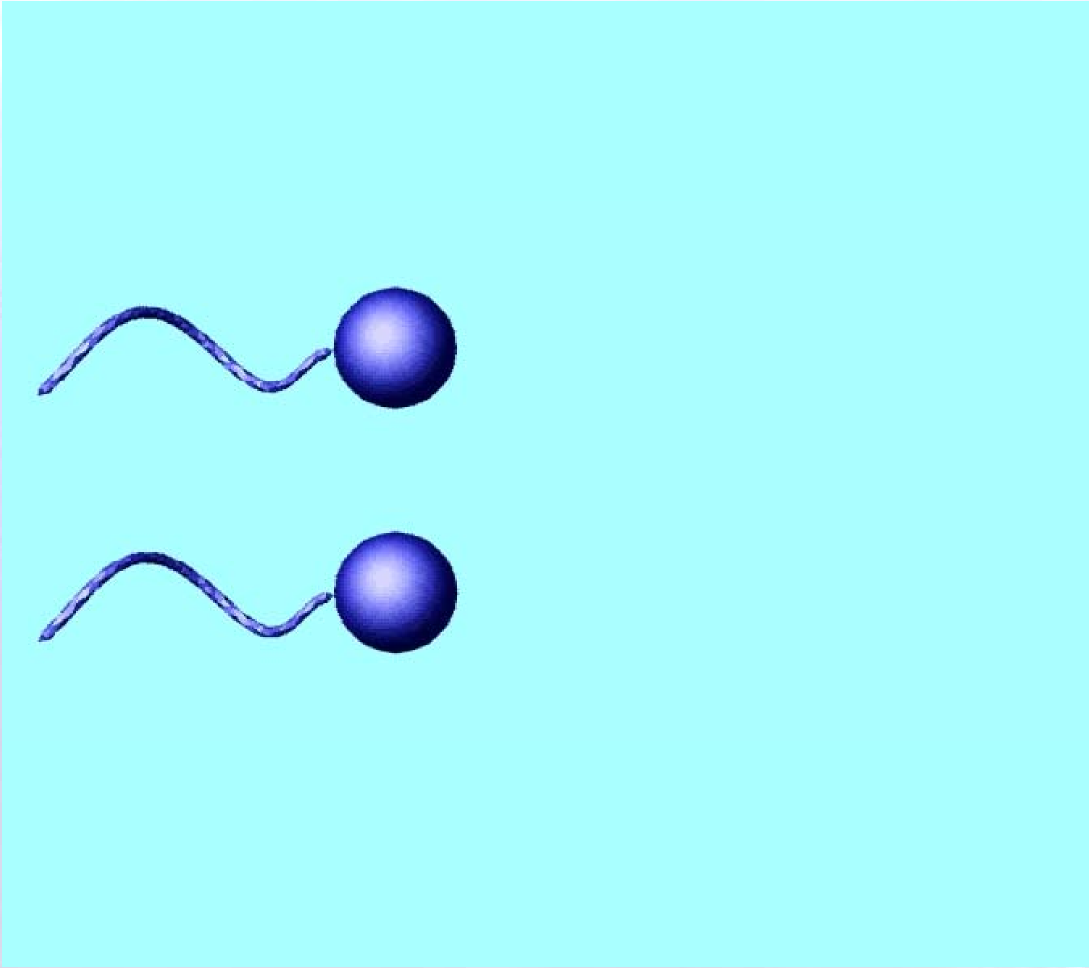
Mechanism is again hydrodynamics.

Drescher *et al.*, *Phys. Rev. Lett.* (2009)

Bacterial swimming

Two Bacteria Interaction



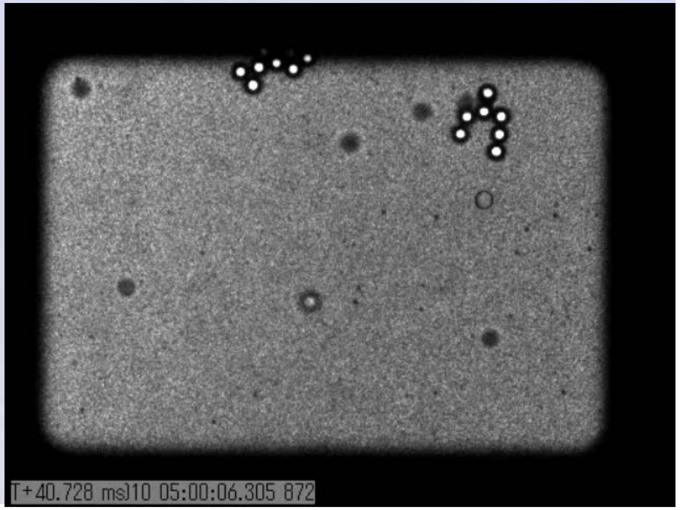


Ishikawa *et al*., *Biophys. J.* (2007)

Bacterial interaction can also be analyzed.

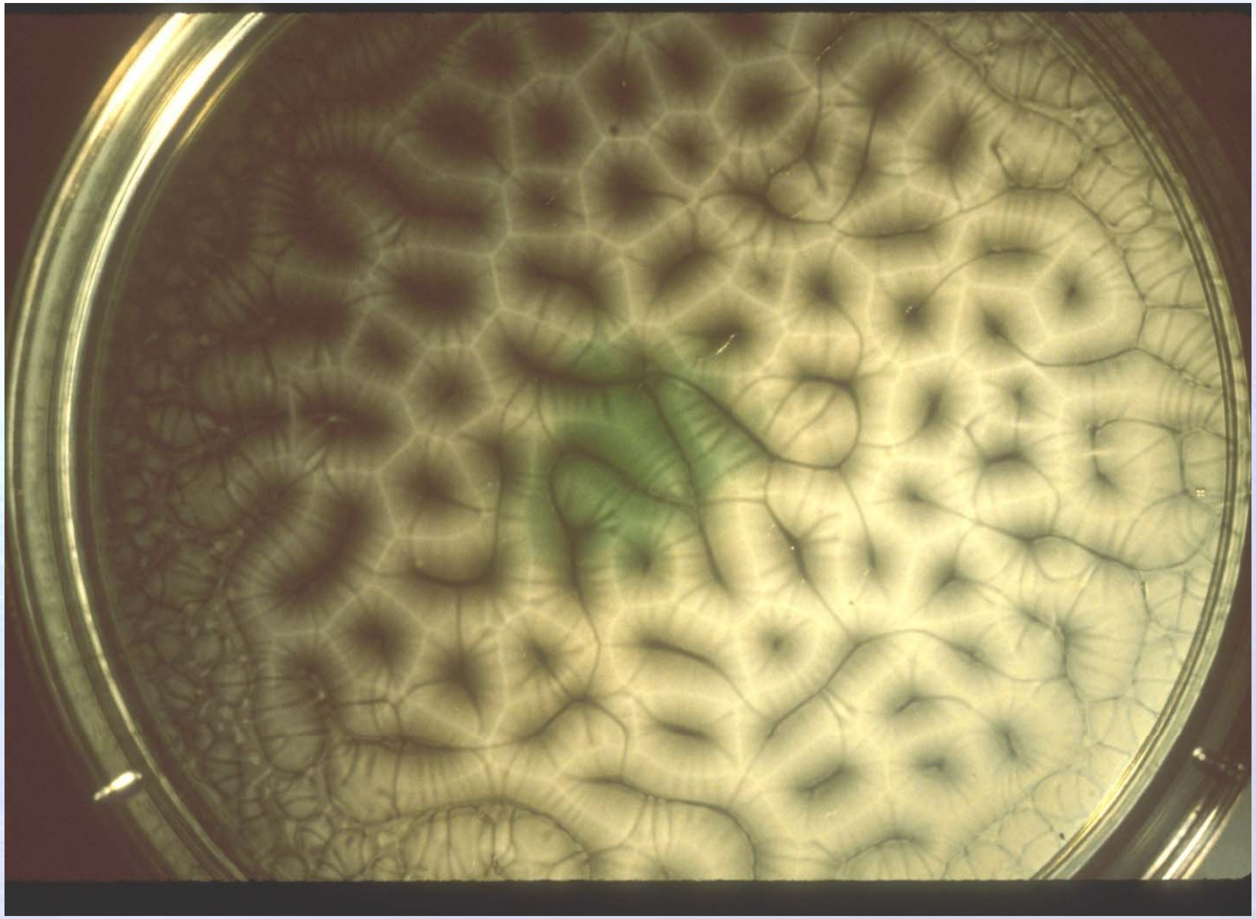
Outline of the talk

- Introduction
- Biomechanics of an **individual** and a **pair** of micro-organisms
- **Collective swimming** in meso-scale
- **Macroscopic properties** of a suspension of micro-organisms
- Conclusions



Bio-convection

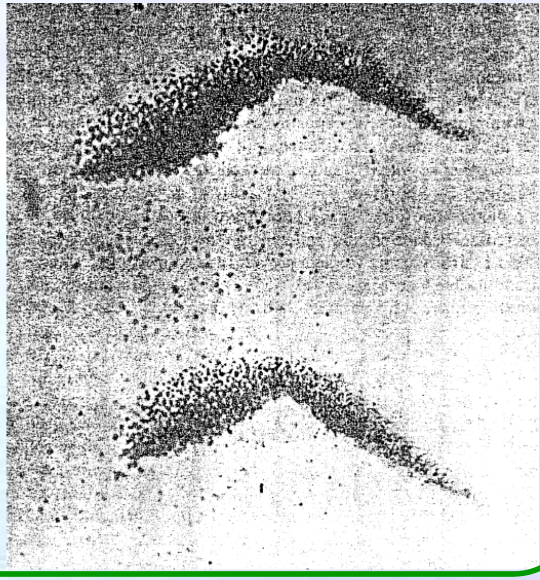
Suspensions of *B. subtilis* : Pedley & Kessler (1992)



Mechanism : upswimming of cells that are slightly denser than water generates unstable density stratification which leads to overturning

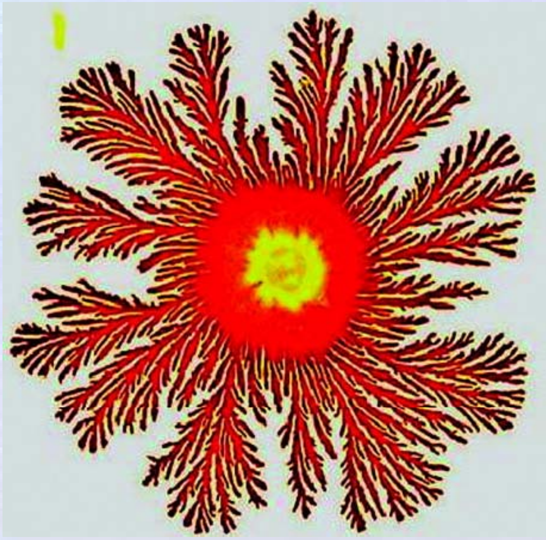
Other collective motions

Band formation



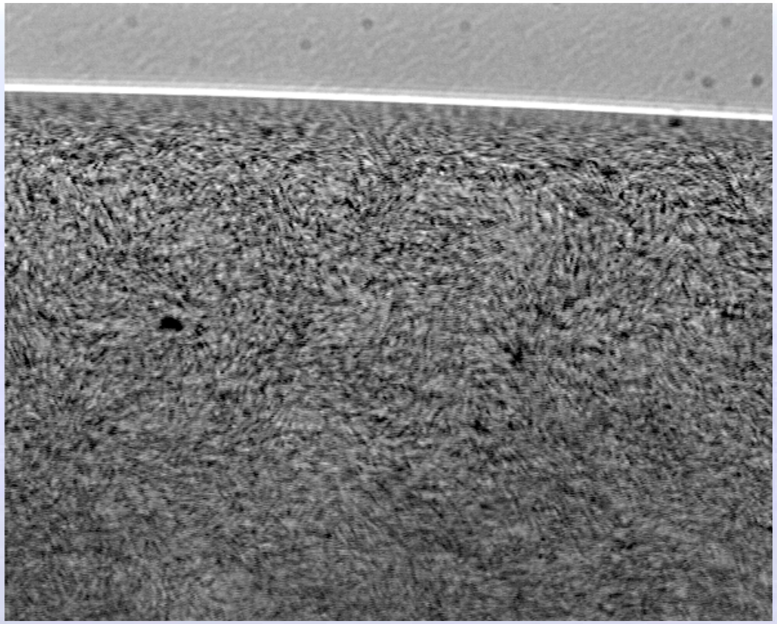
Band formation of magnetotactic bacteria. Picture from Guell *et al.*, *J. Theor. Biol.* (1988)

Colonies on agar gel



Complex patten of bacterial colonies. Picture from Ben-Jacob & Levine (2006)

Slow turbulence



Cell motions of *Bacillus subtilis*. Movie from Goldstein Lab, University of Cambridge

Mechanism
= Physics? Mechanics?

Mathematical Models

Continuum Model:

Pedley & Kessler (1992) : Boussinesq approximation

Simha & Ramaswamy (2002), etc. : A kinetic theory
Saintillan & Shelley (2008) : A kinetic theory
Aranson et al. (2005), etc. : A two-dimensional master equation
Wolgemuth (2008) : A two-phase model

Discrete Model:

Vicsek et al. (1995), Czirok et al. (1997), Gregoire & Chate (2004), Sambelashvili et al. (2007) : The hydrodynamics were not incorporated

Hopkins & Fauci (2002), Llopis & Pagonabarraga (2006), Hernandes-Ortiz et al. (2005), Underhill et al. (2008), Saintillan & Shelley (2007): Far-field hydrodynamics were incorporated

Ishikawa et al. (2007), etc. : Far- and Near-field hydrodynamics were incorporated precisely.

Stokesian-dynamics simulation

Micro-organism : Spherical squirmer model

Multipole Expansion of the boundary integral equation

$$u_i(\mathbf{x})-\langle u_i(\mathbf{x})\rangle = -\frac{1}{8\pi\mu}\sum_{\alpha=1}^{N}\int_{A_\alpha} J_{ij}(\mathbf{x}-\mathbf{y})q_j(\mathbf{y})dA_y \quad : \textit{Ewald sumation}$$

$$=\frac{1}{8\pi\mu}\left[\left(1+\frac{a^2}{6}\nabla^2\right)J_{ij}F_j^\alpha + R_{ij}L_j^\alpha + \left(1+\frac{a^2}{10}\nabla^2\right)K_{ijk}S_{jk}^\alpha + \nabla_k\nabla_l J_{ij}Q_{klj}^\alpha + \cdots\right]$$

+

Faxen Laws

$$U_i^\alpha - \langle u_i(\mathbf{x}^\alpha)\rangle = \frac{F_i^\alpha}{6\pi\mu a} + \frac{2}{3}B_1^\alpha e_i^\alpha + \left(1+\frac{a^2}{6}\nabla^2\right)u_i'(\mathbf{x}^\alpha)$$

$$\Omega_i^\alpha - \langle \omega_i(\mathbf{x}^\alpha)\rangle = \frac{L_i^\alpha}{8\pi\mu a^3} + \frac{1}{2}\varepsilon_{ijk}\nabla_j u_k'(\mathbf{x}^\alpha)$$

$$-\langle E_{ij}(\mathbf{x}^\alpha)\rangle = \frac{S_{ij}^\alpha}{\frac{20}{3}\pi\mu a^3} + \frac{1}{5}\mu a^2 B_2^\alpha\left(3e_i^\alpha e_j^\alpha - \delta_{ij}\right) + \frac{1}{2}\left(1+\frac{a^2}{10}\nabla^2\right)\left(\nabla_j u_i'(\mathbf{x}^\alpha) + \nabla_i u_j'(\mathbf{x}^\alpha)\right)$$

Ishikawa *et al.*, *J. Fluid Mech.* (2008)

Stokesian-dynamics simulation

Then, inclusion of near-field lubrication forces

$$\begin{pmatrix} \mathbf{F} \\ \mathbf{L} \\ \mathbf{S} \end{pmatrix} = \left[\mathbf{R}^{far} - \mathbf{R}_{2B}^{far} + \mathbf{R}_{2B}^{near} \right] \begin{pmatrix} \mathbf{U} - \langle \mathbf{u} \rangle \\ \mathbf{\Omega} - \langle \boldsymbol{\omega} \rangle \\ -\langle \mathbf{E} \rangle \end{pmatrix}$$

$$+ \left[\mathbf{R}^{far} - \mathbf{R}_{2B}^{far} \right] \begin{pmatrix} -\frac{2}{3} B_1 \mathbf{e} + \mathbf{Q}_{sq} \\ 0 \\ -\frac{1}{5} B_2 (3\mathbf{ee} - \mathbf{I}) \end{pmatrix} + \begin{pmatrix} \mathbf{F}_{sq}^{near} \\ \mathbf{L}_{sq}^{near} \\ \mathbf{S}_{sq}^{near} \end{pmatrix}$$

Database compiled by BEM

Effect of squirming motion

cf. Brady & Bossis, *Annu. Rev. Fluid Mech*. (1988)

For details : Ishikawa *et al.*, *J. Fluid Mech.* (2008)

Results: Aggregation

Monolayer

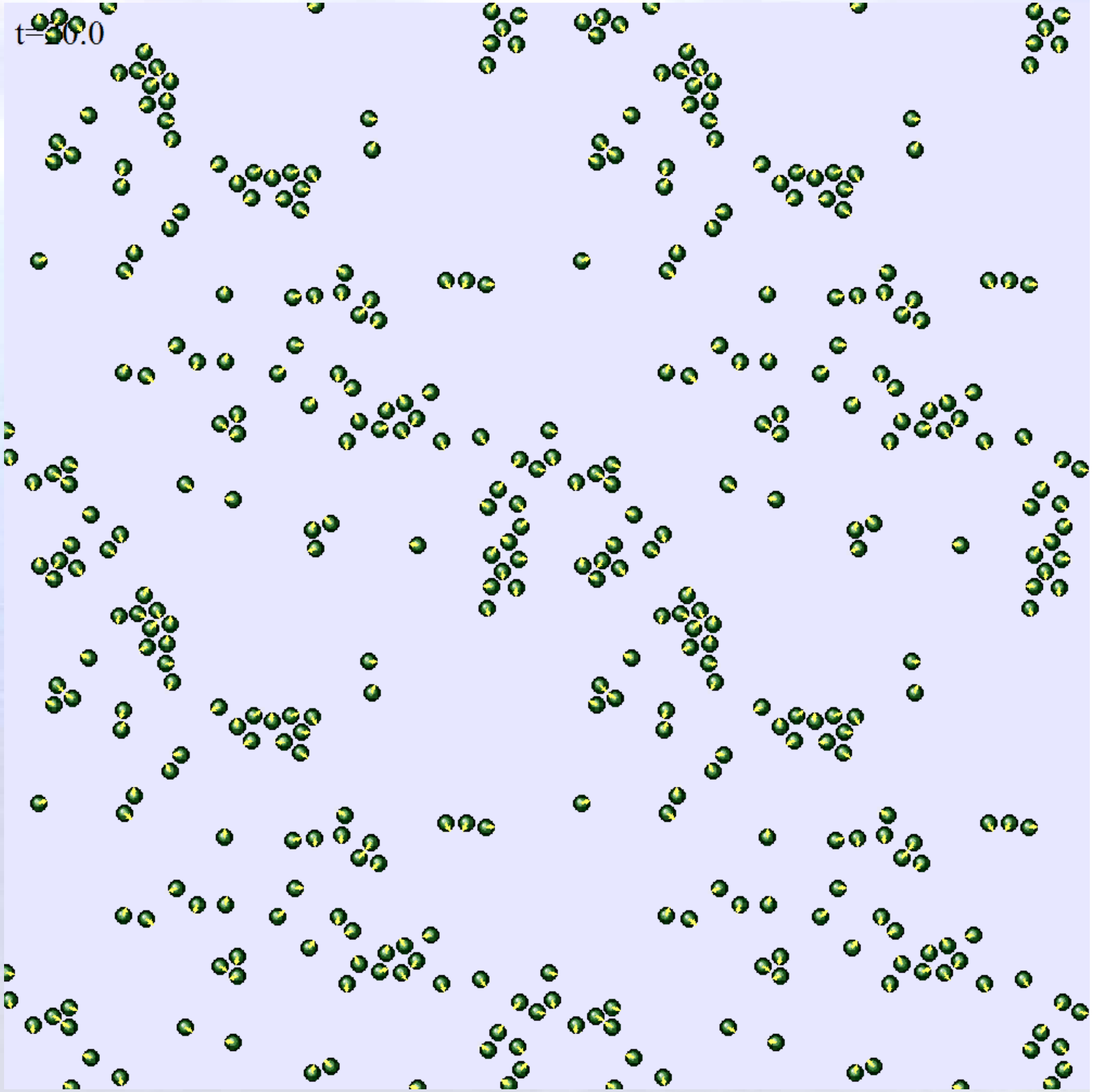
Non-bottom-heavy

ϕ_a=0.1

Periodic B.C.

Hydrodynamic interaction only

Ishikawa & Pedley
Phys. Rev. Lett. (2008)



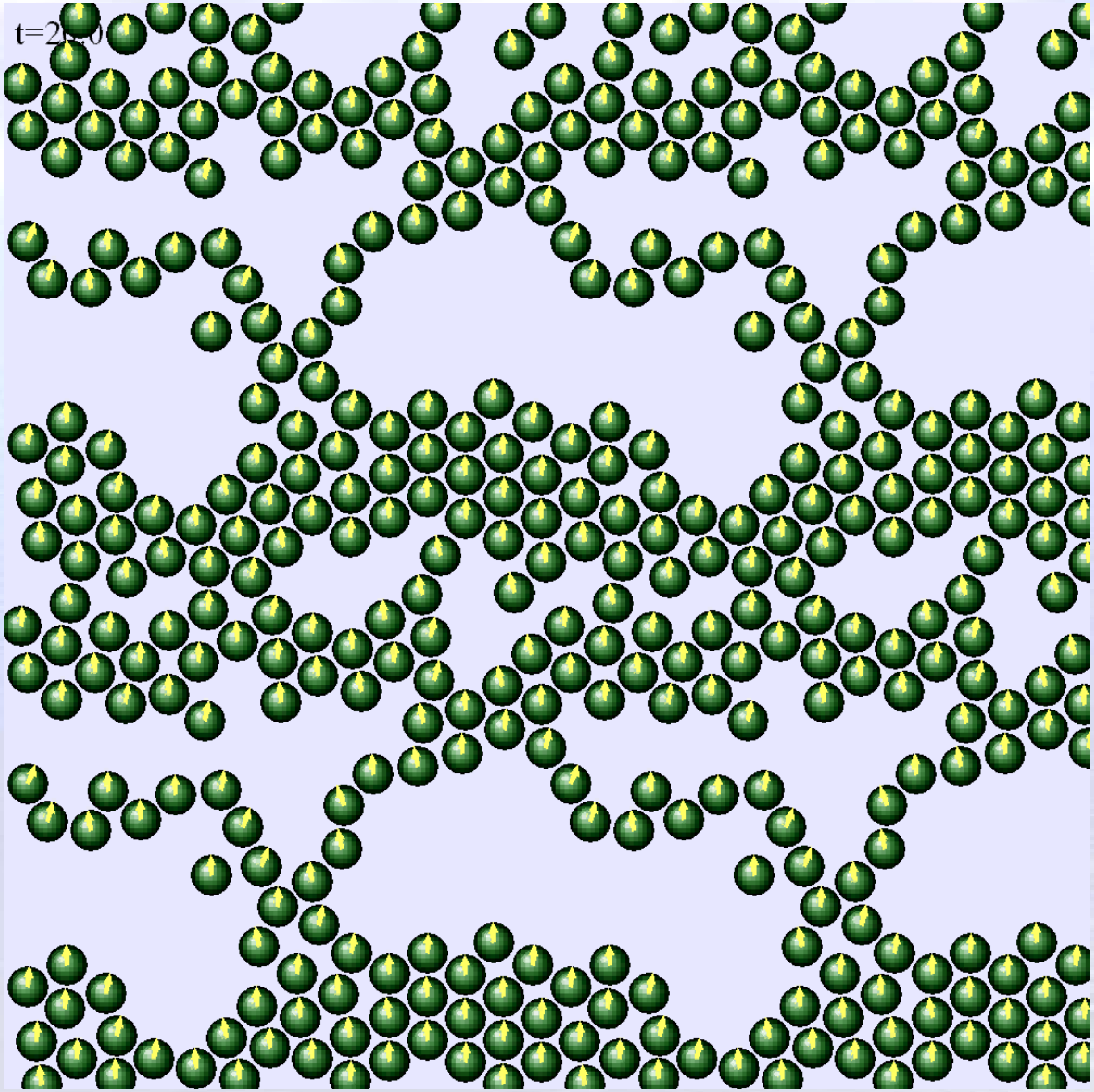
Results: Band formation

Monolayer

Bottom-heavy

ϕ_a=0.5, G_{bh} =100

$$G_{bh} = \frac{2\pi\rho g a h}{\mu B_1}$$



Ishikawa & Pedley
Phys. Rev. Lett. (2008)

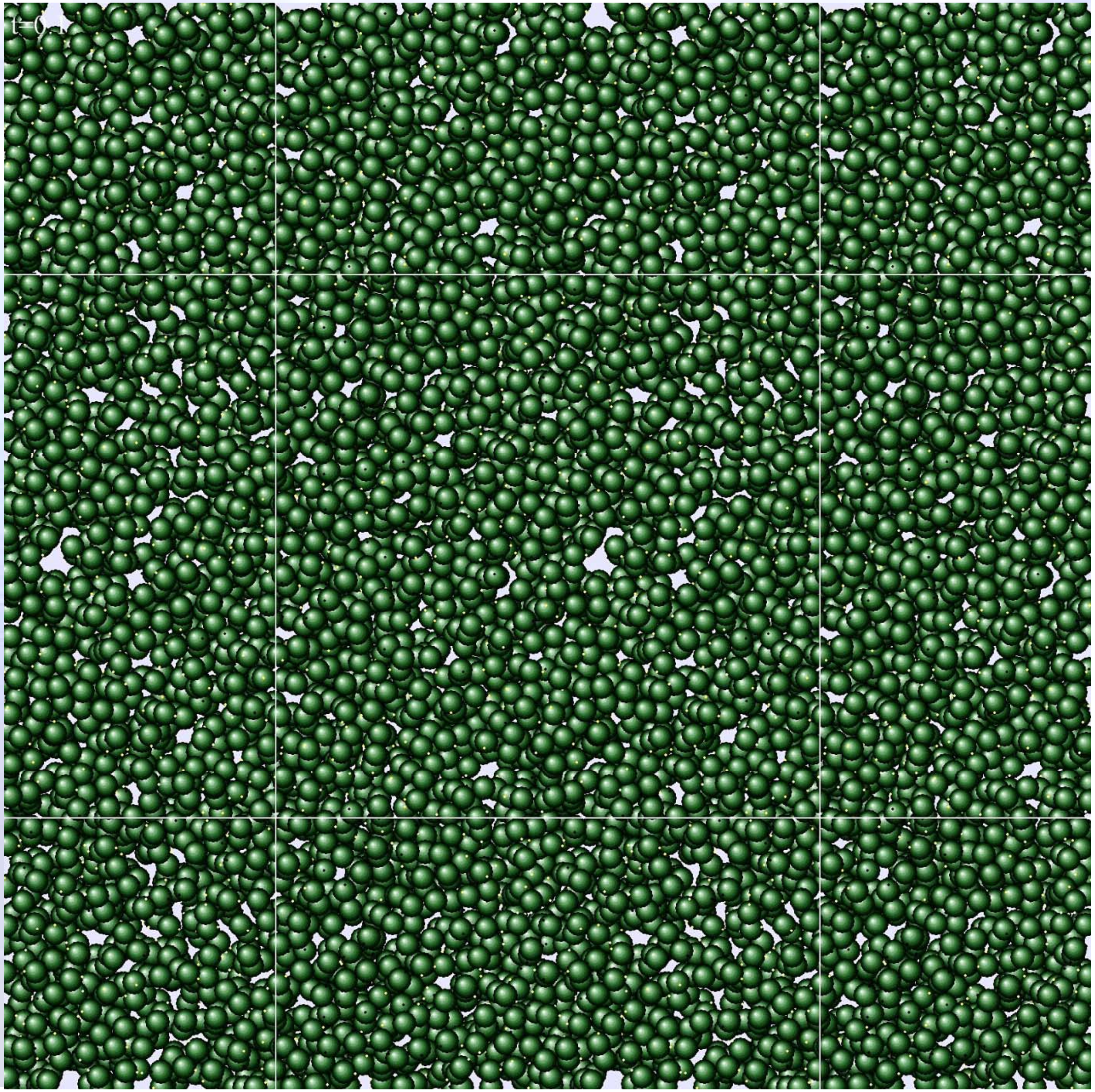
Results: 3D Large scale

3D isotropic suspension

2000 cells

Non-Bottom-Heavy

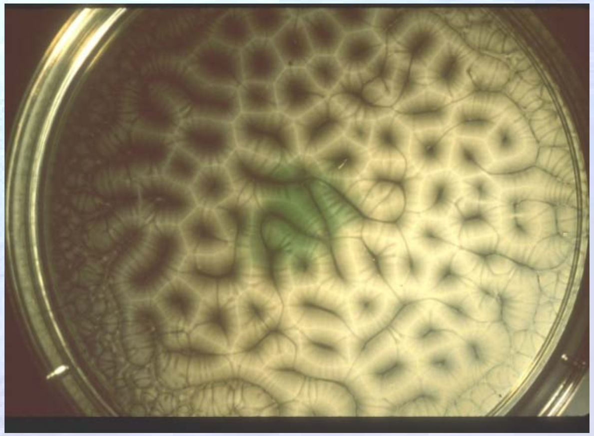
Periodic B.C.

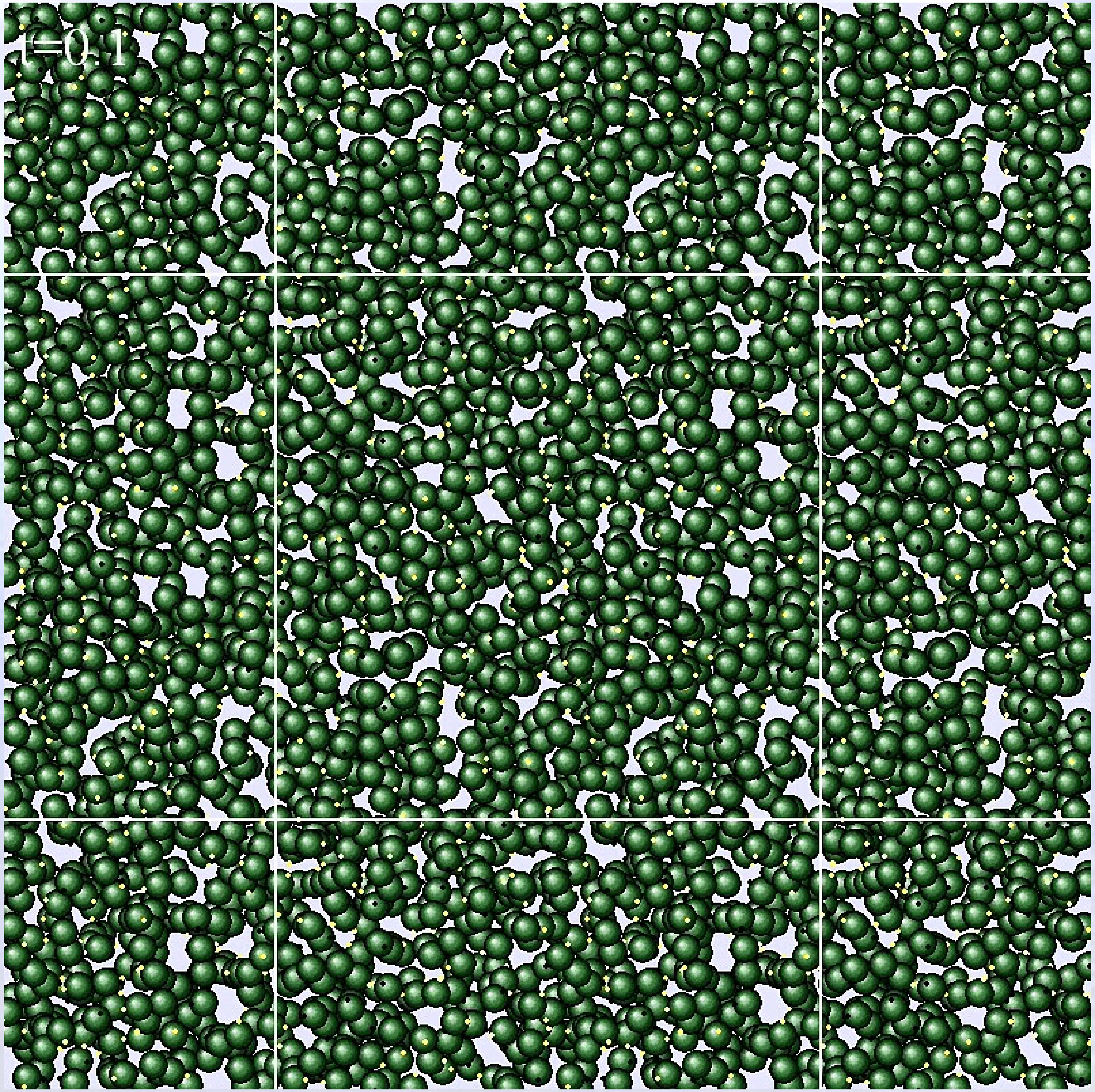


Results: 3D Large scale

Bioconvection

Bottom-Heavy
Sedimentation
Periodic B.C.

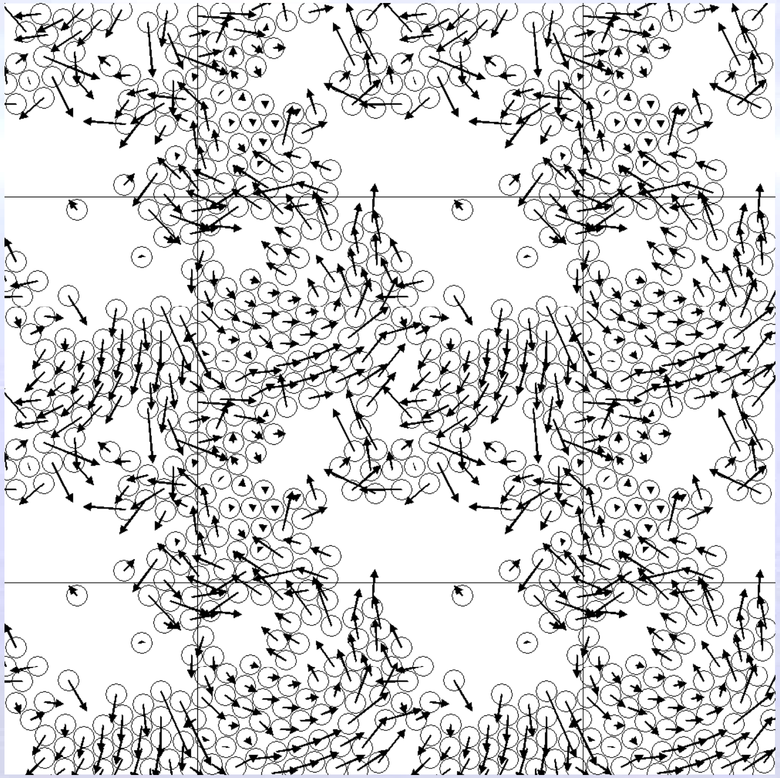




Coherent structures

Various collective motions observed in former experiments can be expressed

- Meso-scale spatiotemporal motion
- Ordered motion



<u>How the coherent structre affect transport phenomena?</u>

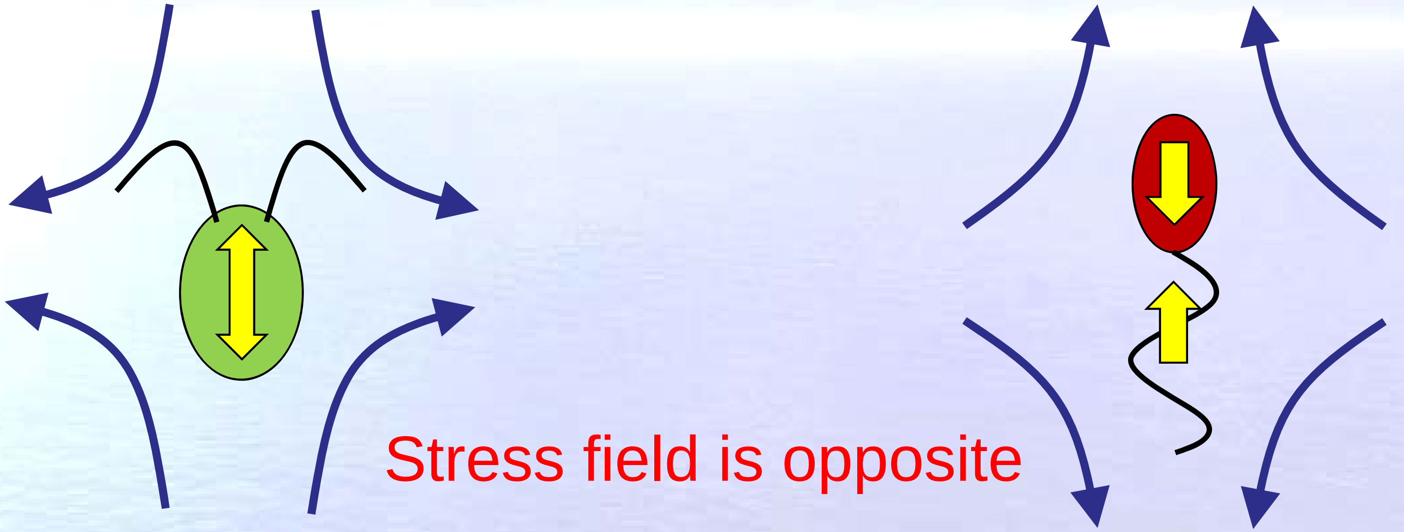
- Diffusion of particles Wu & Libchaber (2000)
- Energy is transported towards larger scale?

Outline of the talk

- Introduction
- Biomechanics of an **individual** and a **pair** of micro-organisms
- **Collective swimming** in meso-scale
- **Macroscopic properties** of a suspension of micro-organisms
- Conclusions

Rheological properties

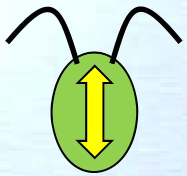
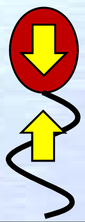
Stress field generated by a solitary cell



A bottom-heavy cell in a shear flow

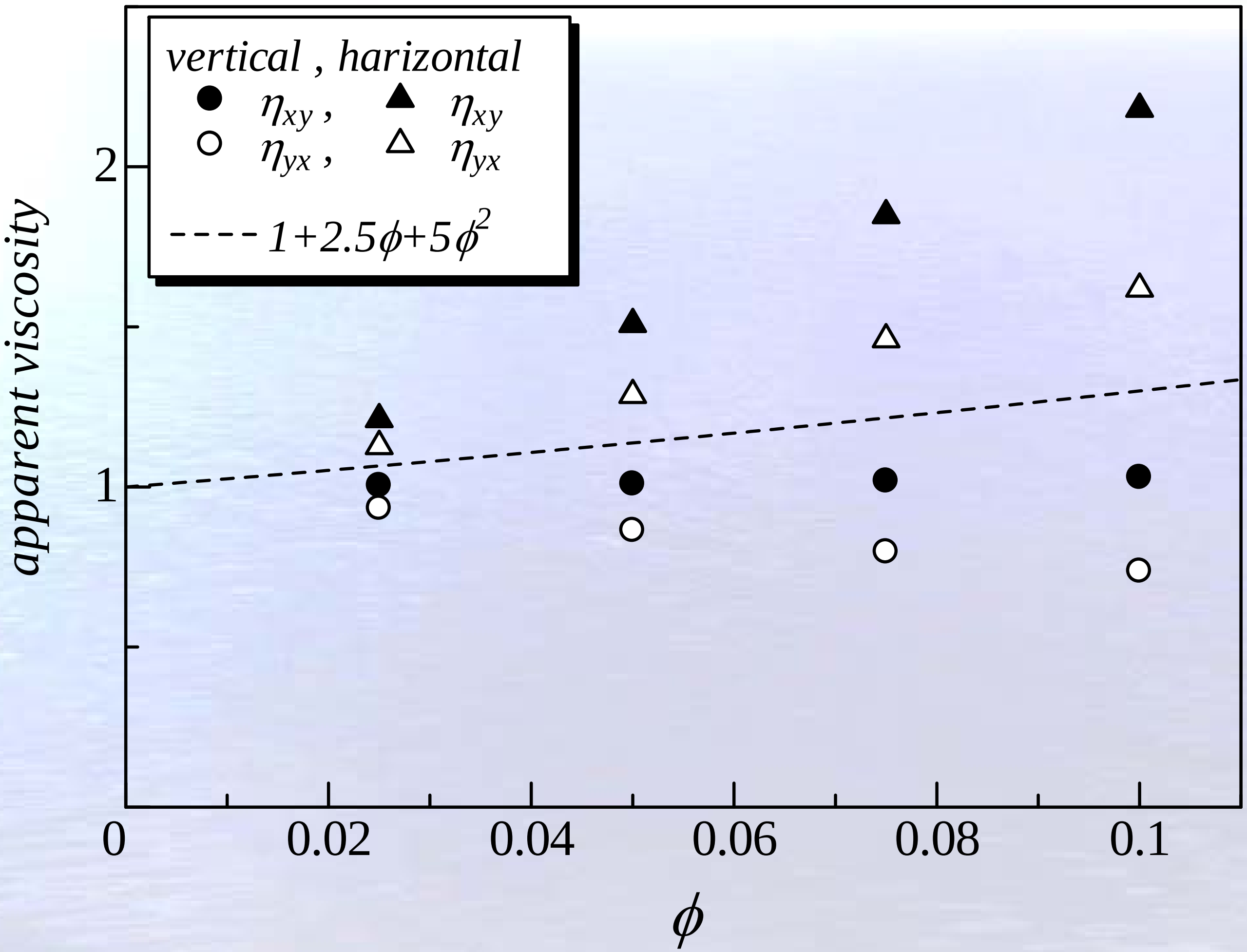
Rheological properties

Shear viscosity (compared to dead cell suspensions)

	Horizontal shear	Vertical shear
	Increase	Decrease
	Decrease	Increase

Rheological properties

Shear viscosity : Stresslet is a part of the problem



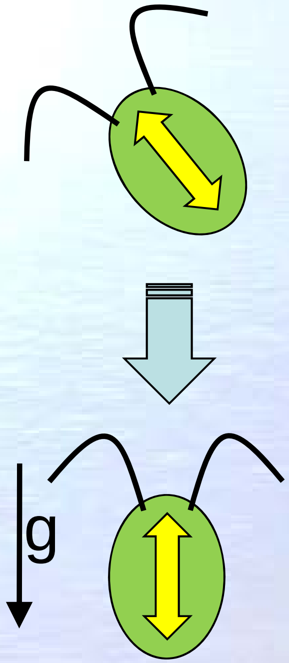
Ishikawa & Pedley, *J. Fluid Mech*. (2007)

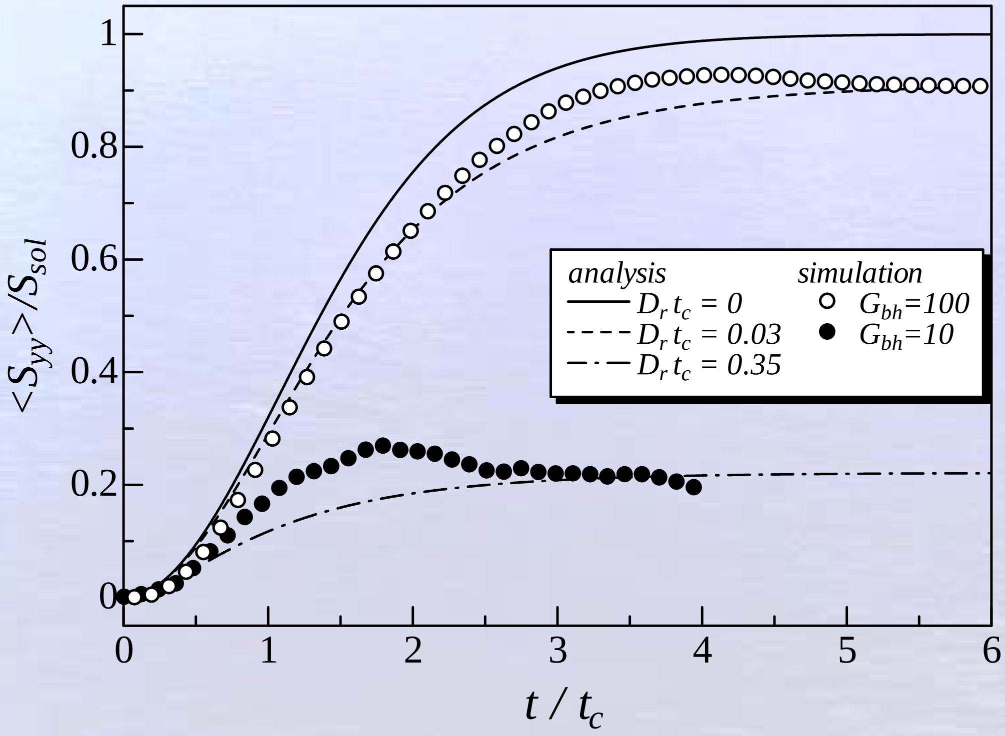
Rheological properties

Normal stress differences appears

Relaxation time of the stress field

Ishikawa *et al.*, *J. Theor. Biol.* (2007)



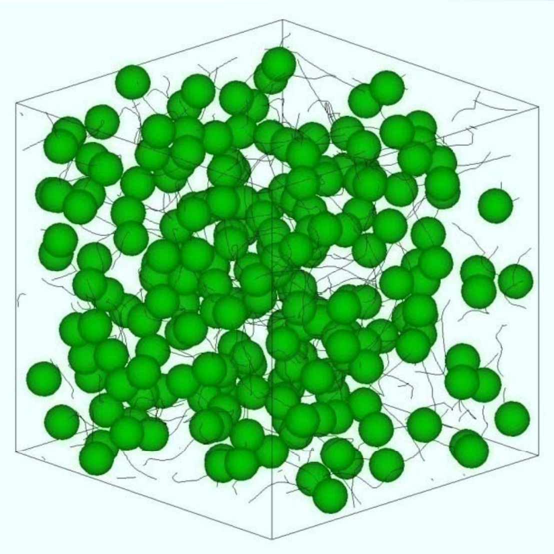


Shows strong non-Newtonian property

Diffusion properties

Cell Conservation (continuum model)

$$\frac{Dn}{Dt} = -\nabla \cdot \left(n\mathbf{V}_c + \mathbf{J}_r \right) \quad \text{[+ birth, death, etc]}$$



where $\mathbf{V}_c$ = mean cell swimming velocity,

$\mathbf{J}_\mathrm{r}$ = flux due to random cell swimming

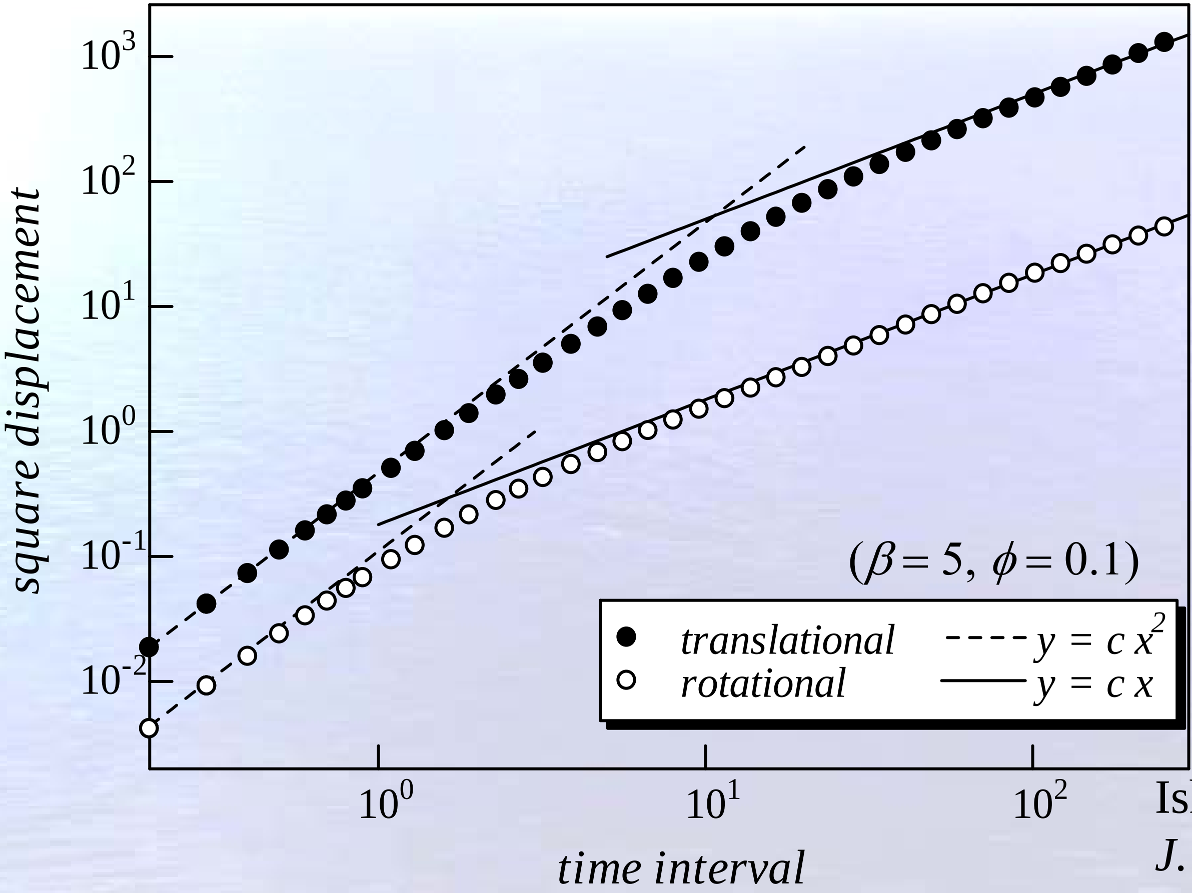
$$\boxed{\mathbf{J}_r = -\mathbf{D} \cdot \nabla n \ \ ?}$$

Definition of **D**

$$\mathbf{D} = \lim_{t \to \infty} \frac{\left\langle \left[\mathbf{r}(t+t_0) - \mathbf{r}(t_0) \right] \left[\mathbf{r}(t+t_0) - \mathbf{r}(t_0) \right] \right\rangle}{2t}$$

Diffusion properties

Self-diffusion of cells



Ishikawa & Pedley, *J. Fluid Mech.* (2007)

The spreading is correctly described as a diffusive process

Diffusion properties

Diffusion of fluid particles

Fluid particle motions : hybrid SDM-BEM

Ishikawa and Yamaguchi, *Phys. Rev. E* (2008)

$$u_i(x) - \langle u_i(x) \rangle = \boxed{\frac{1}{8\pi\mu}\sum_{\alpha=1}^{N}\left[\left(1+\frac{a^2}{6}\nabla^2\right)J_{ij}F_j^{\alpha} + R_{ij}L_j^{\alpha} + K_{ijk}S_{jk}^{\alpha} + \nabla_k\nabla_l J_{ij}Q_{klj}^{\alpha}\right]}$$

$$\boxed{-\frac{1}{8\pi\mu}\sum_{\alpha=1}^{N_{near}}\sum_{m=2}^{\infty}\int_{A_\alpha}\left[\frac{(-1)^m}{m!}\frac{\partial^m}{\partial k_1\cdots\partial k_m}J'_{ij}(y_{k_1}-x_{k_1}^{\alpha})\cdots(y_{k_m}-x_{k_m}^{\alpha})q_j(y)\right]dA_y}$$

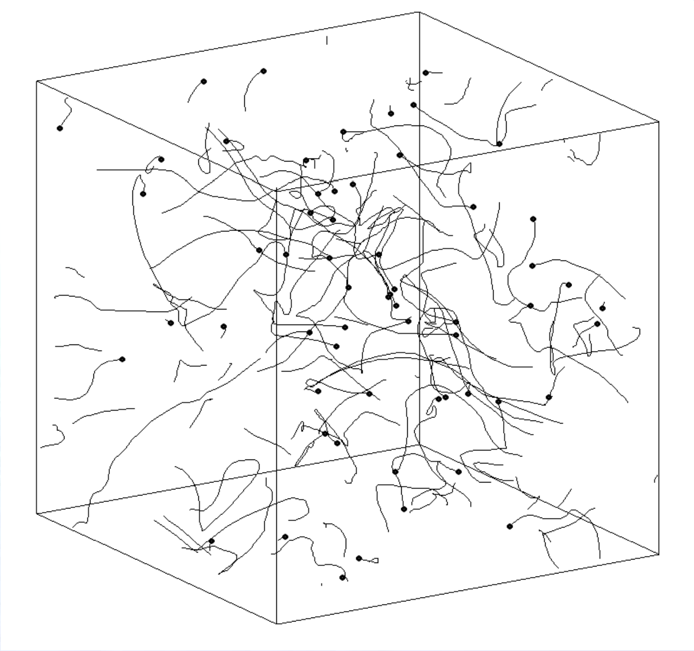
Near field effect is calculated by BEM and L.T.

N_{near}=2 : semi-dilute : compiled a database in priori

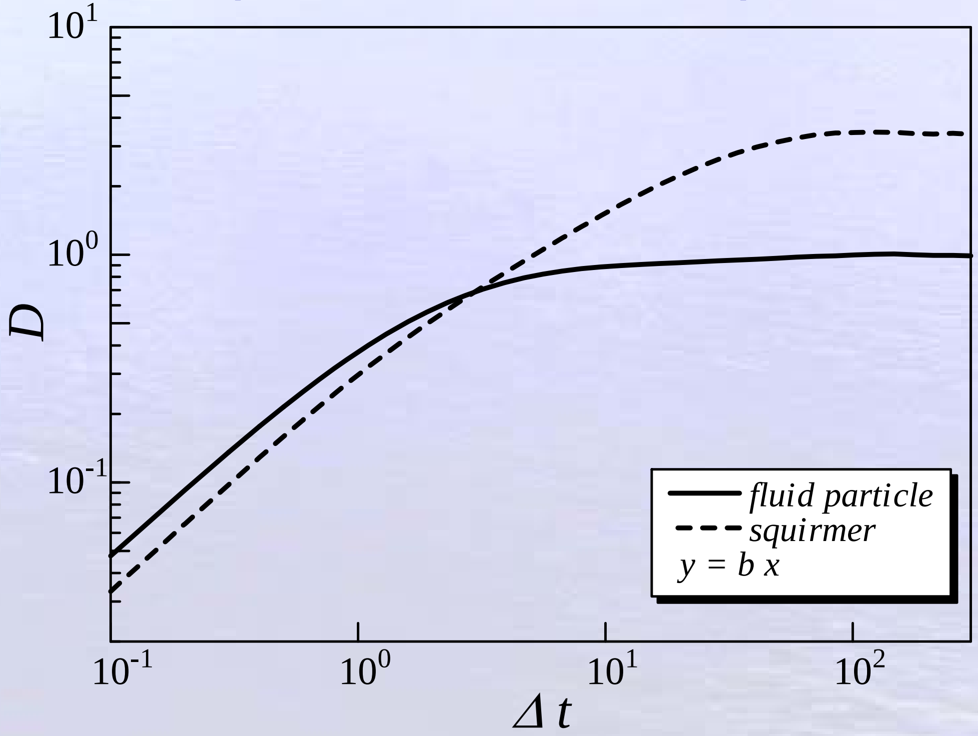
Diffusion properties

Diffusion of fluid particles

Trajectories of fluid particles



Diffusion coefficient of squirmers and fluid particles

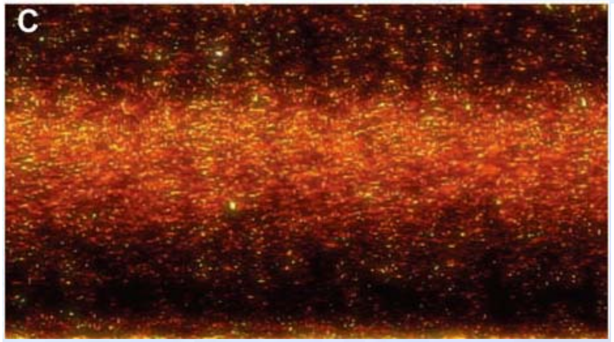


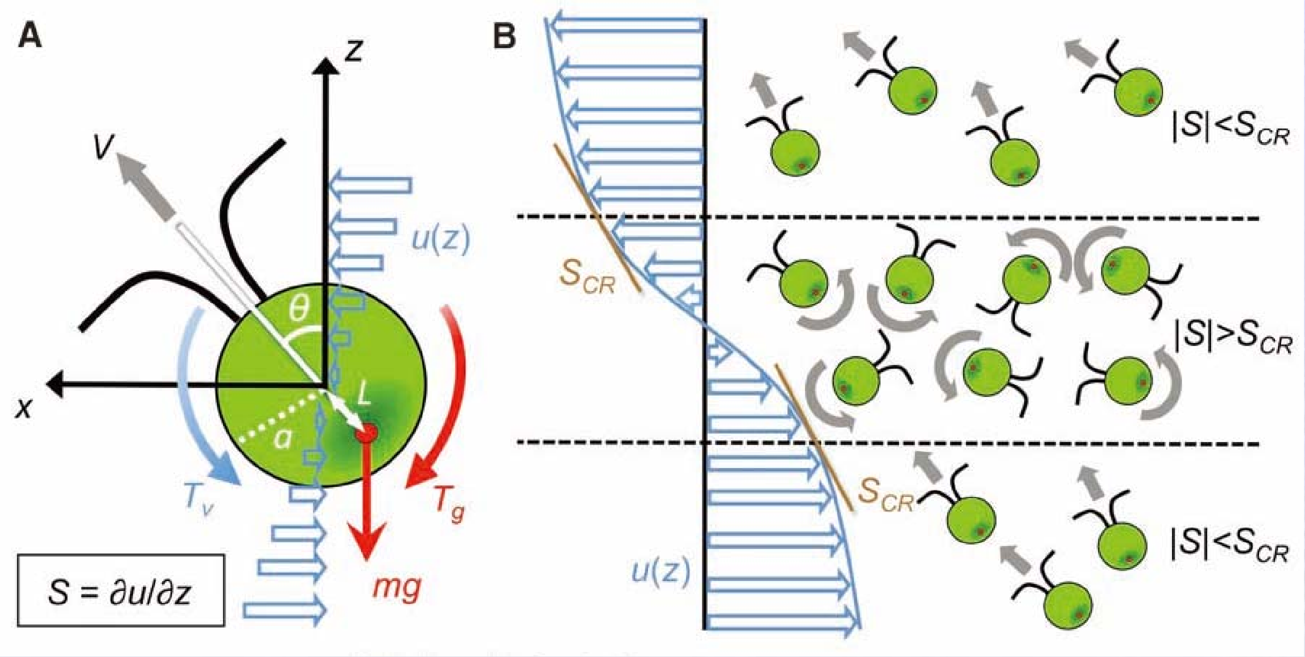
Ishikawa *et al.*, *Phys. Rev. E* (2010)

Spreading of fluid particles is also a diffusion process

Large Scale Example in Nature

Thin layers of plankton are important hotspots of ecological activity



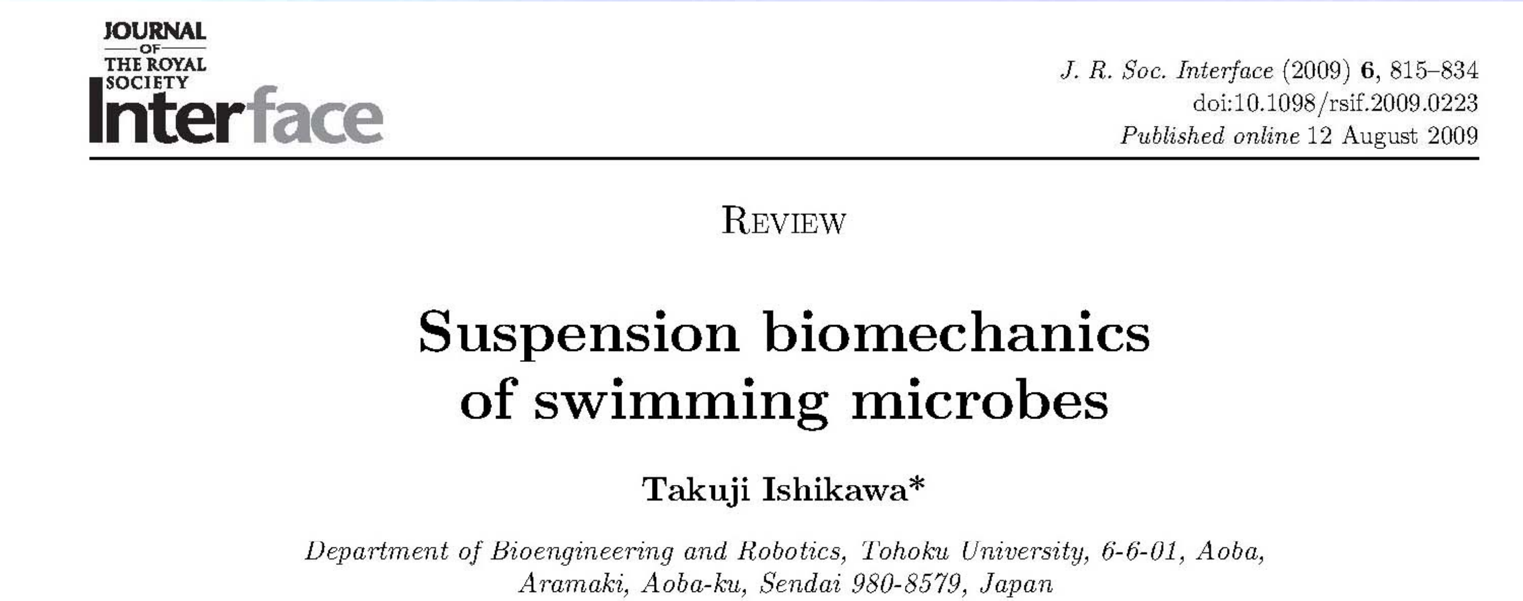


Durham *et al.*, *Science* (2009)

Conclusions

By using the bottom-up strategy, suspension biomechanics of swimming microbes can be clarified much further.

We need more biophysics and biomechanics to understand various phenomena of micro-organisms.



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REVIEW

Suspension biomechanics of swimming microbes

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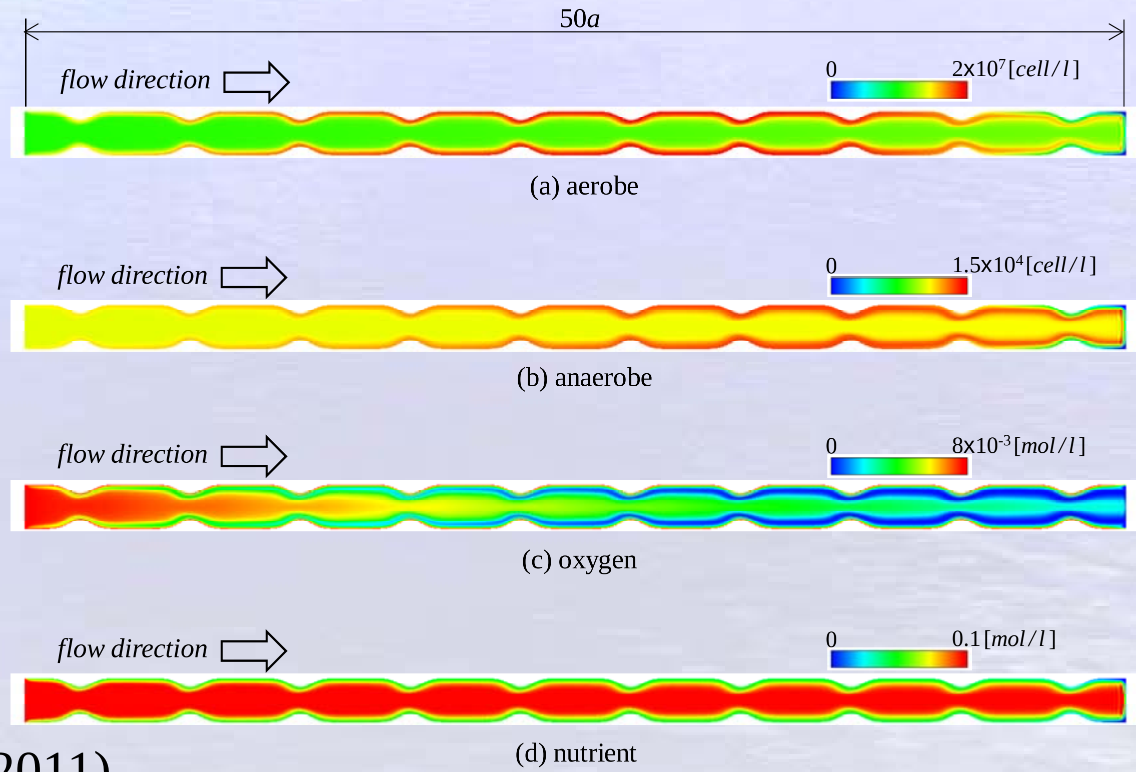
Recent review paper on this topic

Conclusions

Applications: microbial flora, for instance



Simultaneously solving:

- Flow field generated by peristalsis
- Concentrations of oxygen and nutrient
- Densities of anaerobes and aerobes

Ishikawa *et al.*, *J. Theor. Biol.* (2011)

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Dr. J. T. Locsei (Cambridge)

Volvox experiments

Prof. R. E. Goldstein (Cambridge)

Coherent structures

Dr. E. Lauga (UCSD)

Prof. T. Yamaguchi (Tohoku Univ.)

Lab members

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Tohoku Univ. Global COE Program

Thank you for your listening