Physics of Biological Systems

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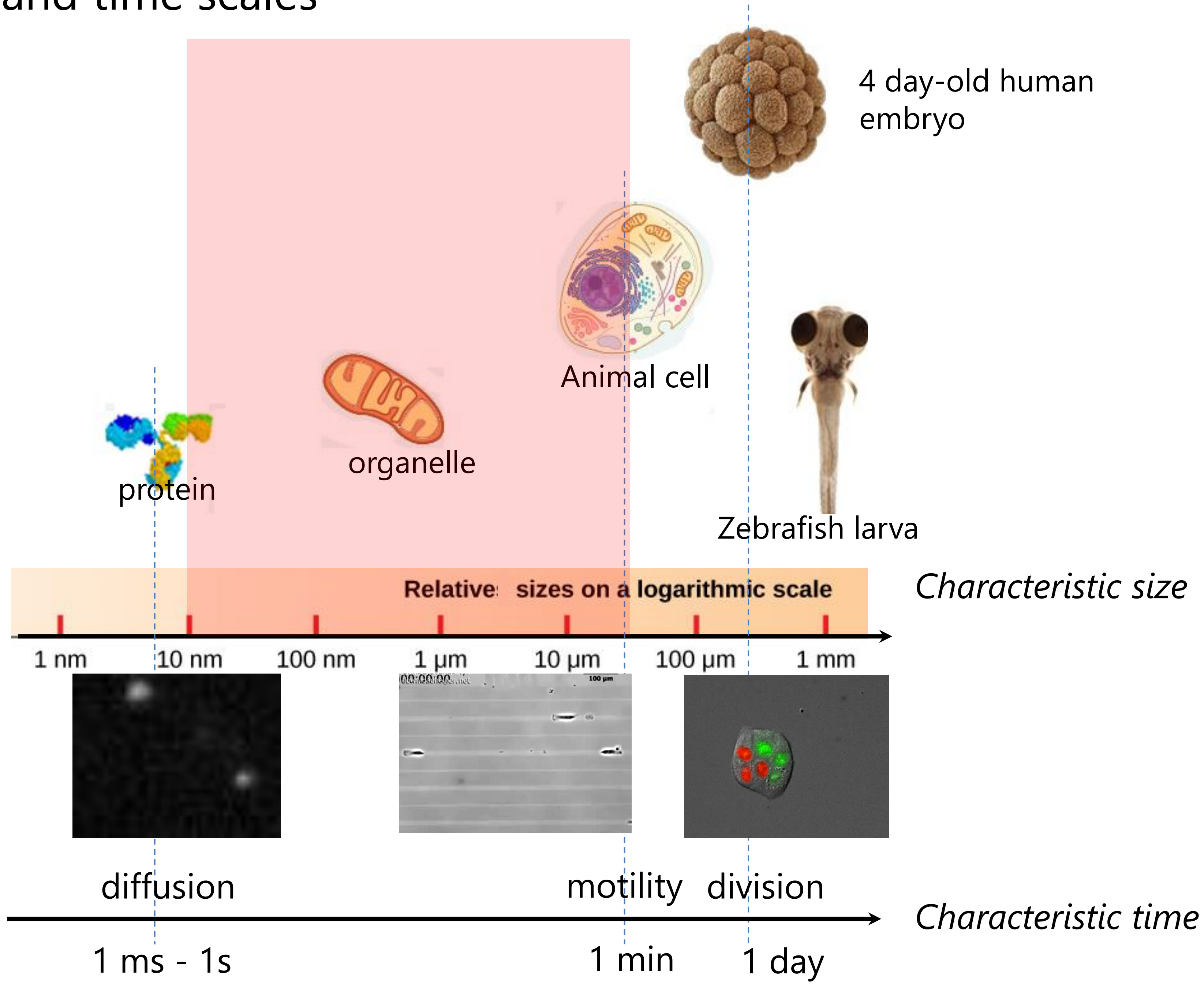
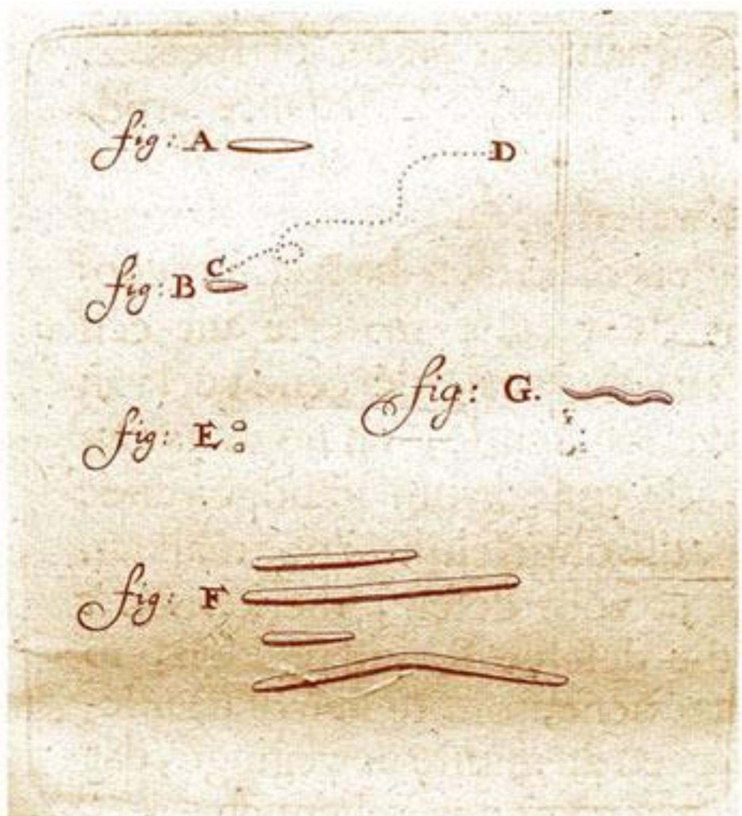
Space and time scales



Transport in biology

- Diffusion – Brownian motion
- Active transport
- Cell motility

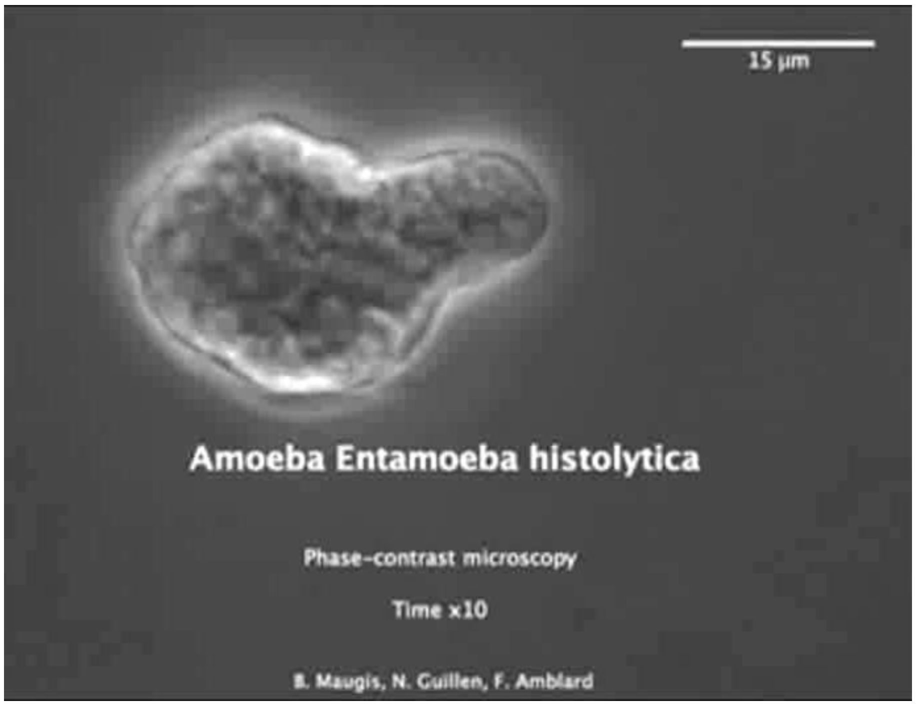
First report of cell movements



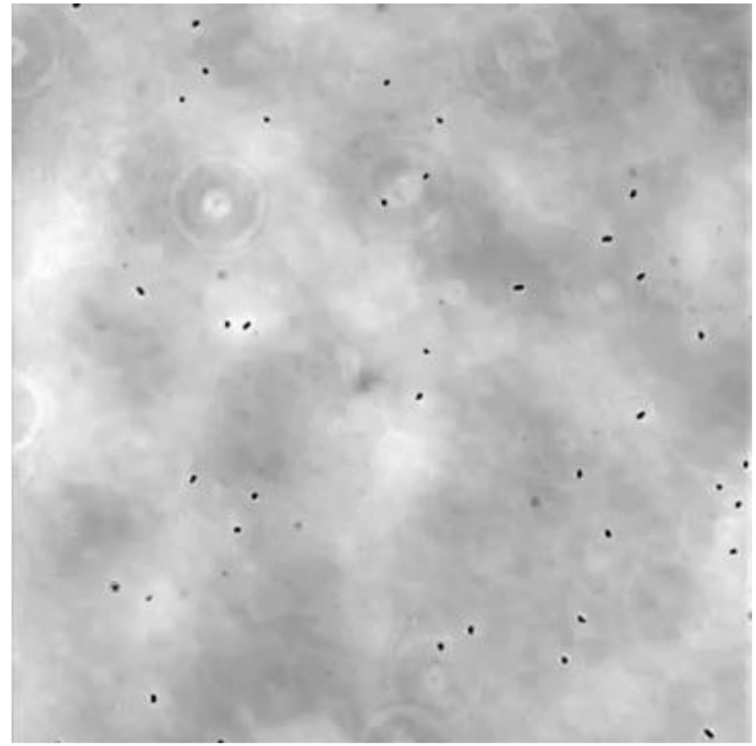
Nature Reviews | Molecular Cell Biology

van Leeuwenhoek's description of microbes in tooth plaque in a letter to Francois Aston in 1683 contains a drawing that shows the short rods of bacilli and bacteria, the spheres of micrococci, and the corkscrew spirillum.

Cell motility



Migration of an amoeba
(on a surface)



Swiming of E.Coli bacteria
(low mag)

Q: Can we quantitatively characterize these movements?

Molecular origin of Brownian motion from a macroscopic experiment

Brownian motion

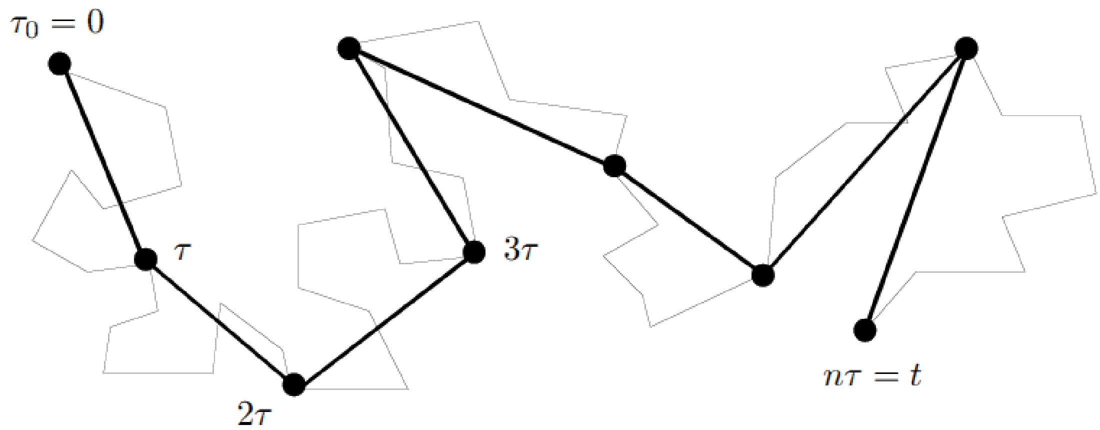
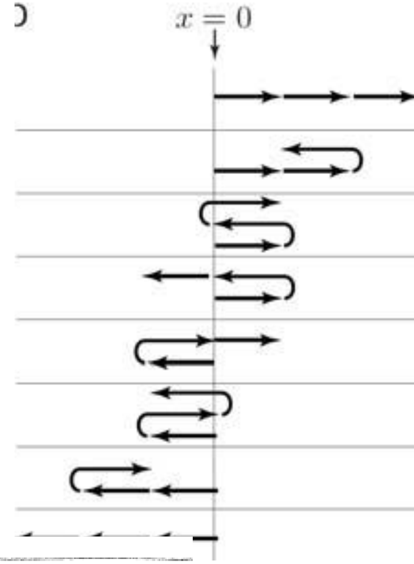


FIG. 1.1 – Trajectoire brownienne aux différentes échelles de temps. La particule brownienne subit beaucoup de collisions pendant l'intervalle de temps τ.

Thermal velocity $$V = \sqrt{\frac{3k_B T}{M}}$$

1D Random walks



$\{x_1, x_2, x_3\}$, cm	x_2k_3, cm	
$+1, +2, +3$	$+2$	0
$+1, +2, +1$	-2	
$+1, \ 0, +1$	0	0
$+1, \ 0, -1$	0	
$-1, \ 0, +1$	0	0
$-1, \ 0, -1$	0	
$-1, -2, -1$	-2	0
$-1, -2, -3$	$+2$	

- What is mean displacement?

$$\langle R \rangle = \left\langle \sum_{i=1}^{N} x_i \right\rangle = \sum_{i=1}^{N} \langle x_i \rangle = 0 \quad \text{since } \langle x_i \rangle = 0$$

- Variance?

$$\langle R^2 \rangle = \left\langle \sum_{i=1}^{N} \sum_{j=1}^{N} x_i x_j \right\rangle = \sum_{i=1}^{N} \langle x_i^2 \rangle + \sum_{i \neq j}^{N} \langle x_i x_j \rangle$$

- for the 2nd term $\langle x_i x_j \rangle = 0$ since the steps are independent and there are equal # of $+a^2$ and $-a^2$ terms.

- 1st term $\langle x_i^2 \rangle = a^2$ so

$$\langle R^2 \rangle = N a^2$$

- or RMSD

$$\boxed{\sqrt{\langle R^2 \rangle} = a\sqrt{N}}$$

$$\langle x_j \rangle = 0$$

$$\langle x_j^2 \rangle \neq 0$$

Random walks

$$\left\langle (x_N)^2 \right\rangle = NL^2 \qquad N = t/\Delta t$$

Define the **diffusion constant** as: $D = L^2/(2\Delta t)$

The **mean square displacement** of a 1D random walk increases linearly in time: $\boxed{\left\langle (x_N)^2 \right\rangle = 2Dt}$

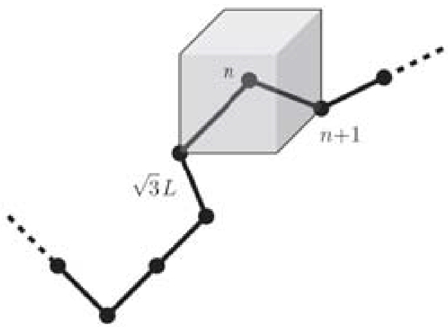
2D: $\left\langle (\vec{r}_N)^2 \right\rangle = 4Dt$

3D: $\left\langle (\vec{r}_N)^2 \right\rangle = 6Dt$

The diffusion law is universal, as long as we have some distribution of random, independent steps

Random walks and Polymers

Mathematics of random walks is also the appropriate language to understand the conformation of many biological macromolecules



$$\sqrt{\left\langle \vec{r}_N^{\,2} \right\rangle} = \sqrt{\left\langle x_N^2 \right\rangle + \left\langle y_N^2 \right\rangle + \left\langle z_N^2 \right\rangle}$$
$$= L\sqrt{3N}$$

Random coil polymers are loose structures

Assumption that a monomer is equally likely to occupy all the adjacent spaces breaks down when the monomers are strongly attracted to one another (see protein folding).

Self avoidance in 3D results in an exponent 0.58

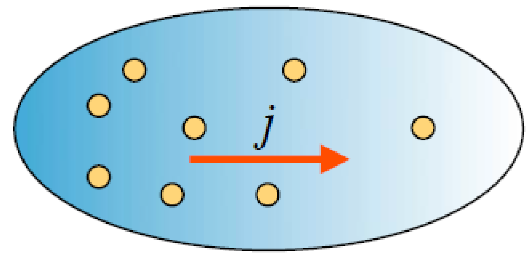
In all cases: $\boxed{R \propto LN^{\nu}}$ (ν is called Flory exponent)

Diffusion

Brownian motion leads to diffusion

Fick's law $$j = -D\frac{\partial c}{\partial x}$$

continuity equation $$\frac{\partial c}{\partial t} = -\frac{\partial j}{\partial x}$$



$$\boxed{\frac{\partial c}{\partial t} = D\frac{\partial^2 c}{\partial x^2}}$$

diffusion equation

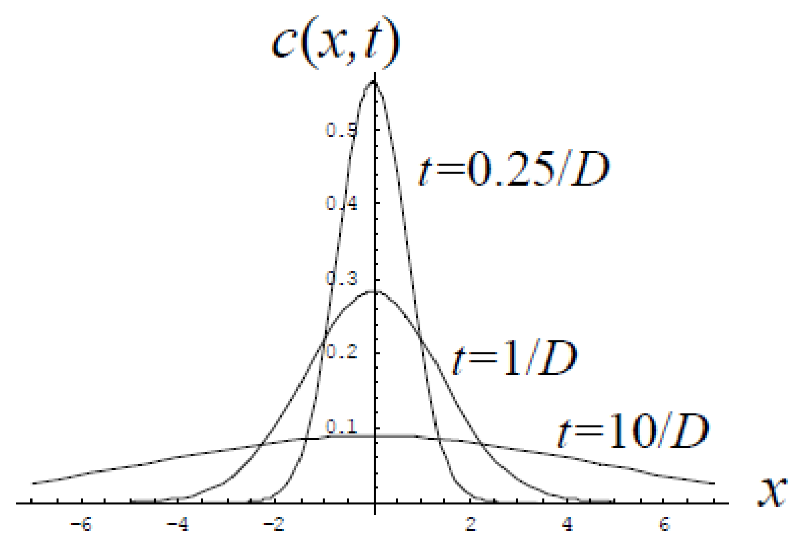
Only true for large numbers!

j: number flux [$m^{-2}\,s^{-1}$]; c: number density or concentration [m^{-3}]; D: diffusion constant [$m^2\,s^{-1}$]

Pulse solution

$$\left.\begin{aligned} &\frac{\partial c}{\partial t} = D\frac{\partial^2 c}{\partial x^2} && \text{(Diffusion equation)} \\ &c(x,0) = \delta(x) && \text{(initial condition)} \end{aligned}\right\}$$

$$\boxed{c(x,t) = \frac{1}{\sqrt{4\pi Dt}}\,e^{-x^2/4Dt}}$$



Deriving the Einstein relation

What is the relation between friction and diffusion?

Flux in the presence of a driving force: $j_F(x) = v_{drift}c = \frac{F}{\xi}c$

Total flux including diffusion: $j(x) = -D\frac{dc}{dx} + \frac{F}{\xi}c$

Steady-state: $D\frac{dc}{dx} = \frac{F}{\xi}c$ $\qquad D\xi\frac{dc}{c} = Fdx = -dU$

$$\frac{c(x)}{c(0)} = \frac{e^{-U(x)/D\xi}}{e^{-U(0)/D\xi}}$$

This is the Boltzmann distribution we expect if we set: $\boxed{D\xi = kT}$

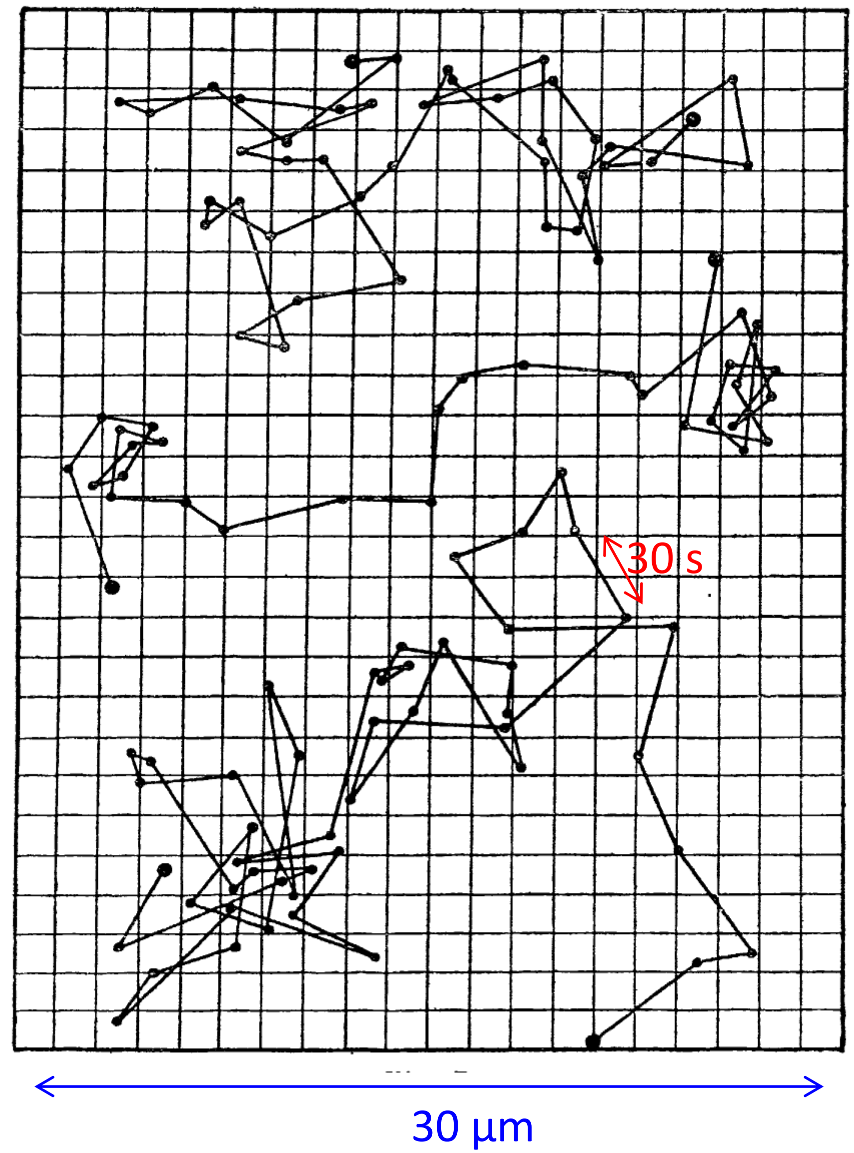
Einstein relation

ξ: friction coefficient [kg s^{-1}]

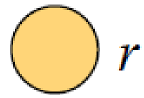
Robert Brown, 1827*: "Having found motion in the particles of the pollen of all the living plants which I had examined, I was led next to inquire whether this property continued after the death of the plant, and for what length of time it was retained*

Putty (mastic in french) grains (0.2 µm)

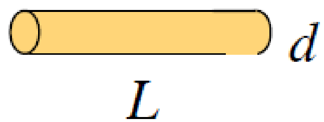
Jean Perrin, 1909



Friction coefficients and diffusion times

sphere 

$$\zeta = 6\pi\eta r$$

cylinder 

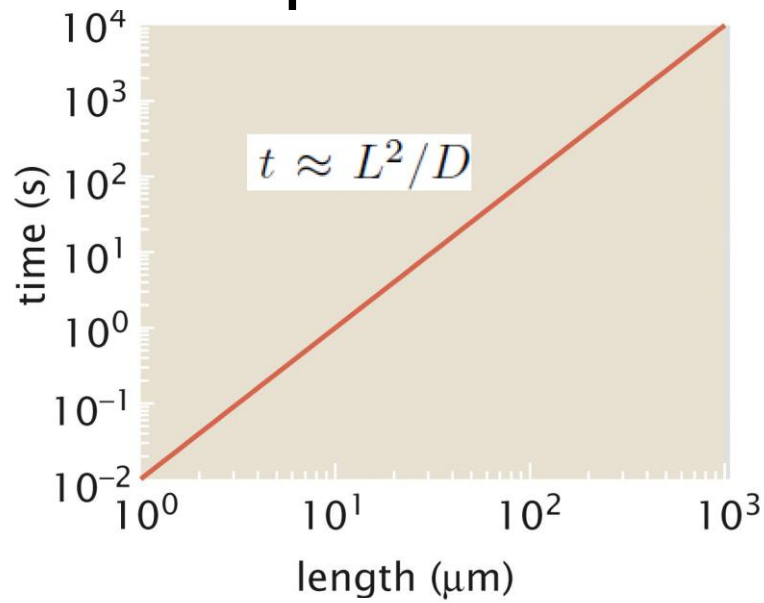
$$\zeta_{\parallel} = \frac{2\pi\eta L}{\ln(L/d)} \qquad \zeta_{\parallel} = \frac{4\pi\eta L}{\ln(L/d)}$$

		1 μm	100 μm	10 mm
ions	(0.1 nm)	0.2 ms	2s	7 hr
proteins	(3 nm)	5 ms	50 s	6 days
organelles	(500 nm)	1 s	3 hr	3 years

Diffusion is the dominant form of material transport at submicrometer scales

r: radius [m]; *L*: length [m]; *d*: diameter [m]; η: viscosity [kg m^{-1} s^{-1}]

Passive *vs* active transport



globular protein

$$D = 100\,\mu\mathrm{m}^2/\mathrm{s}$$

In 1 s, a protein or a small molecule "visits" a whole cell of 10µm in diameter. But, on neurons, long of several cm to 1m...

$$L \approx 10^6\,\mu\mathrm{m}\ (1\,\mathrm{m})$$

$$t=10^{10}\mathrm{s}=300\mathrm{y}$$

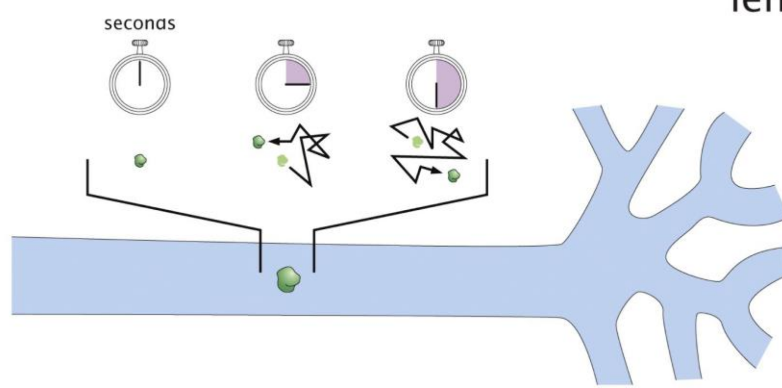


Figure 13.5a Physical Biology of the Cell, 2ed. (© Garland Science 2013)

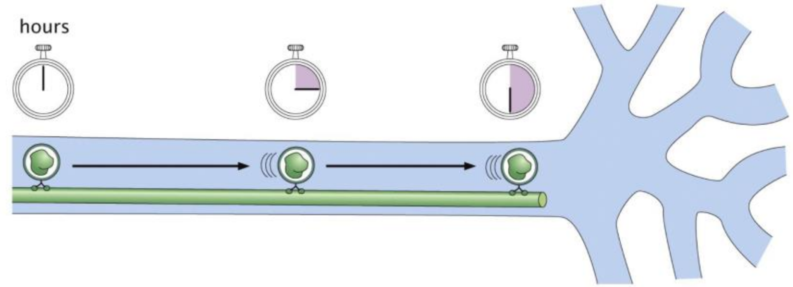


Figure 13.5b Physical Biology of the Cell, 2ed. (© Garland Science 2013)

Diffusive & directed motions of RNA polymerase

Passive: Free RNA polymerase molecule diffusing in a bacterial cell

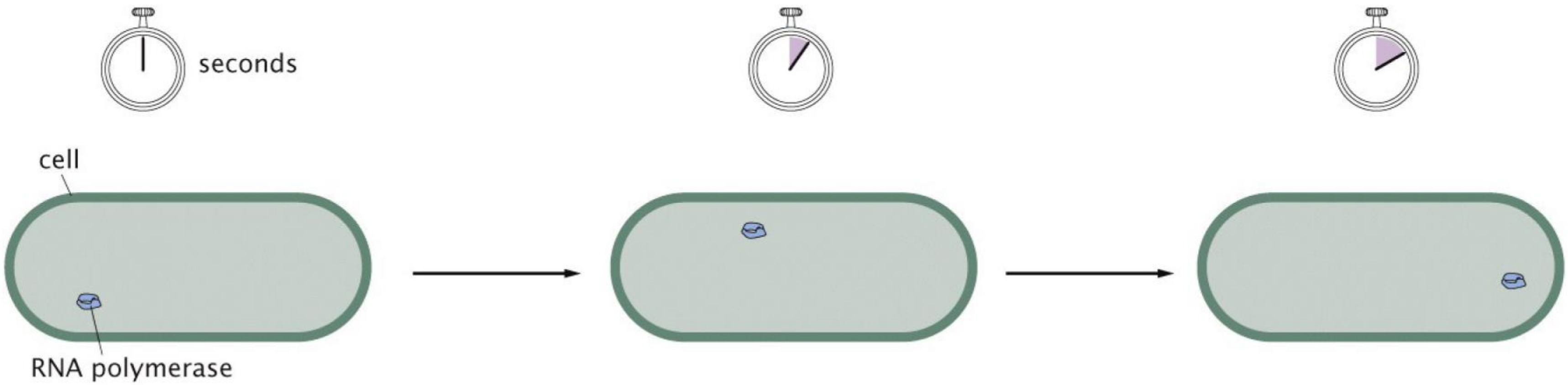


Figure 13.2a Physical Biology of the Cell, 2ed. (© Garland Science 2013)

Active: One-dimensional motion of RNA polymerase along DNA

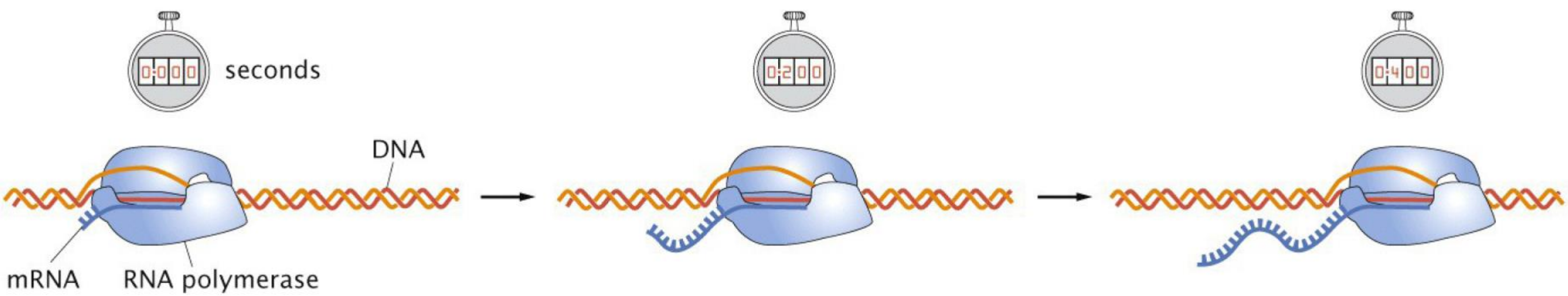


Figure 13.2b Physical Biology of the Cell, 2ed. (© Garland Science 2013)

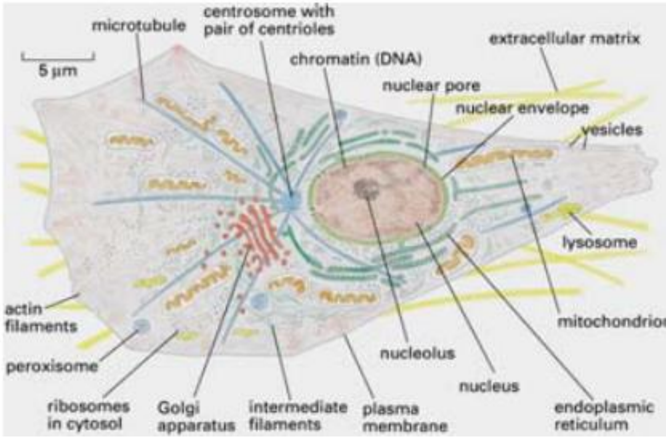
Compartmentalization and active transport

Eukaryotic cells tackle problem of organization by compartmentalization

Nucleus:	DNA replication & transcription
Mitochondria:	energy production
Endoplasmic reticulum:	protein synthesis
Golgi apparatus:	protein sorting
Lysosymes:	protein degradation and recycling
Plasma membrane:	extracellular signalling

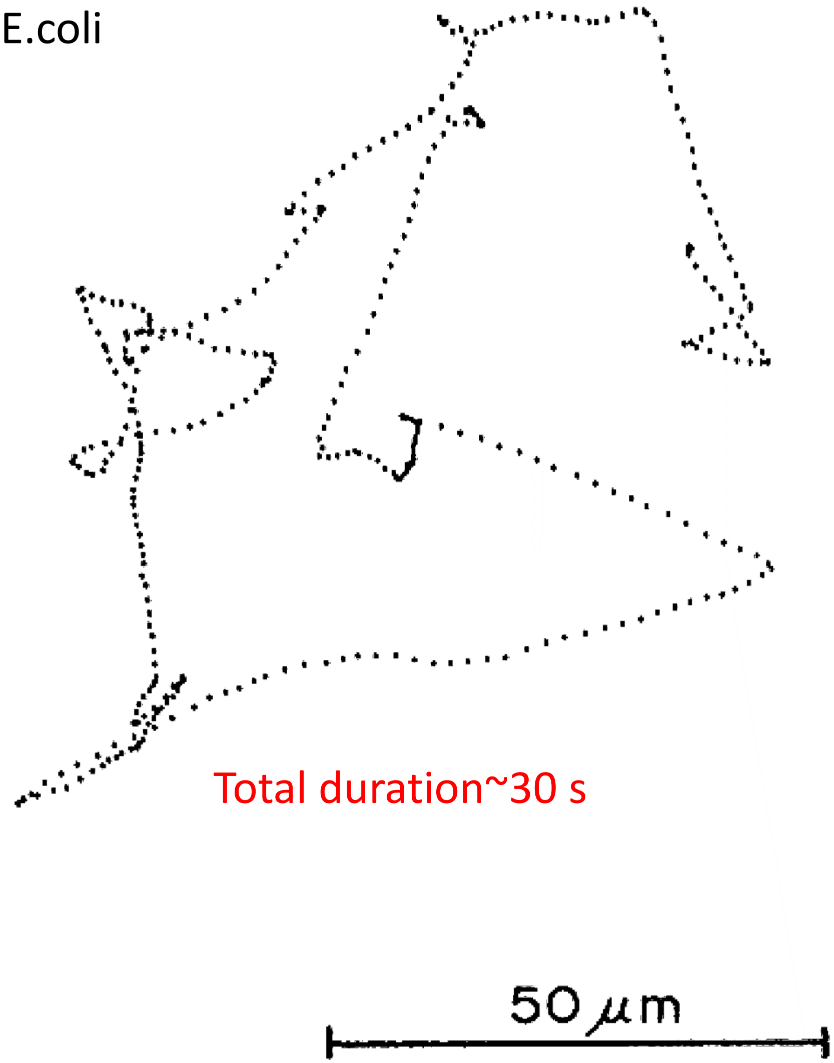
Requires active transport to move components between compartments

- Vesicle trafficking: endocytosis / exocytosis
- Cytoskeleton: filaments and motor proteins



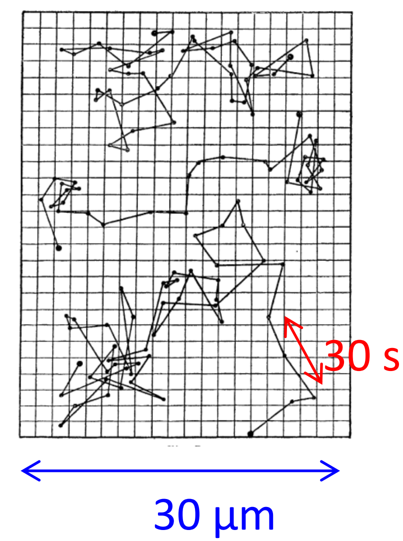
Bacteria swimming

E.coli



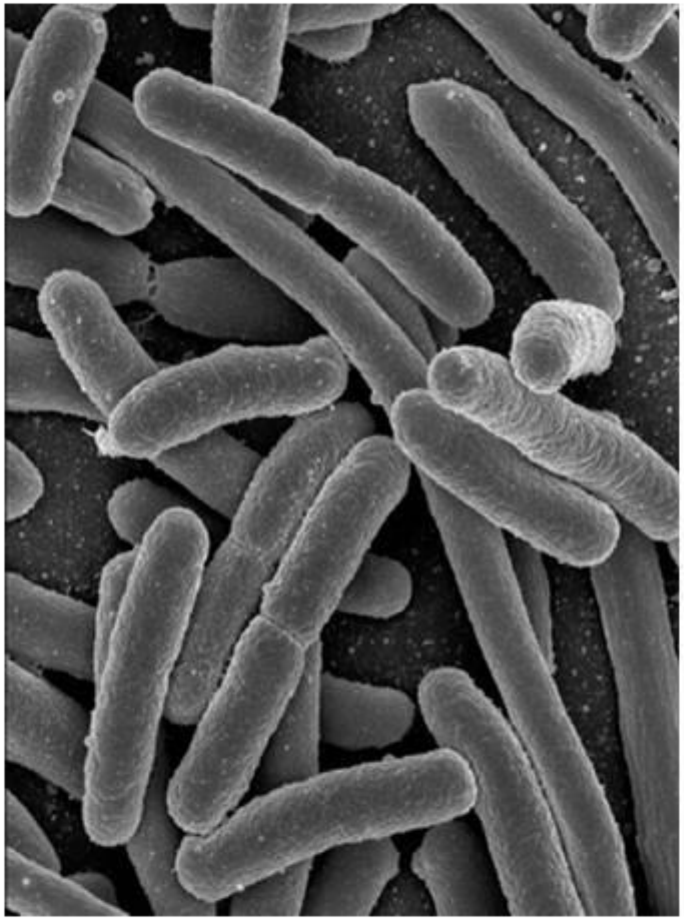
H. C. Berg & D. A. Brown, 1972

Grains de mastic
(0,2 μm)

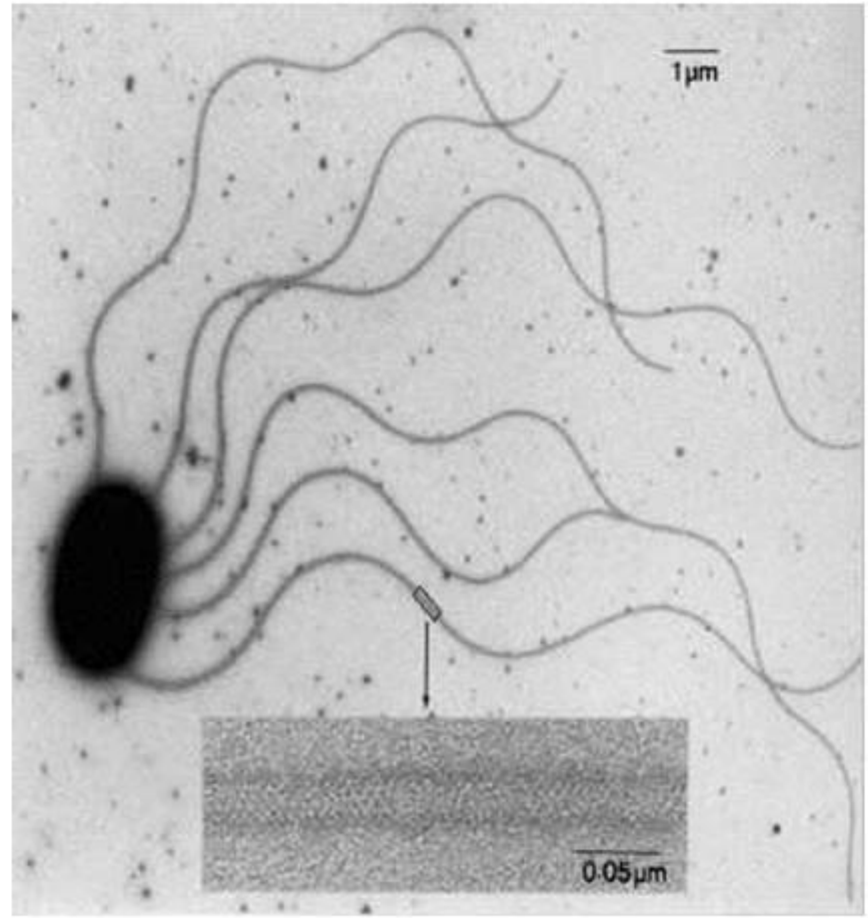


Jean Perrin, 1909

E. Coli

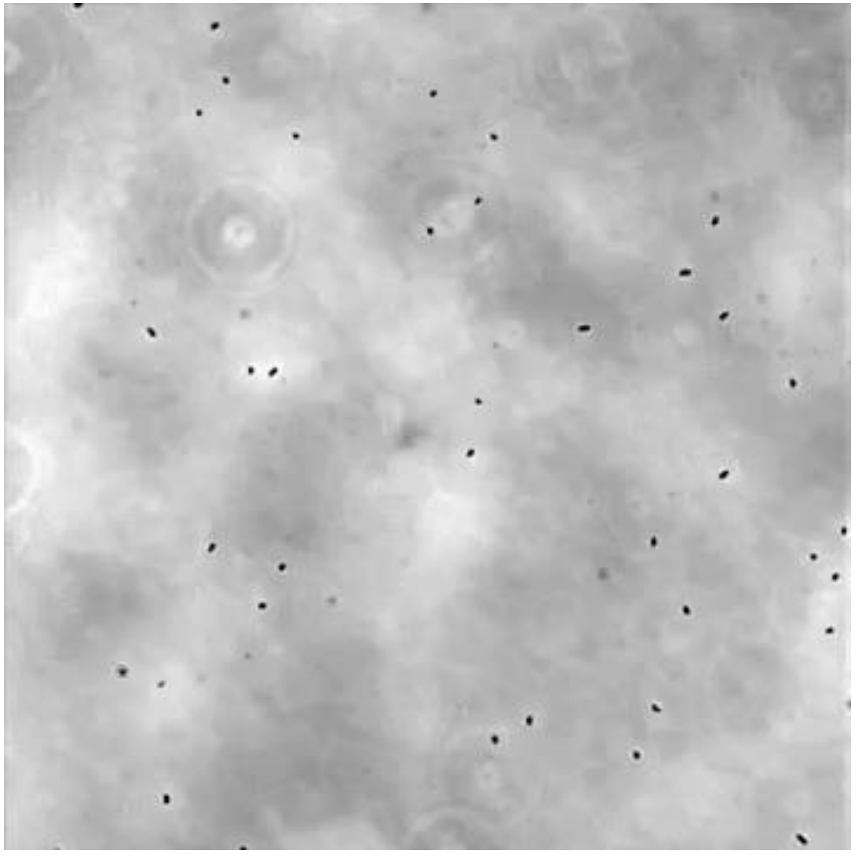


1 μm in diameter
2 μm in length



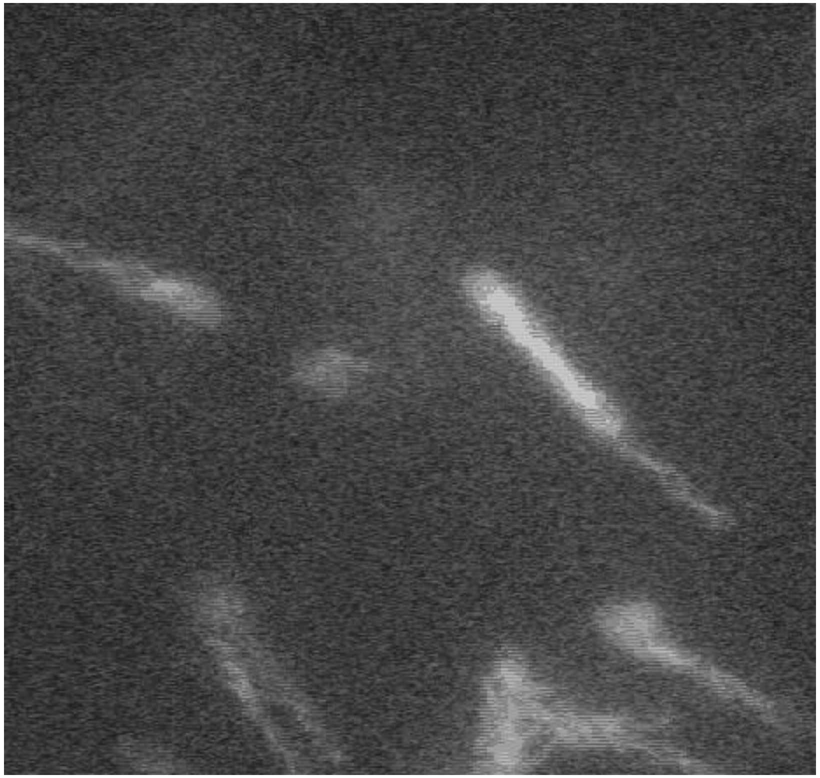
~ 6 flagella
10 μm in length

Low magnification video and trajectory tracking



E.Coli swims straight in segments (“runs”), then undergoes a random tilt (“tumble”) that causes it to change direction

High magnification video (and by fluorescence)

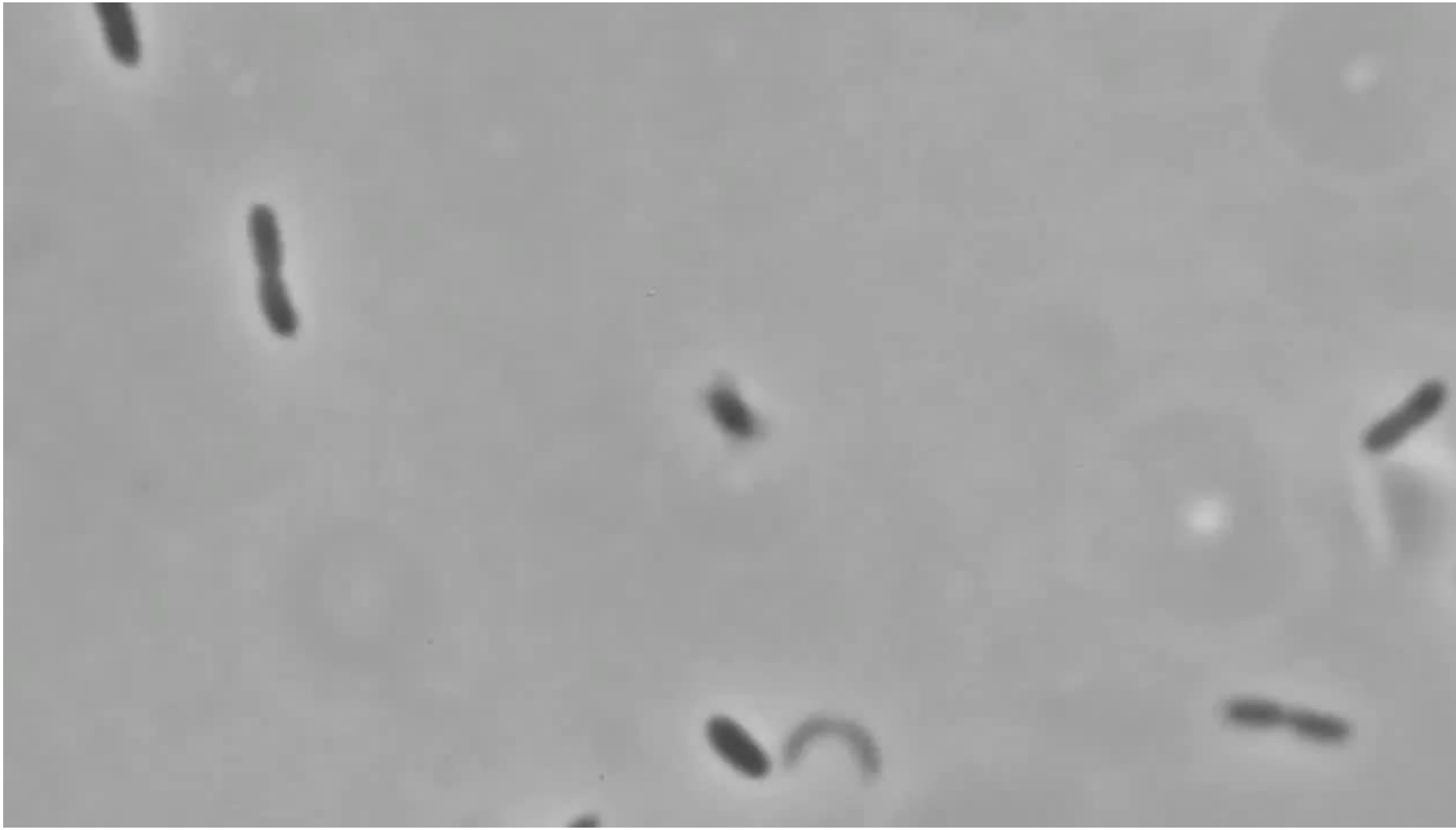


Inversion de contraste

During periods of “run”, the flagella form a bundle that has a helical rotation

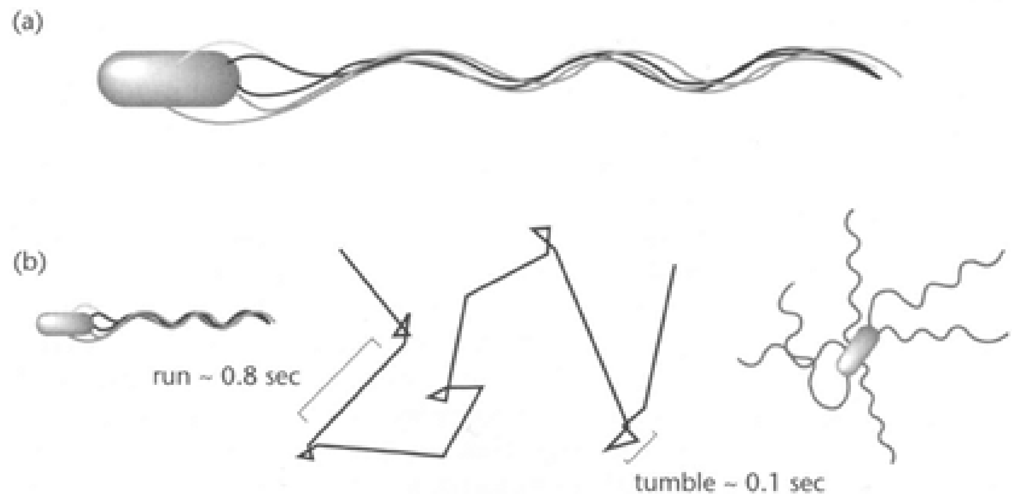
http://www.rowland.harvard.edu/labs/bacteria/index_movies.html (H. Berg)

Evidence of flagellum rotation in E.coli

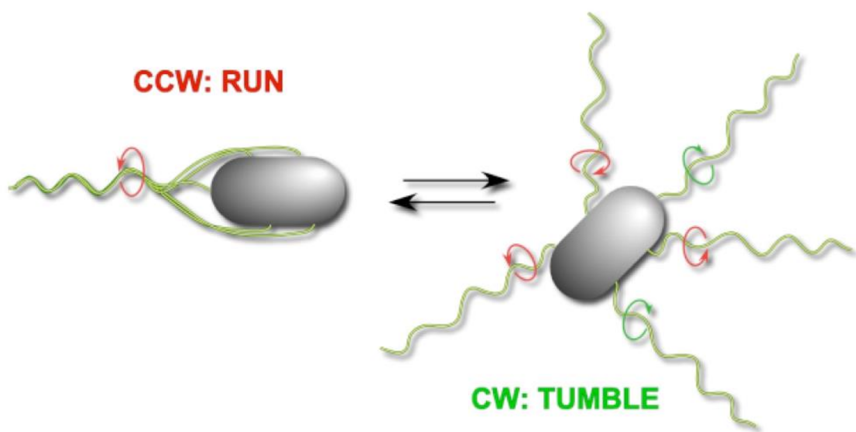


Run and tumble Sequences

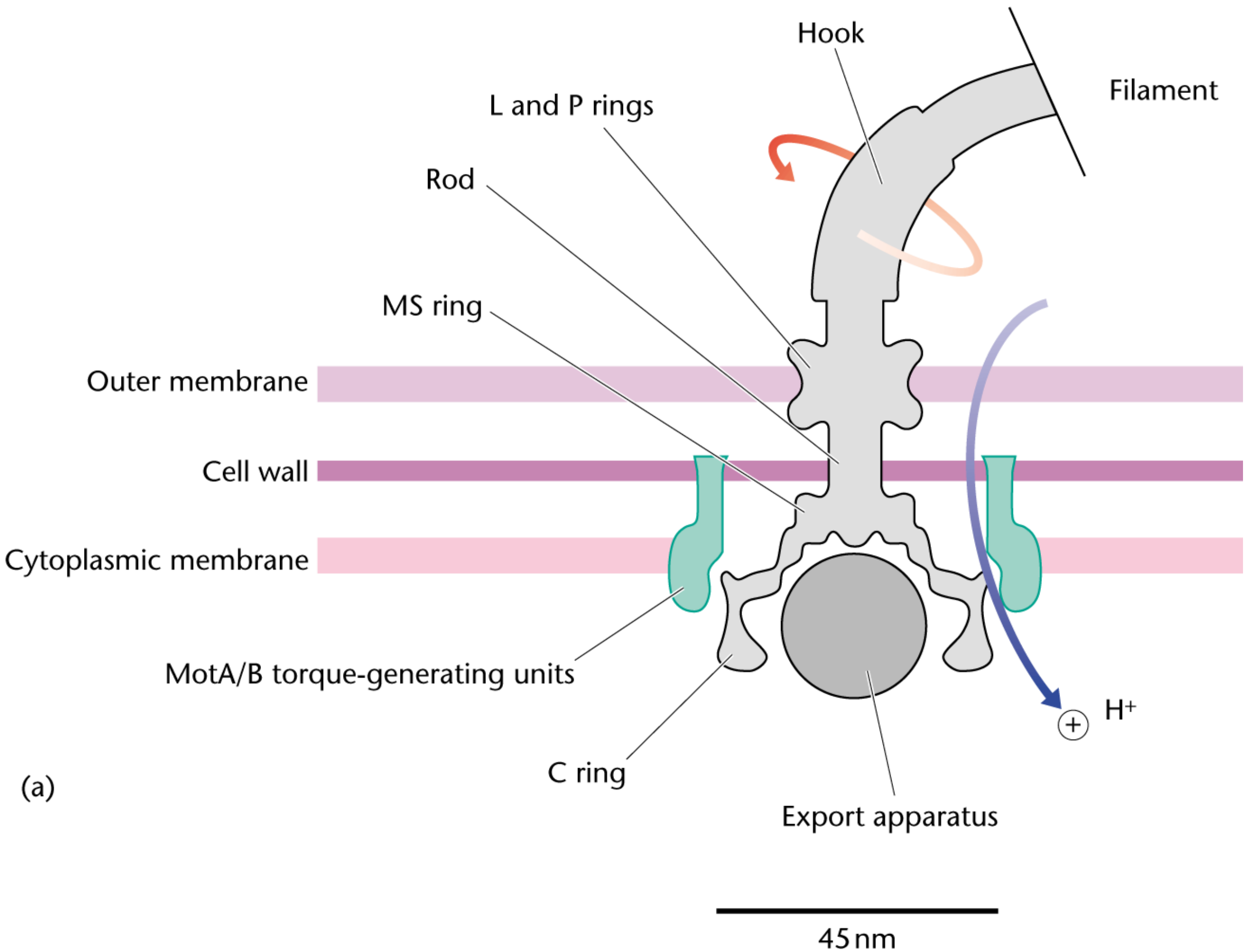
Figure 1-5 Bacterial swimming.
(a) Schematic diagram of *Escherichia coli* swimming. The cell body is a cylinder about 2 μm long and 0.5 μm in diameter and has 6–10 flagella on its surface, each up to 10 μm in length. Coordinated rotation of flagella drives the cell at speeds of about 30 μm/sec through water. (b) Under normal conditions, the bacterium alternates between periods of smooth swimming ('runs') and intermittent chaotic changes in direction ('tumbles').

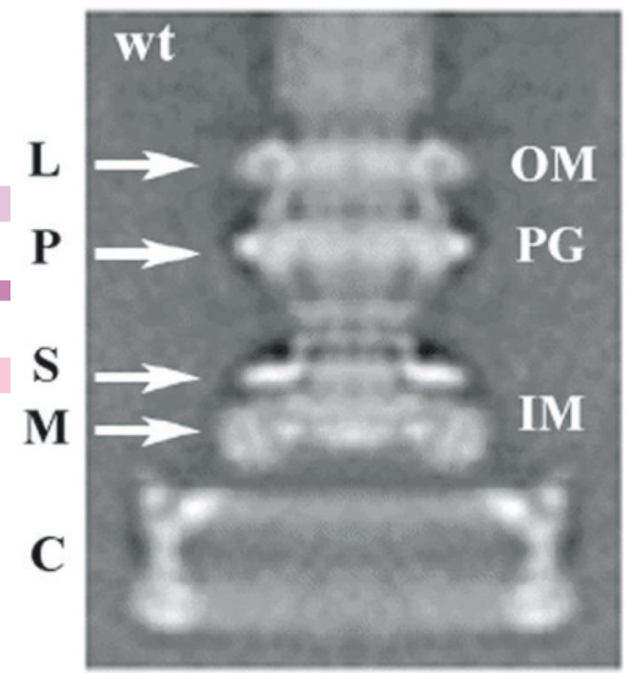


... controlled by the rotation of the motors



Anatomy of the rotary engine





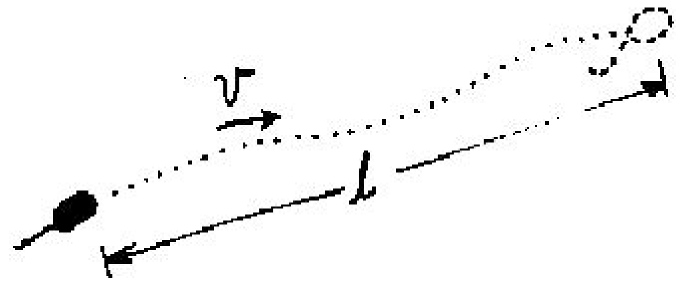
The bacterial flagellum
(animation)

Protons flux

Rotation up to ~10 000 tours/min

Estimating the length of runs

Each run must be long enough to beat the diffusion.
-The length of the runs varies with external conditions.
-Under "bad" conditions, the length of the runs is never smaller than D/v (i.e. 30 µm).



to out-swim diffusion:

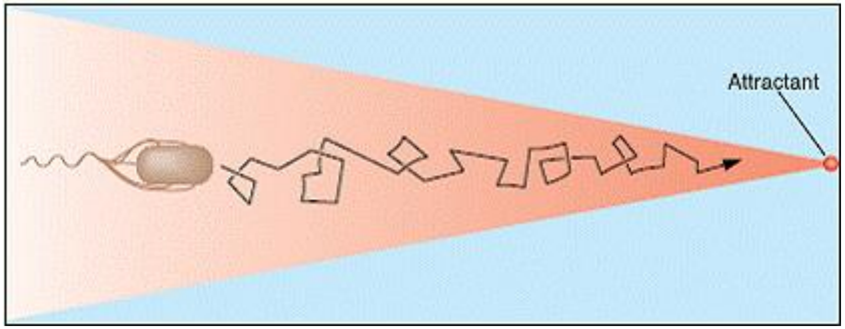
$$\ell \geq D/v$$

if $D = 10^{-5}\ cm^2/sec$, $v = .003\ cm/sec$

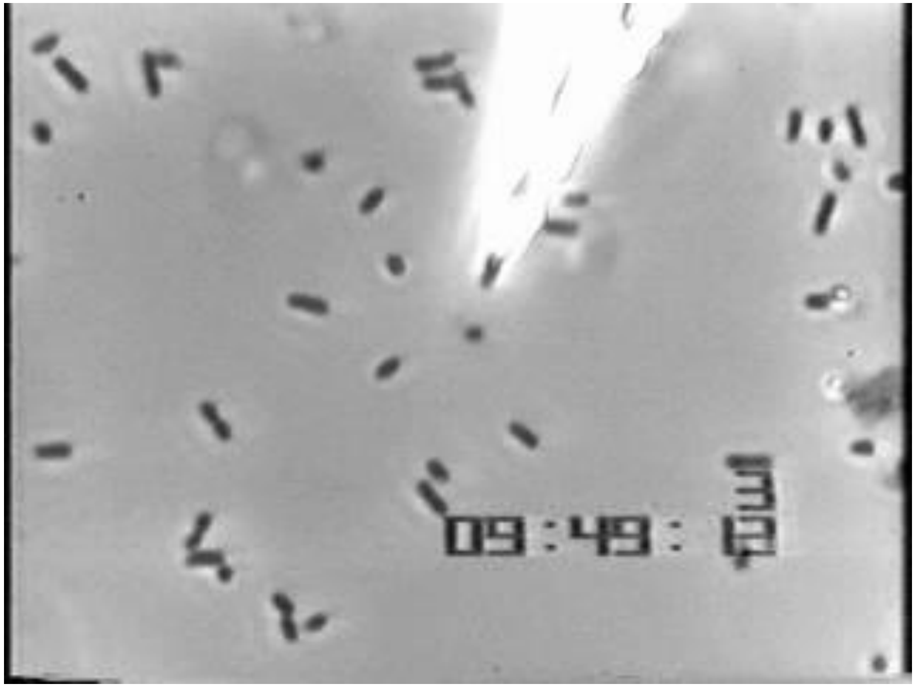
$$\ell \geq 30\ \mu$$

"If you don't swim that far you haven't gone anywhere."

E.Coli in the presence of chemoattractant?

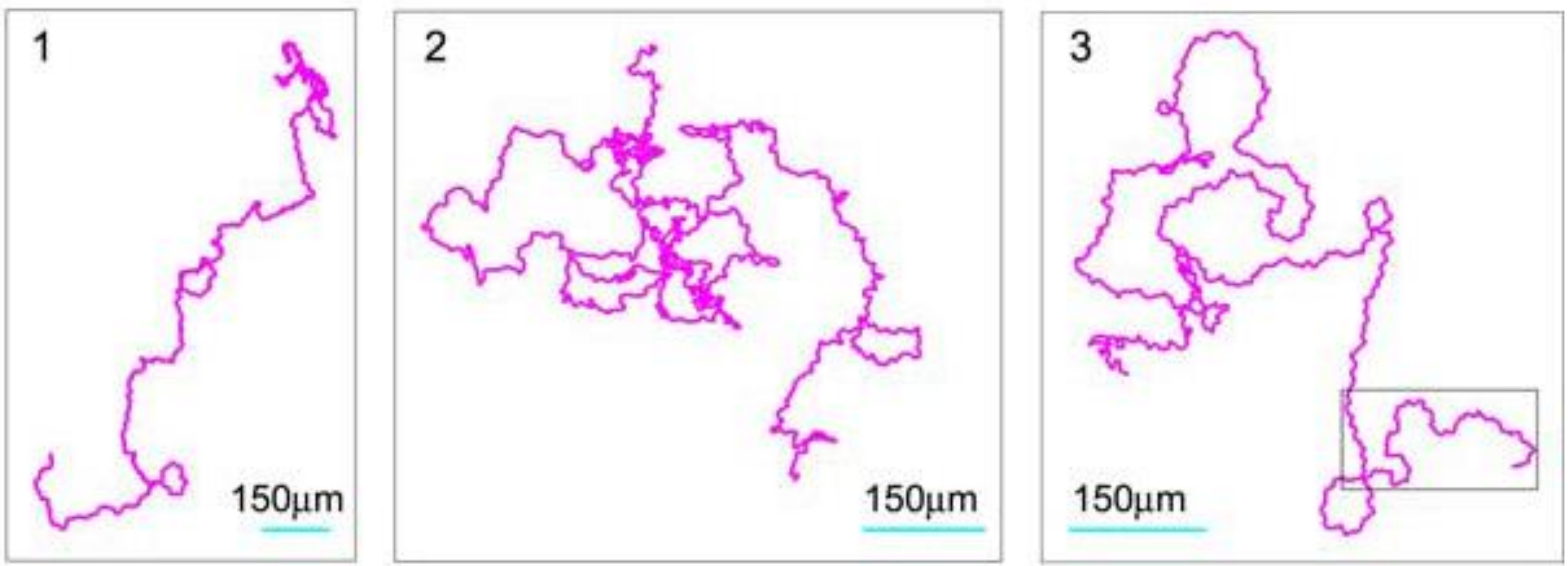


More directed movement?

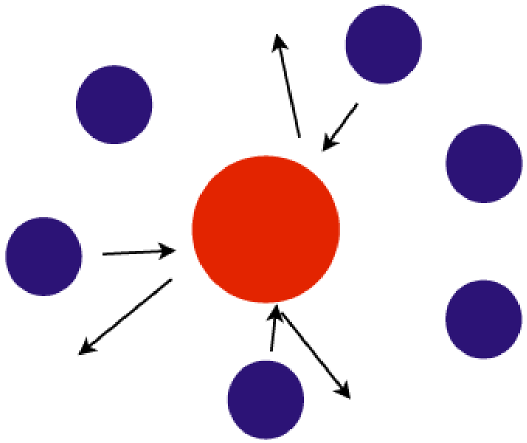


YES!
The time spent in the CCW ("run") state is longer

Migration of eukaryotic cells



Equation de Langevin



$$m\frac{d\mathbf{v}}{dt} = -\xi\mathbf{v} + \mathbf{f}(t)$$

Friction ξ

Force de Langevin

$$\langle \mathbf{f}(t)\rangle = \mathbf{0}. \quad \langle f_i(t) f_j(t')\rangle = A\delta_{ij}\delta(t-t')$$

Pas de corrélations: bruit blanc

Temps caractéristique $\tau = m/\xi$.

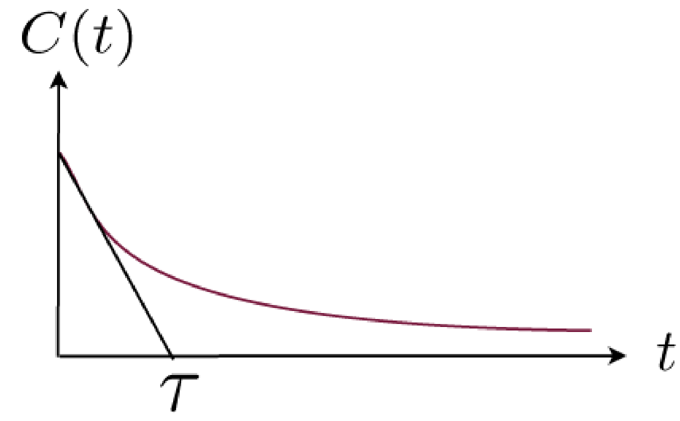
Vitesse:

$$\mathbf{v}(t) = \mathbf{v}_i e^{-t/\tau} + \int_0^t \frac{\mathbf{f}(t')}{m} e^{-(t-t')/\tau} dt'$$

Temps longs mouvement non biaisé: $\langle \mathbf{v}(t)\rangle = \mathbf{0}$

Corrélation des vitesses

$$C(t,t') \equiv \langle \mathbf{v}(t)\mathbf{v}(t') \rangle$$



On obtient (temps longs, pour d=3)

$$C(t,t') = \frac{3A}{2\xi m} e^{-|t-t'|/\tau}$$

Théorème d'équipartition de l'énergie (équilibre): $\frac{1}{2}m\langle \mathbf{v}^2 \rangle = \frac{3}{2}kT$

On déduit $A = 2kT\xi$

Déplacement quadratique moyen

$$\langle \mathbf{r}(t)^2 \rangle = \int_0^t dt_1 \int_0^t dt_2 \langle \mathbf{v}(t_1)\mathbf{v}(t_2) \rangle$$

On obtient (temps longs, pour d=3)

$$\langle \mathbf{r}(t)^2 \rangle \sim \frac{6kT}{\xi} t$$

coefficient de diffusion défini par $\langle \mathbf{r}^2 \rangle = 2dDt$

soit

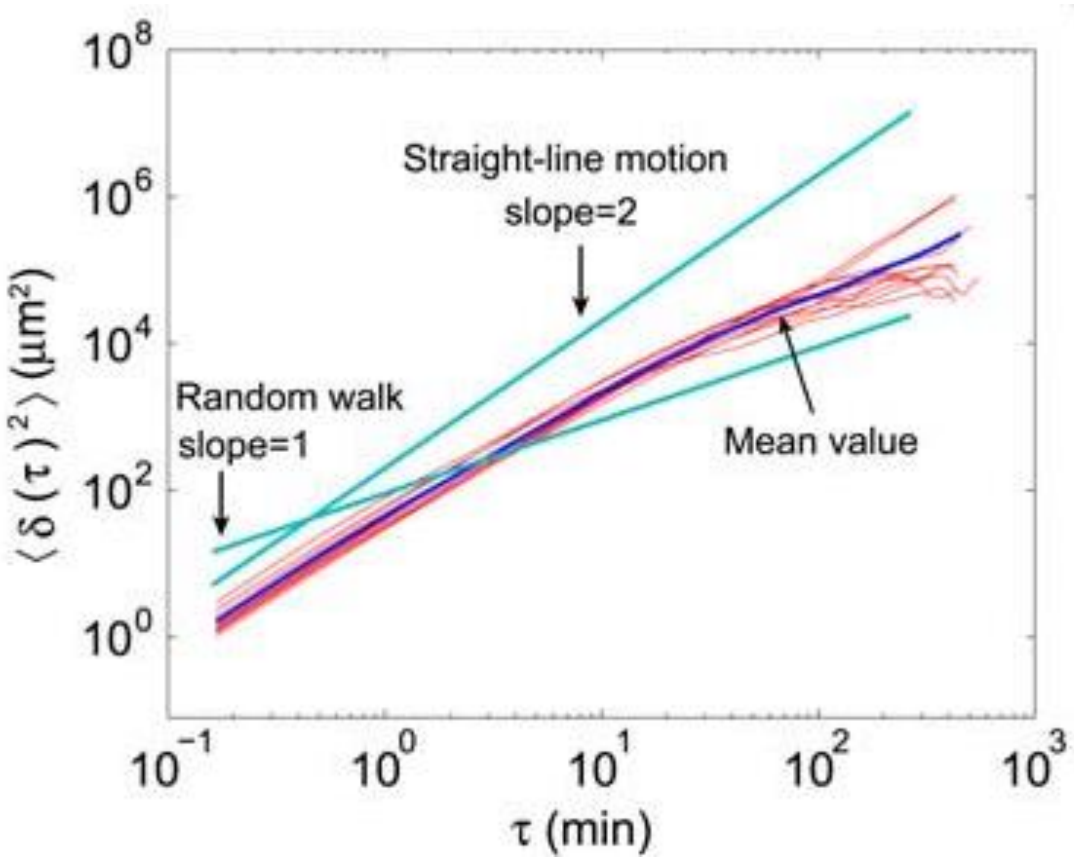
$$D = \frac{kT}{\xi}$$

Relation d'Einstein

Note: sans l'hypothèse « temps longs »

$$< r^2(t) > = 2D \left(t - D \, \frac{1 - e^{-\frac{v^2 t}{D}}}{v^2} \right)$$

Migration of eukaryotic cells



$$< r^2(t) > = 2D\left(t - D\,\frac{1 - e^{-\frac{v^2 t}{D}}}{v^2}\right)$$

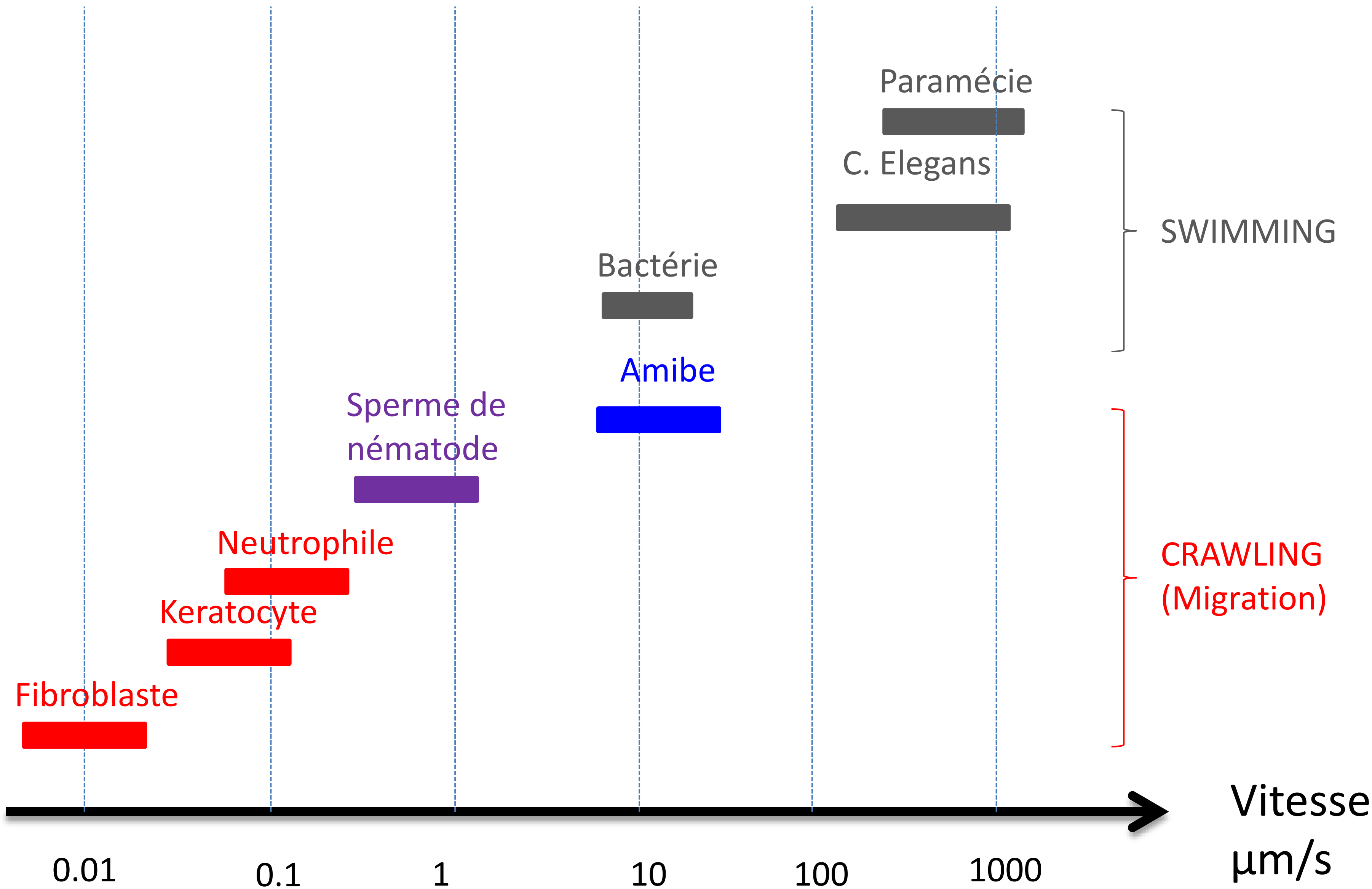
$$MSD(\tau) = < (\overrightarrow{r(t+\tau)} - \overrightarrow{r(t)})^2 >$$

Migration of eukaryotic cells

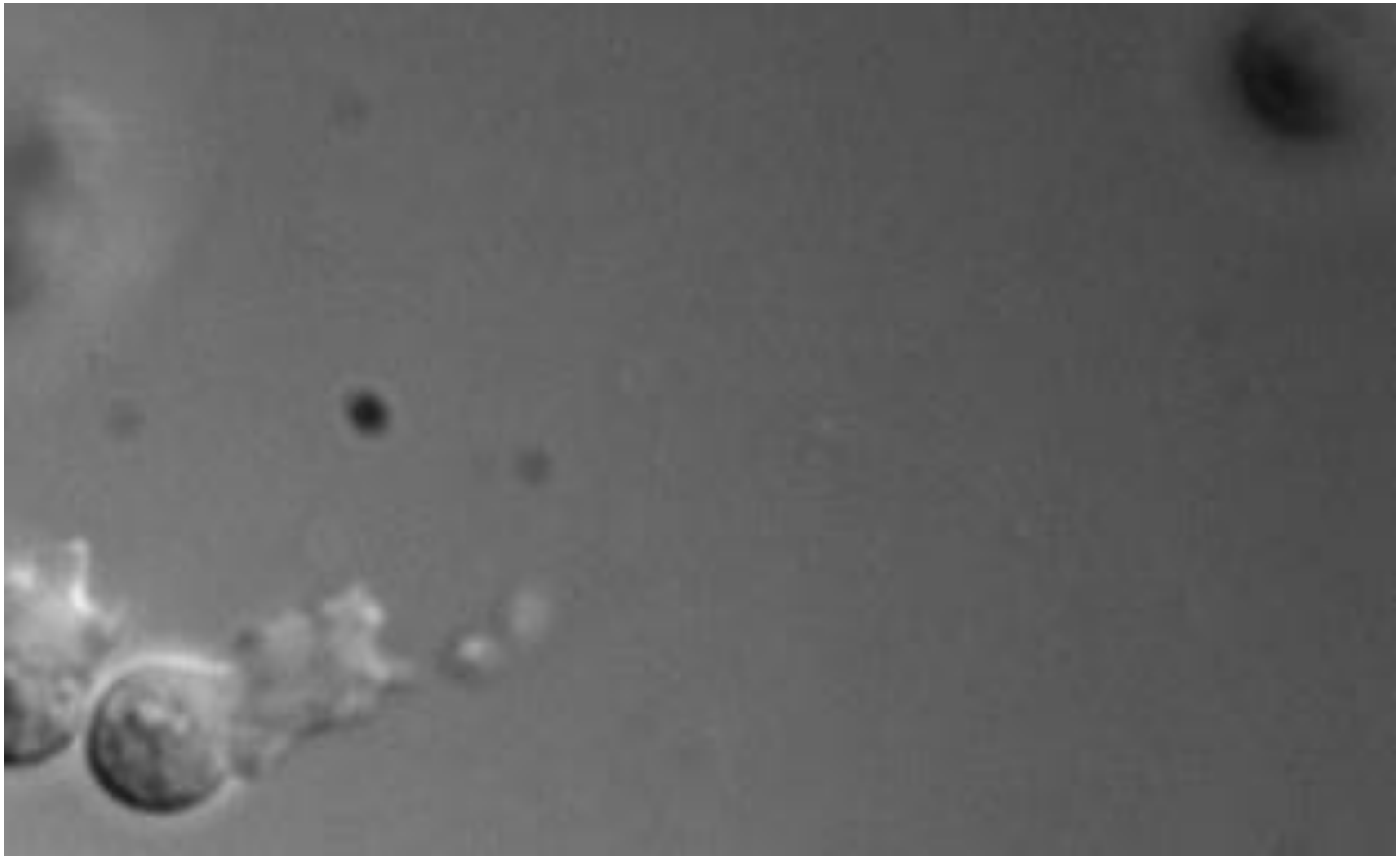
Typical Cell Speeds and Persistence Times			
Cell Type	P (min)	S ($\mu m/min$)	μ (cm^2/s)
Neutrophils	1-4	20	30 x 10-9
Macrophages	30	2	10 x 10-9
Fibroblasts	60	0.5	1.2 x 10-9
Endothelial Cells	300	0.4	
Smooth Muscle Cells	240-300	0.5	6.2 x 10-9
Neurons on laminin		1-3	

P=persistence time: Time before memory of initial direction is lost; S=speed of migration

μ =random motility coefficient

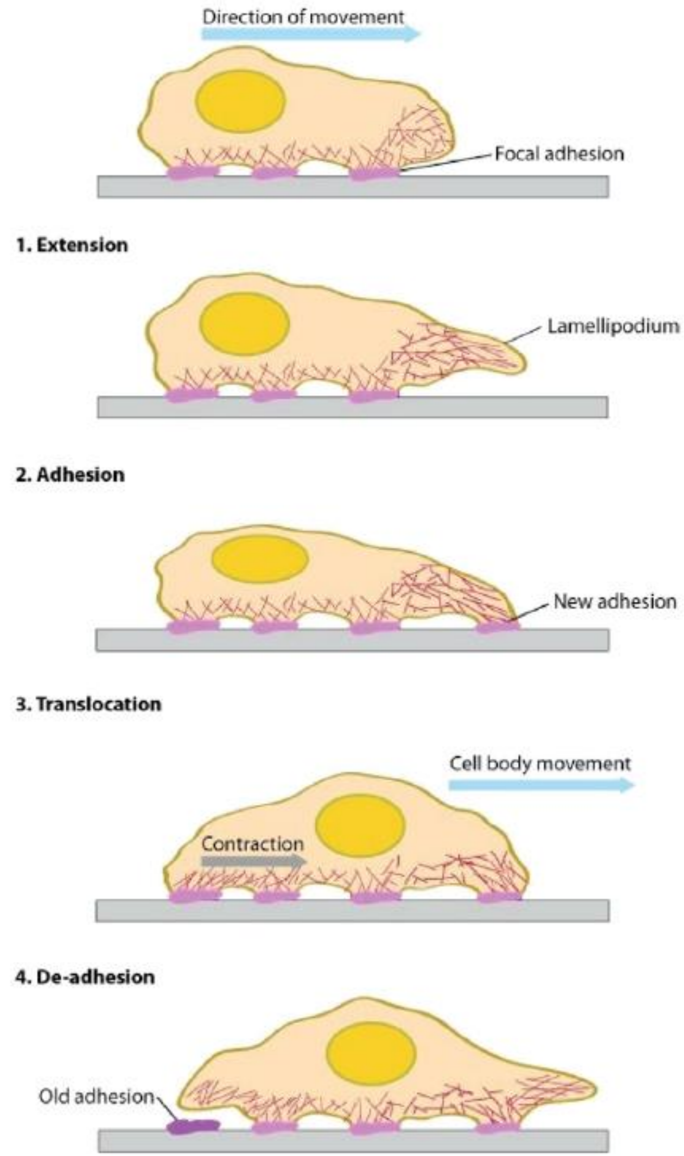

Paramécie
C. Elegans
SWIMMING
Bactérie
Amibe
Sperme de
nématode
Neutrophile
CRAWLING
(Migration)
Keratocyte
Fibroblaste
Vitesse
µm/s
0.01
0.1
1
10
100
1000

Keratocyte



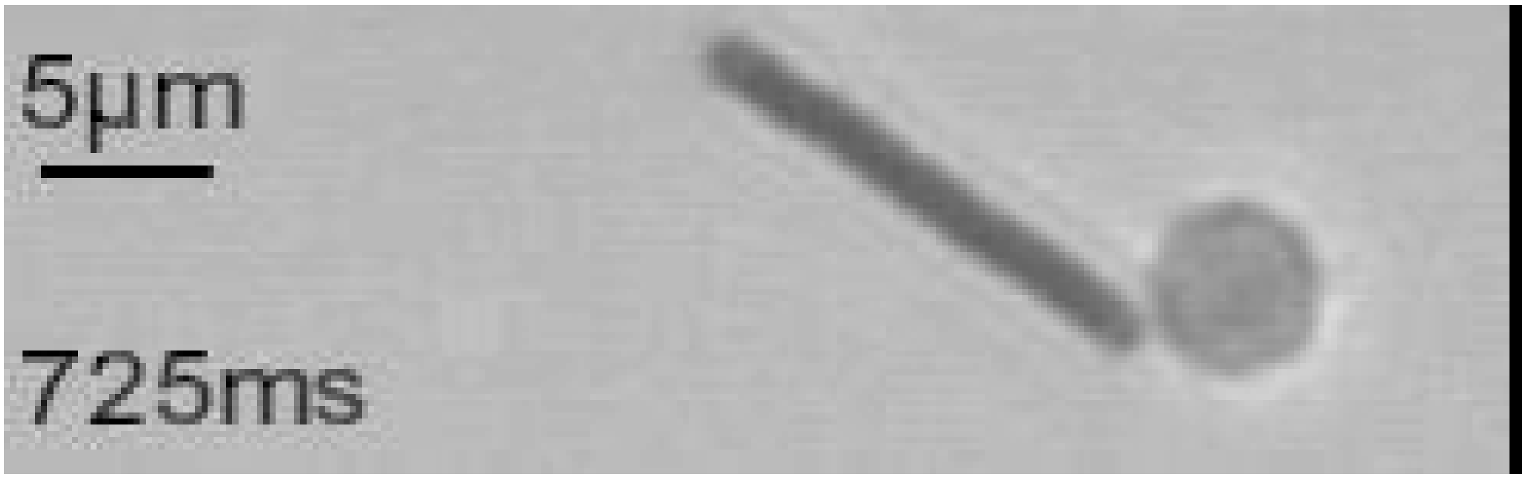
Worm cell

Schematic representation of 2D cell migration



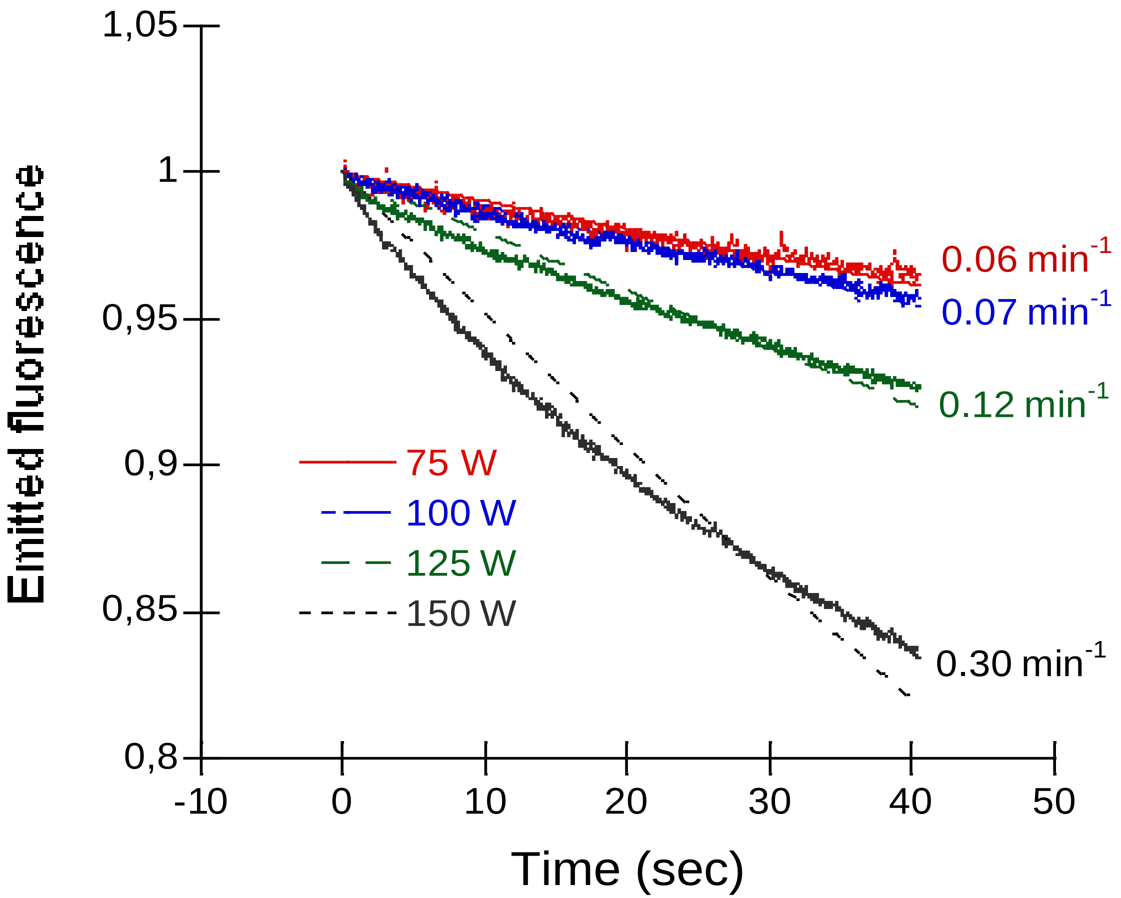
Other 'biological' swimmers
Other uses of flagella and cilia

Building artificial swimmers



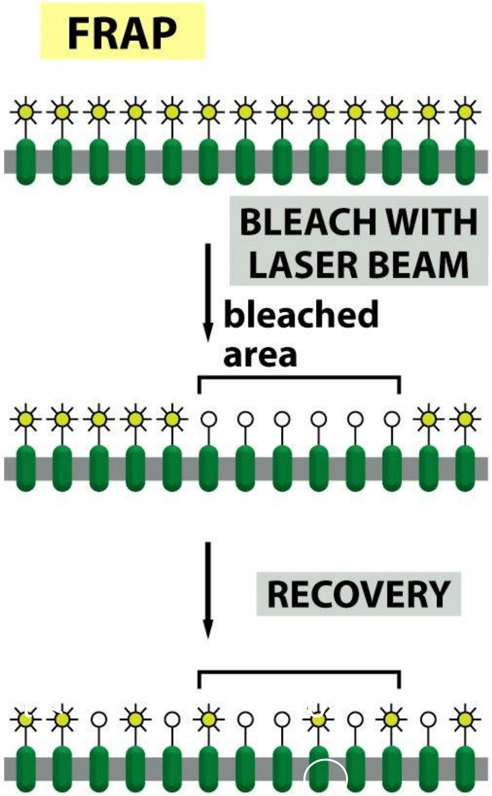
Photobleaching

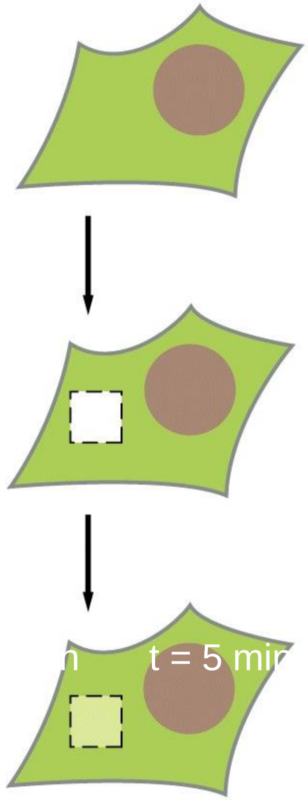
A fluorophore can perform a finite number of absorption/emission cycles, depending on its chemical structure and environment (30,000 for FITC).
Covalent deterioration due to oxidation.

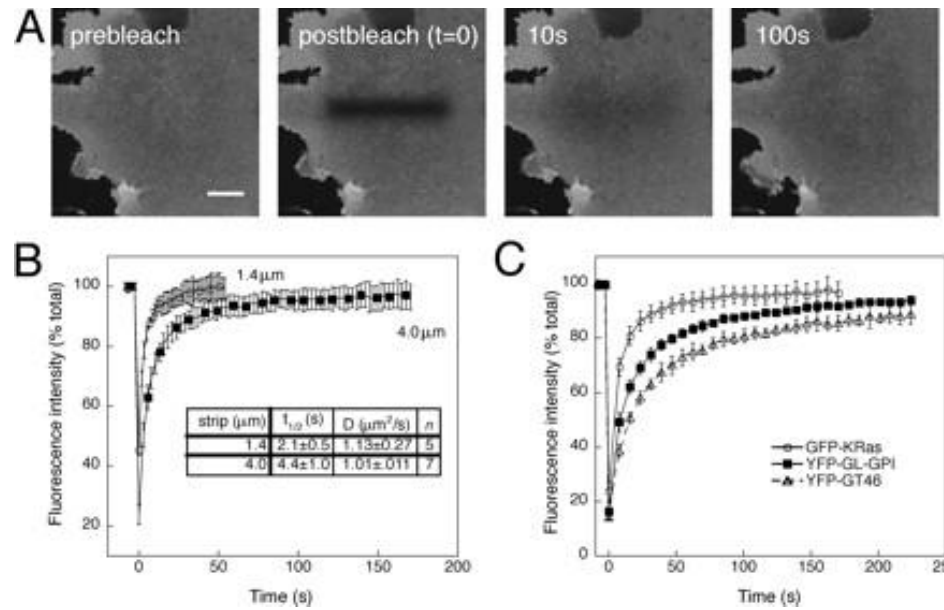


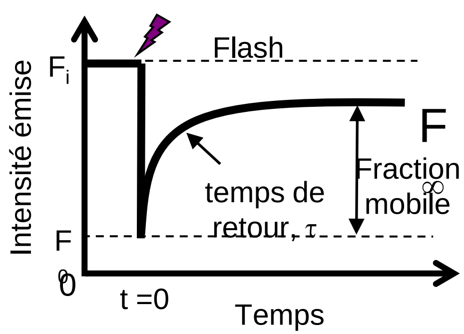
Measurement of diffusion coefficients

<u>Method 1</u>: FRAP Fluorescence Recovery after photobleaching





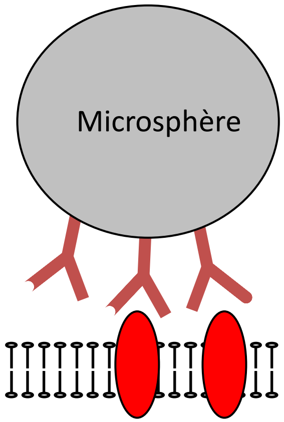




$$\tau_D = \omega^2/4D$$

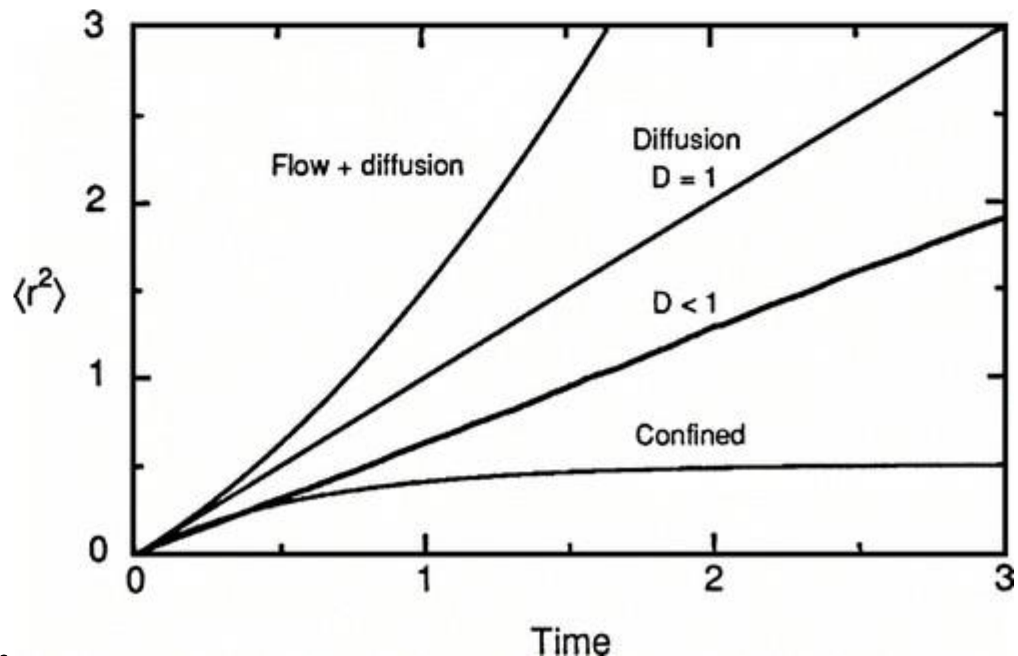
Measurement of diffusion coefficients

<u>Method 2</u>: Single particle tracking



Latex (50 – 500 nm)
Or colloidal
Quantum Dots

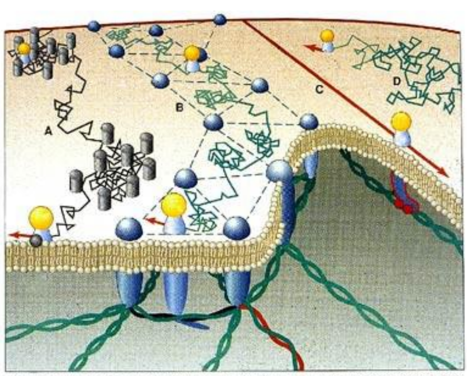
Anticorps ou ligand



Résolution spatiale: contraste interférentiel, fluorescence
Résolution temporelle: caméra rapide

Free:
MSD = 4 D t
Anomalous:
MSD = 4 D t^{α} (α<1)
Confined:
MSD = $L^2/3$ (1-exp(-12Dt/L^2))
Directed:
MSD = 4 D t + V^2 t^2

Mean squared displacement



Lateral transport modes on the cell surface. (A) Transient confinement by obstacle clusters (B) or by the cytoskeleton, (C) directed motion, and (D) free random diffusion