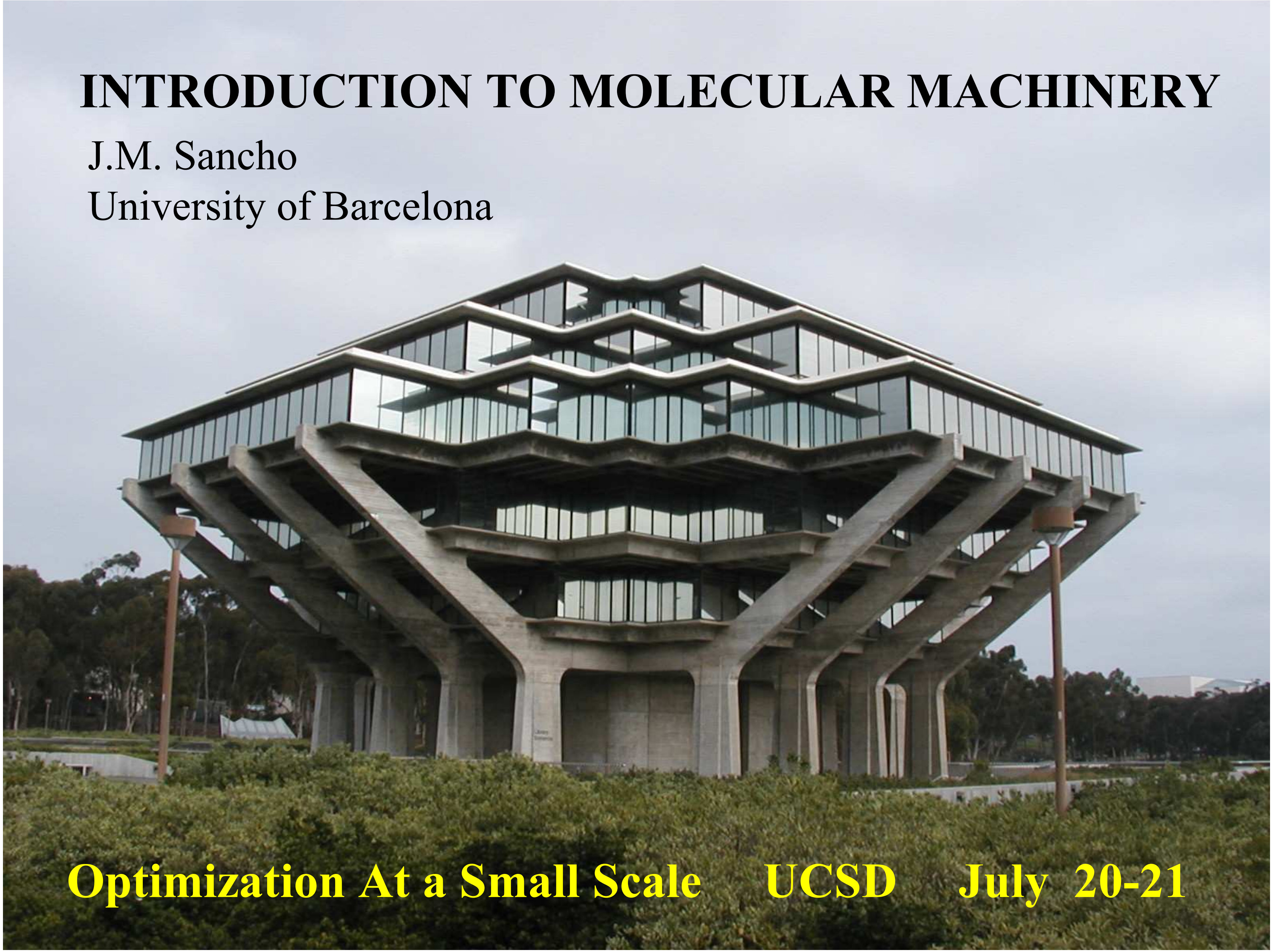


INTRODUCTION TO MOLECULAR MACHINERY

J.M. Sancho
University of Barcelona

Optimization At a Small Scale UCSD July 20-21

... and collaborators

Alex Ciudad ....”***A unified phenomenological analysis of the experimental data velocity curves in molecular motors***”
J. Chem. Phys. **128**, 225107(2008)

Alex Gómez-Marín... ***“Two-state flashing molecular pumps”,***
Europhysics Letters **86**, 40002(2009)

Javier G. Orlandi***...”Theoretical study of a membrane channel gated by ATP”,***
Eur. Phys. J. E, DOI 10.1140/epje/i2009-10483-9 (2009)

Rubén Pérez... ***“Theoretical analysis of the F1-ATPase experimental data”***
Preprint, 2009.....

Slides from:
“ ***Physical Biology of the Cell***” , R.Phillips et al... Garland (2009)

OUTLINE

1. INTRODUCTION
2. MOTORS
3. CHANNELS
4. PUMPS
5. CONCLUSSIONS

MOLECULAR MACHINERY

MOTORS:
- LINEAR: kinesin
- ROTATORY: BFM, Fo, F1..

TRANSPORTERS:
- PUMPS: Fo, Na-K ATPase
- CHANNELS: KscA, Cl(CFTR),..

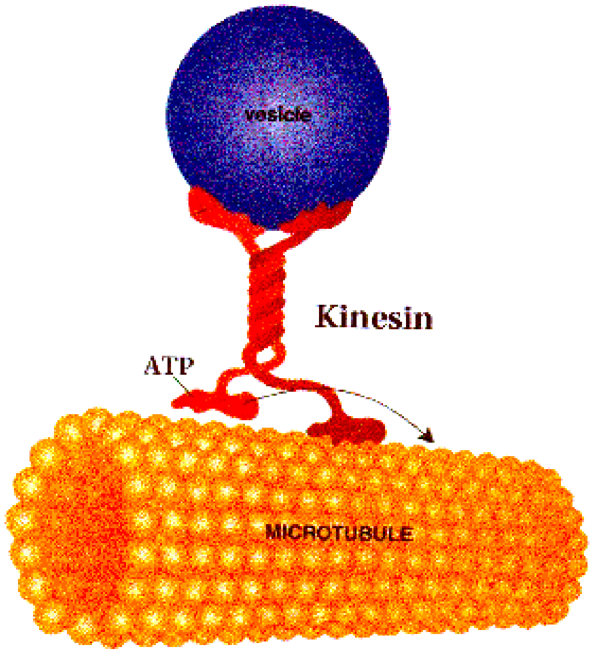
... and more: translocators, Polymerases, Ribosomes,...

and also artificial devices ...

EXAMPLES

LINEAR MOTORS

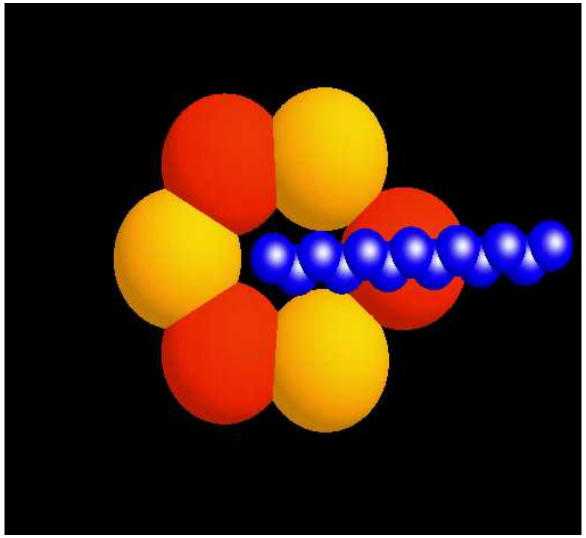
Kinesin



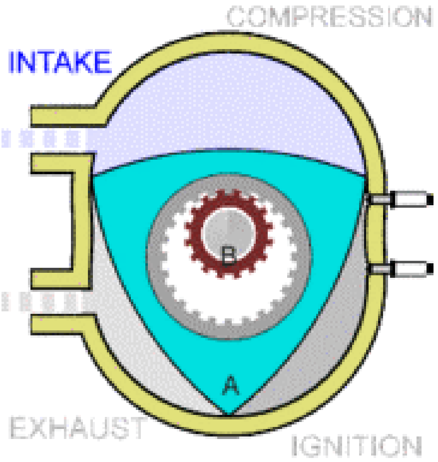
Maglev

ROTATORY MOTORS

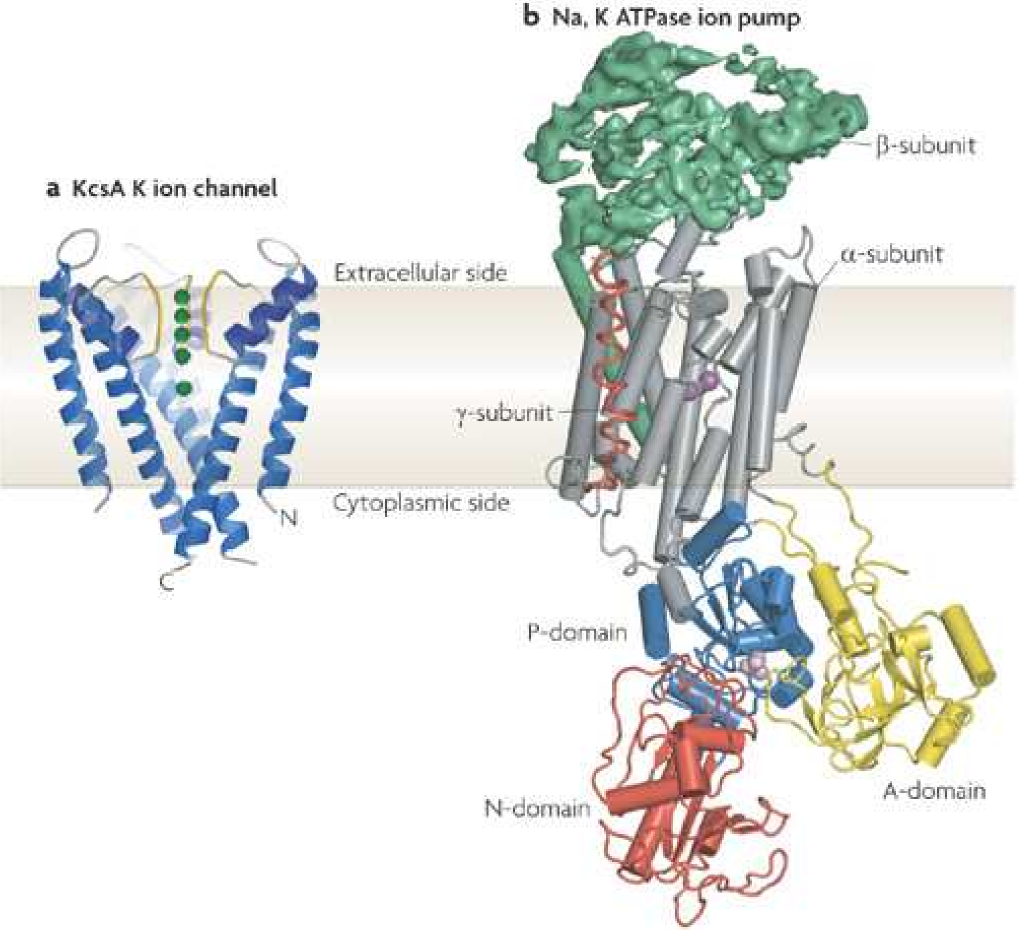
F1-ATPase

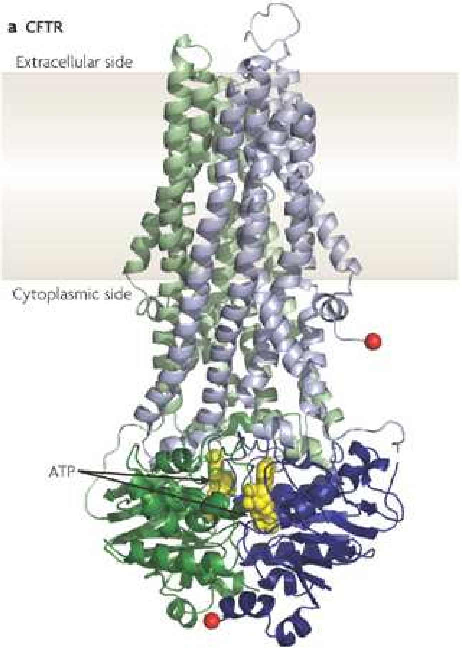


Wankel



K channel Na-K ATP pump Cl (CFTR) ATP channel

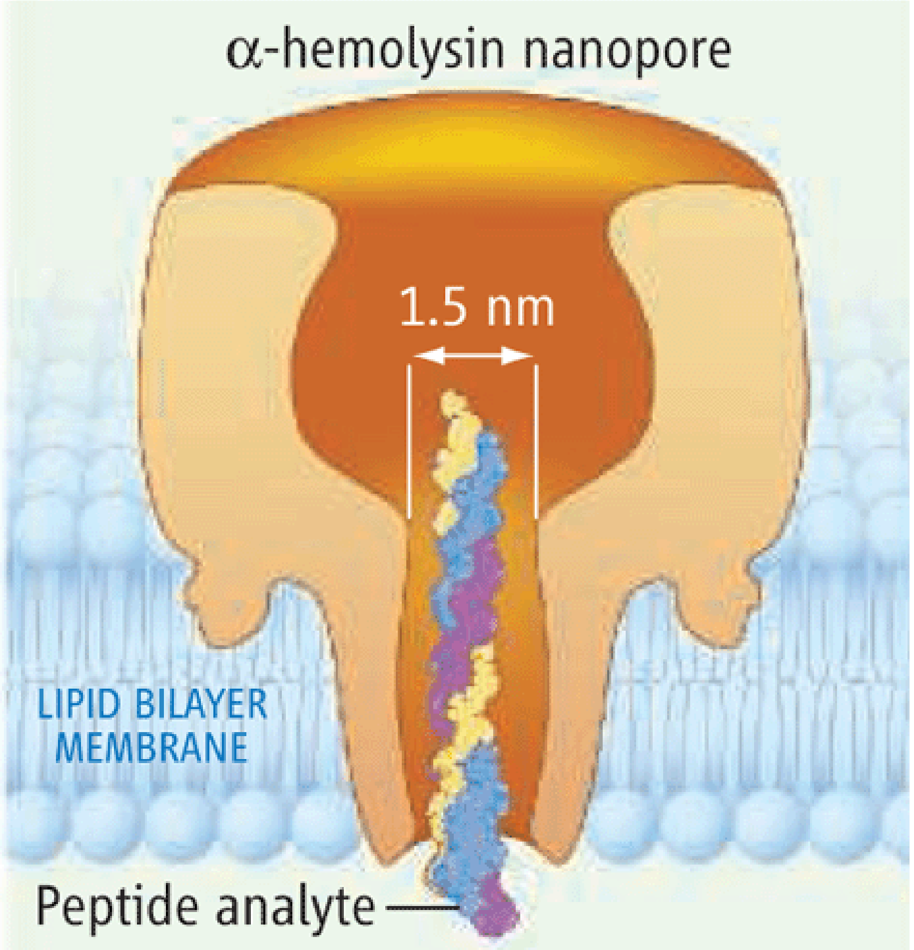




From D.C. Gadsby, Nature Reviews-MCB, 10, 344 (2009)

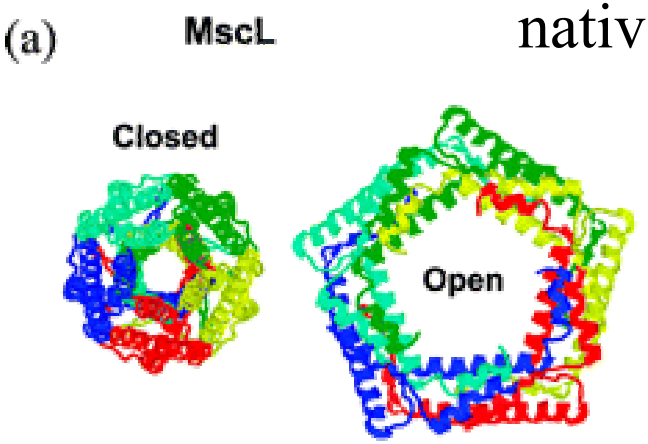
Channels

selectivity

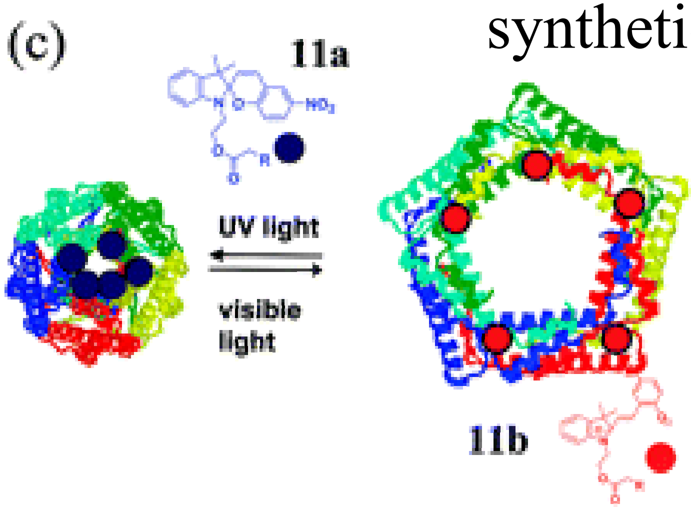


Martin & Siwy, SCIENCE **317**,3313(2007)

native



synthetic



Feringa, JOC Perspective **72**, 6635 (2007)

Channel

Pump

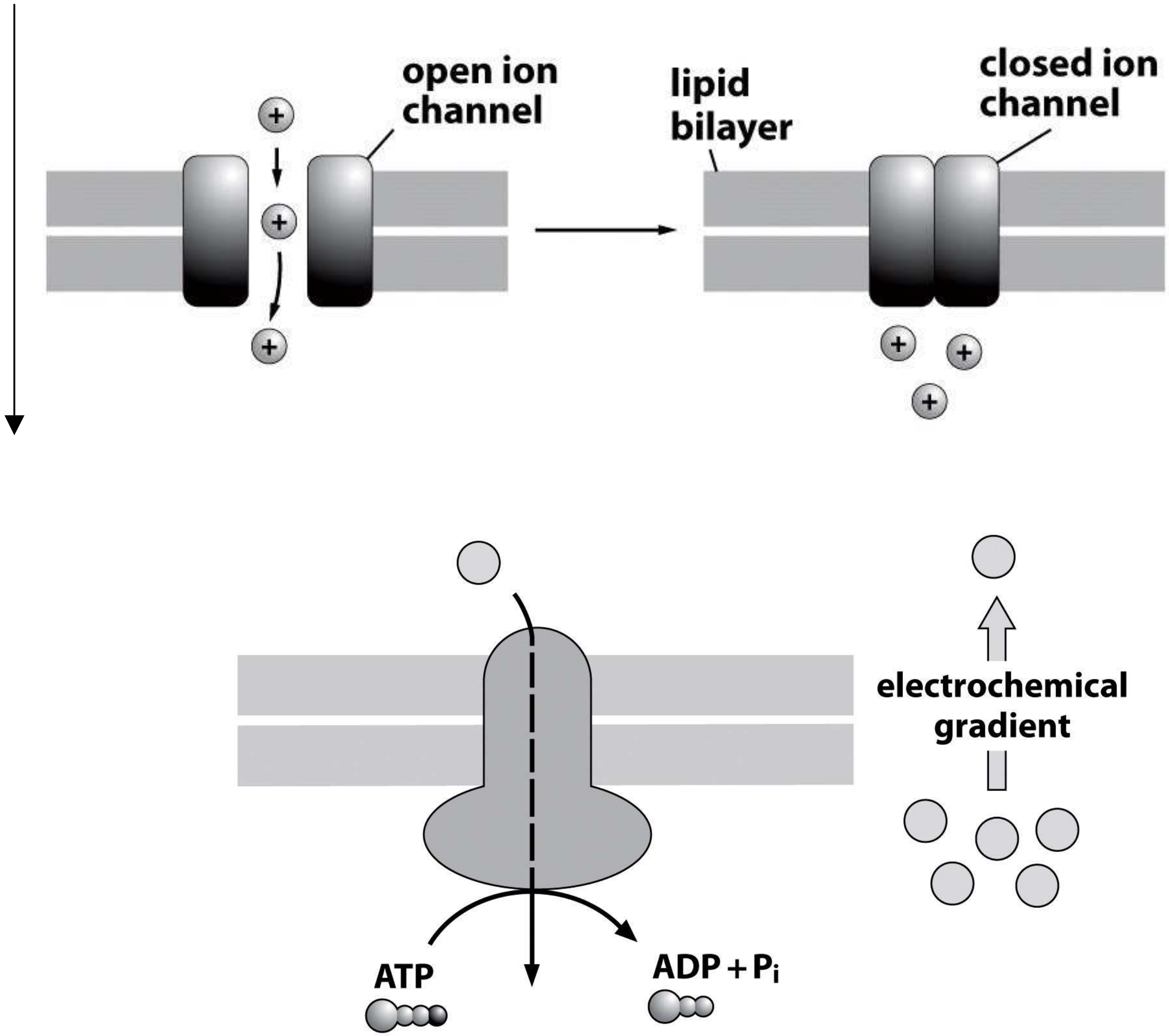
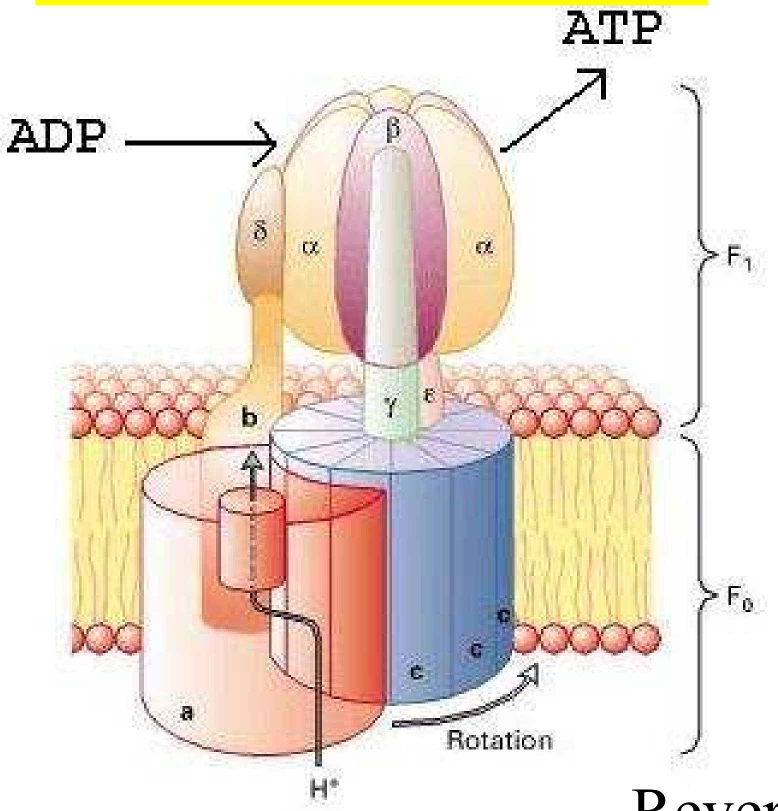


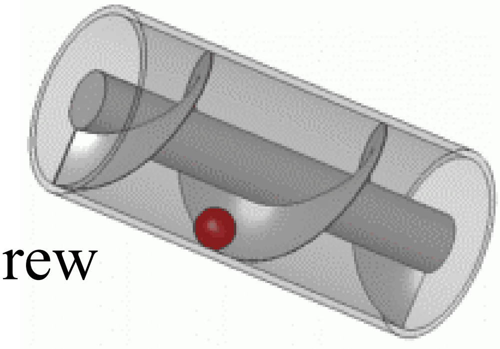
Figure 3.27 Physical Biology of the Cell (© Garland Science 2009)

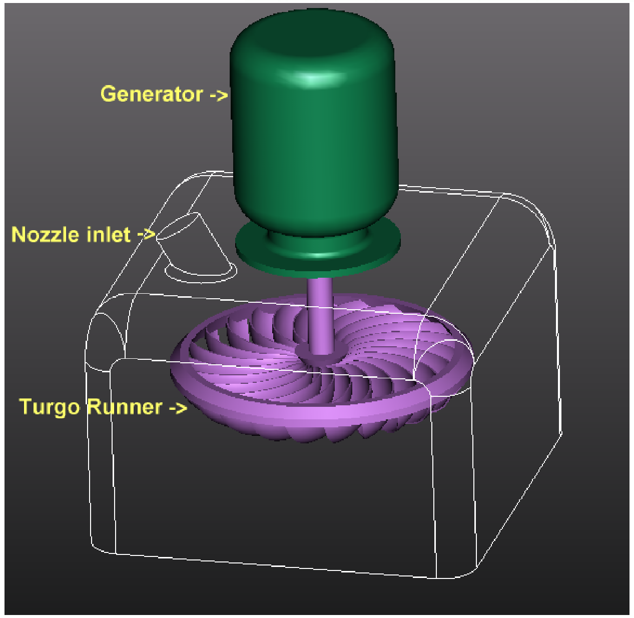
MIXED MOTOR AND PUMP

F0F1-ATP Synthase



Archimedes screw





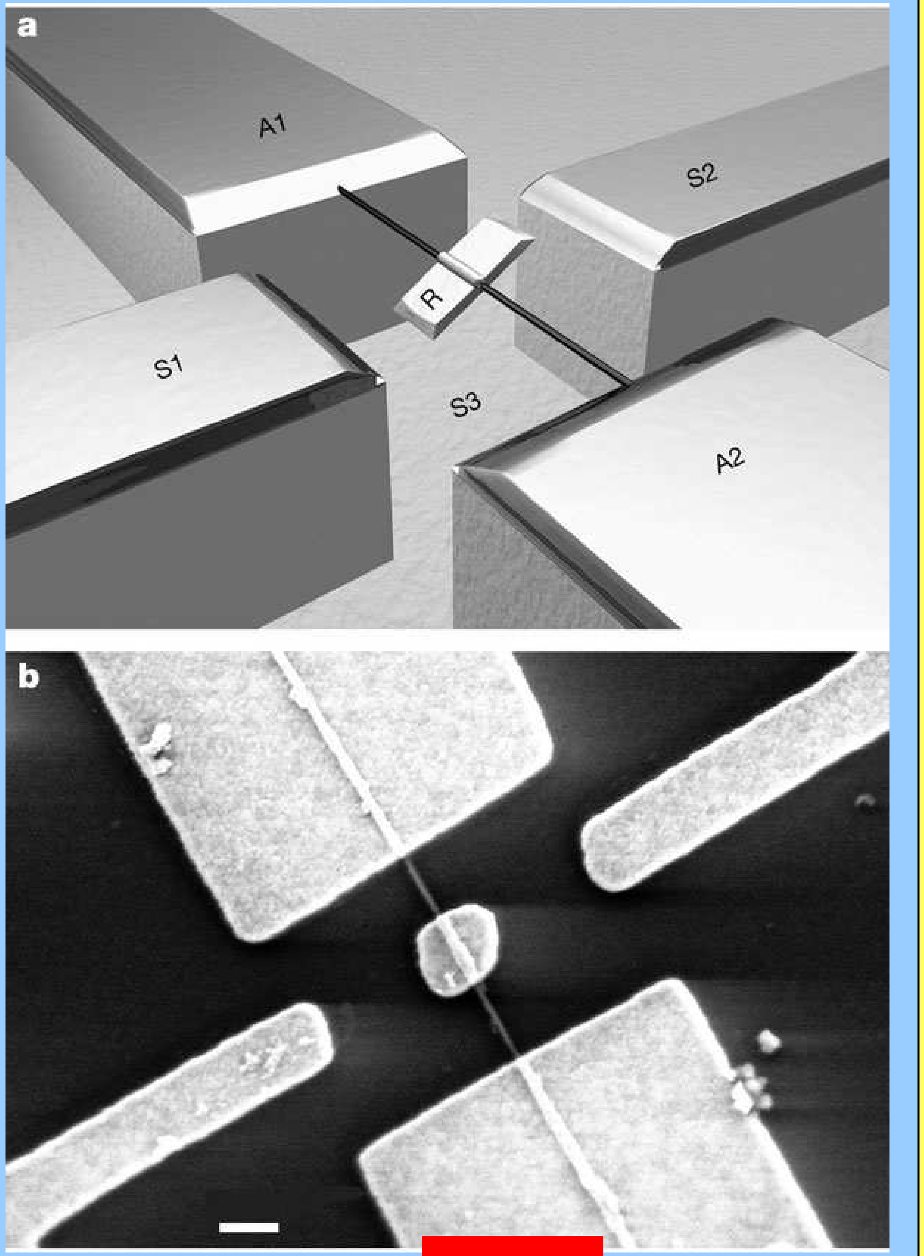
Reversible engine

ATP hydrolysis → proton pump

ATP synthesis ← proton flux

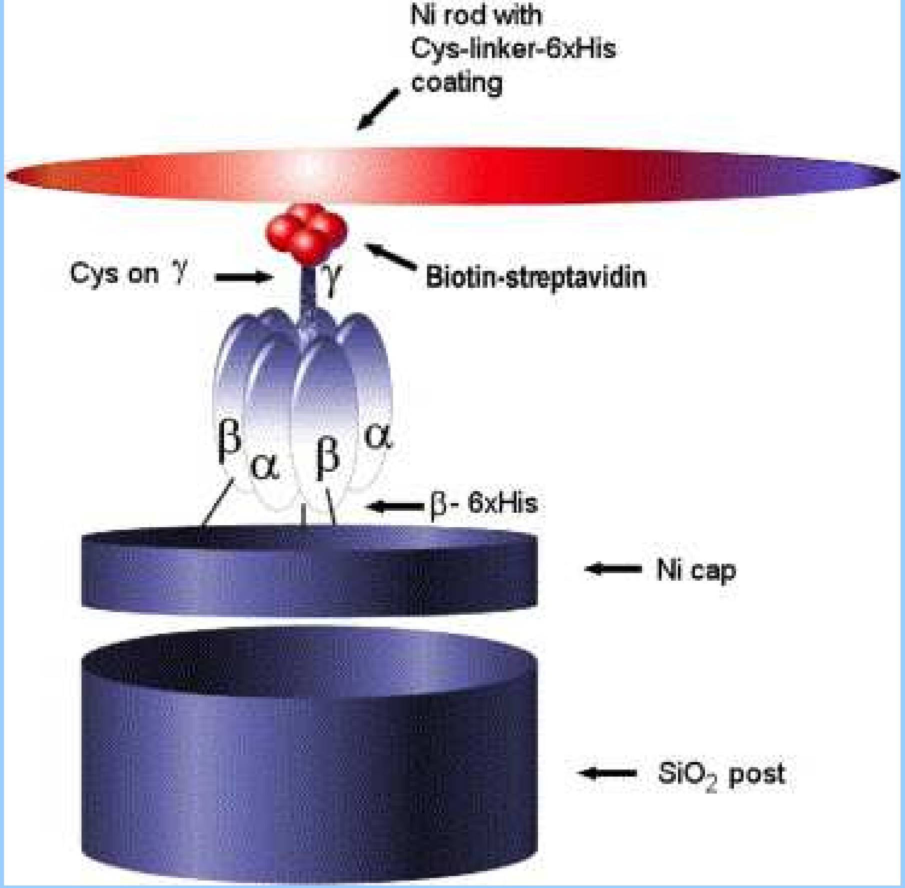
Nano-devices

Fennimore,et al.
Nature **424**, 408 (2003)

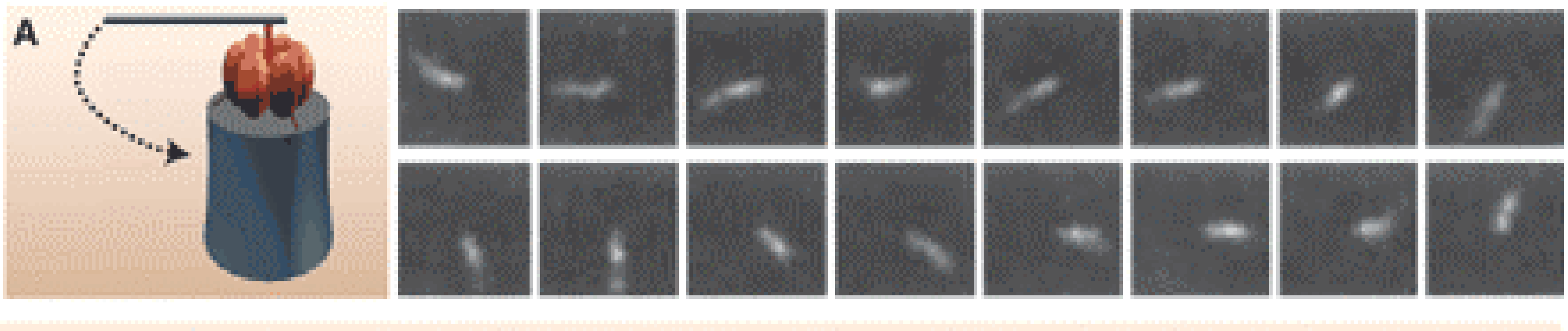


video

Mixed nanomotors: Rotatory. F1-ATPase

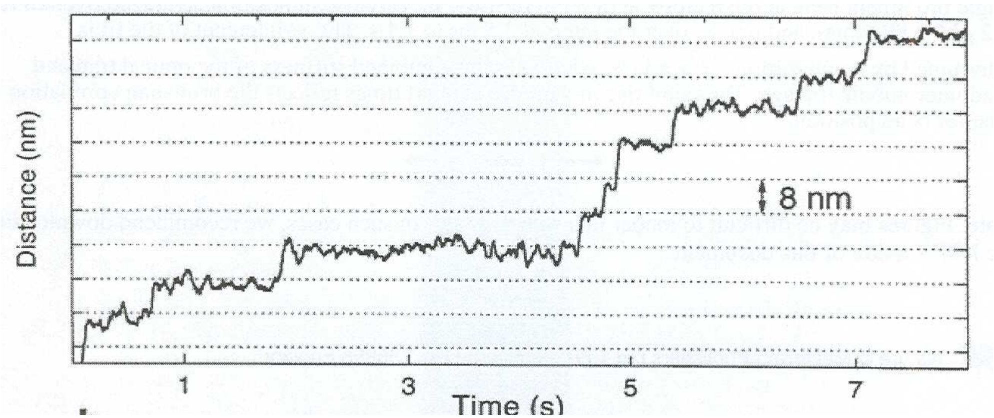


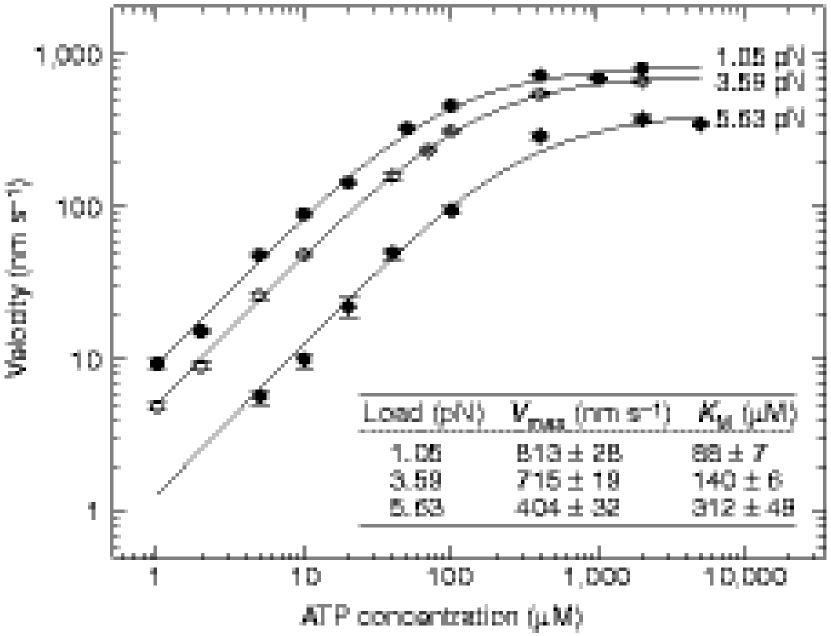
Song et al.,
Science **290**, 1555 (2000)



Levels of analysis

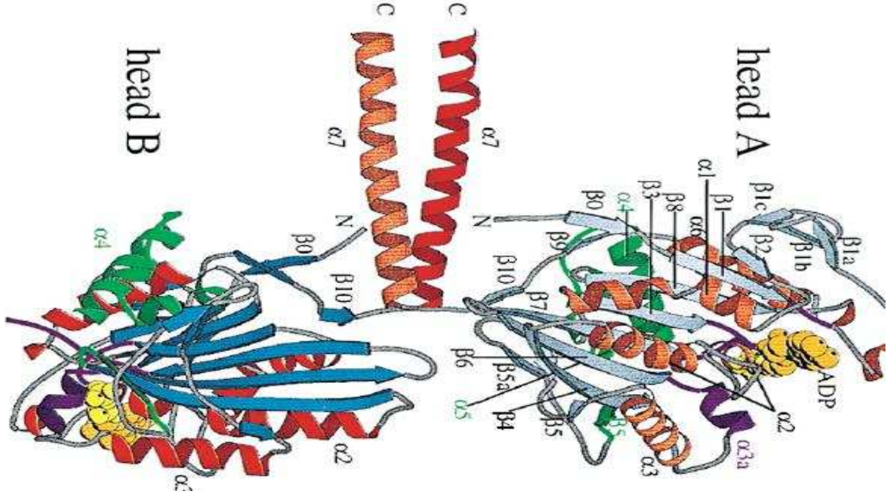
Experimental data: trajectories and mean observables



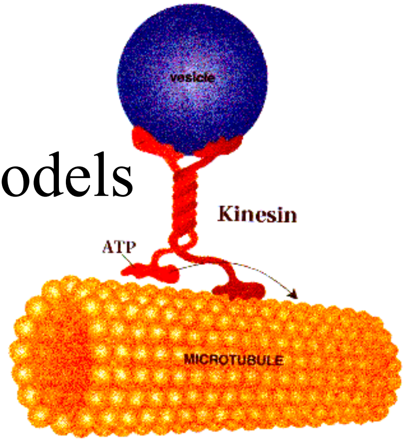


Load (pN)	V_{max} (nm s^{-1})	K_M (μM)
1.05	813 ± 28	88 ± 7
3.59	715 ± 19	140 ± 6
5.63	404 ± 32	312 ± 49

Structure and function



Mesoscopic models



Aim: A theoretical scenario for experimental data

Input or
control parameters:

[ATP] concentration
ATP-energy
Conservative forces: F...
Particle densities
Electrochemical potential
Friction

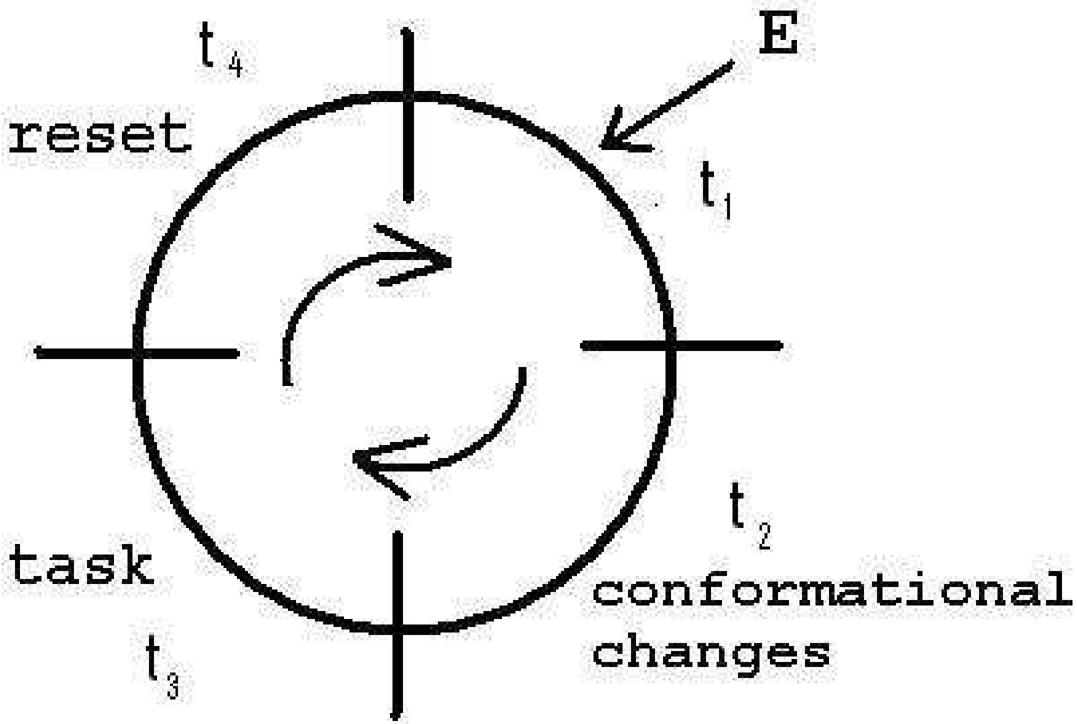
Outputs or observables:

Trajectories
Spatial steps
<v>
Randomnes
Power
Efficiency

$$v([ATP], F)$$

$$J([ATP], \Delta\phi, \rho_{in}, \rho_{out})$$

The starting point: the CYCLE



$$v = \frac{L_0}{t_1 + t_2 + \ldots + t_n}$$

Physical and chemical elements

$$E_{in} \left| \begin{array}{l} \Delta G_{ATP} \sim 20 k_B T \sim 90\, pN.nm \sim 51\, meV \\ smf = -\Delta\phi + k_B T \ln \frac{[Na]_{in}}{[Na]_{ex}} \end{array} \right.$$

Motive force $$F_m = \frac{E_{in}}{L_0}$$

Conservative force $$F_{ex}$$

Times: chemical (waiting and internal) and mechanical times.

Internal time → t_0 Waiting time → $t_0 \frac{k}{[ATP]}$

M-M

MOTORS

Scales: $\Delta G_{ATP} \sim 90\,pN.nm \rightarrow F \sim pN; L_0 \sim nm$

Mechanical time

$$(\gamma_0 + \gamma_L)\frac{L_0}{t_{mech}} = F_m - F_{ex}$$

Analytical formula for motors

$$v = \frac{L_0}{t_0\left(1+\frac{k}{[ATP]}\right)+\frac{L_0(\gamma_0+\gamma_L)}{F_m-F_{ex}}}$$

Too many free parameters!

Kinesin: $t_0 = 0.1\,ms$ $\qquad L_0 = 8\,nm \rightarrow v \sim 800 nm/s$

CHANNELS

FLUX : diffusion down the gradient
driven by the membrane potential

GATING: open and close.
ATP hydrolysis
V membrane potential
density (M-M)
others

$$J = P_{open} * J_{free}$$

FLUX: Free open channel

$$\frac{\partial \rho}{\partial t} = D \frac{\partial^2 \rho}{\partial x^2}\,, \qquad \rho(0) = \rho_0\,,\ \rho(L) = \rho_1$$

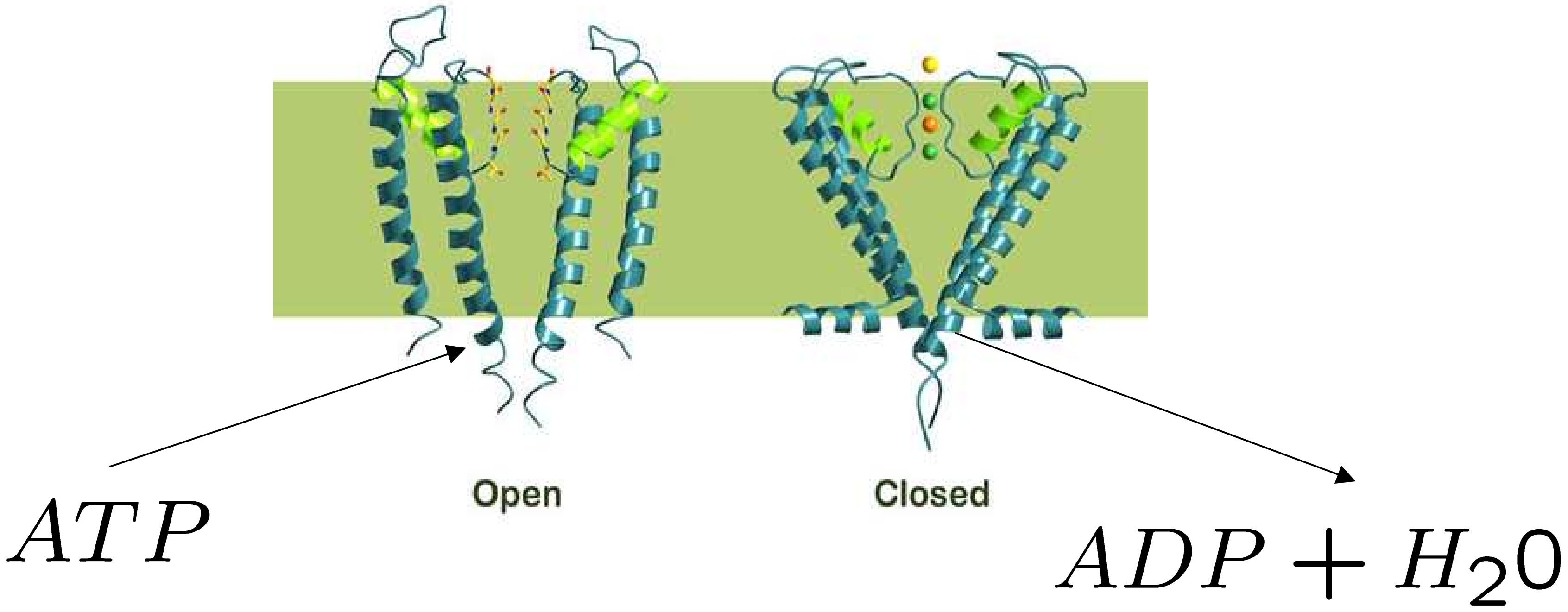
$$J = -\frac{AD}{L}\,(\rho_1 - \rho_0) \qquad \rho_0 > \rho_1$$

$$Cl^-\,,\ A = 0.2\,nm^2\,,\ L = 5\,nm\,,\ \Delta\rho = 0.1\,M$$

$$D = \frac{K_B T}{6\pi r_o \eta} = 1.1\,10^9\,nm^2/s$$

$$J \sim 6.10^6\,ions/s \sim 0.4\,pA$$

ATP gated channel → two states : open and closed.



ATP

$ADP + H_2 0$

ATP-binding

$$\omega_1 = \omega_0 \frac{[ATP]}{k_M + [ATP]}$$

ATP-Hydrolysis

$$\omega_0$$

$$J_{ATP} = -\frac{\sigma}{1+2\sigma}\frac{DA}{L}\left(\rho_1 - \rho_0\right)$$

$$\sigma = \frac{[ATP]}{k_M}$$

Free channel with potential membrane

$$J = \frac{AD}{L}\bar{v}\,\frac{\rho_0-\rho_1 e^{-\bar{v}}}{1-e^{-\bar{v}}}\,, \qquad \bar{v} = \frac{\Delta\phi}{k_B T}$$

Goldman-Hodkin-Katz eq.

Voltage gated channel: Na

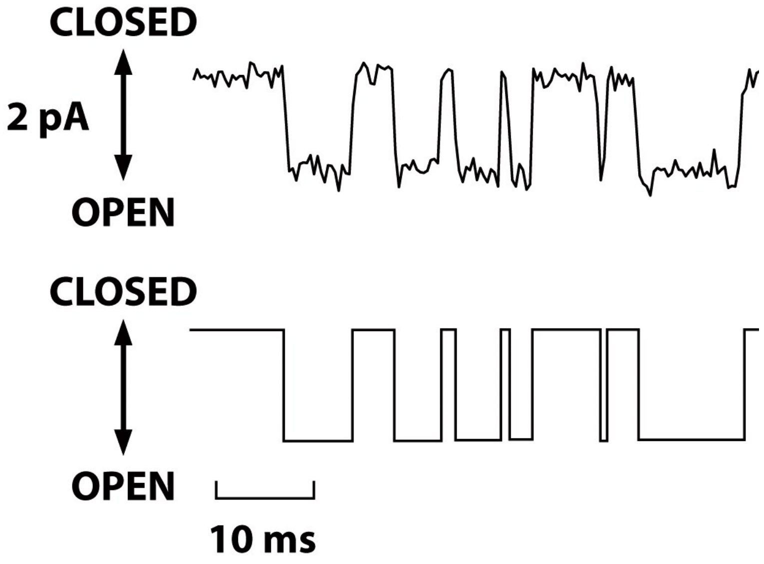


Figure 7.2a Physical Biology of the Cell (© Garland Science 2009)

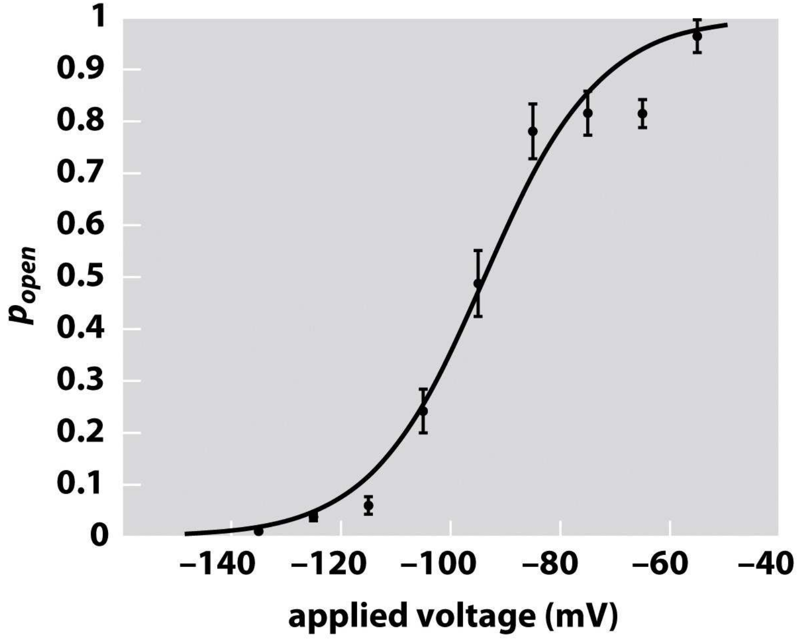
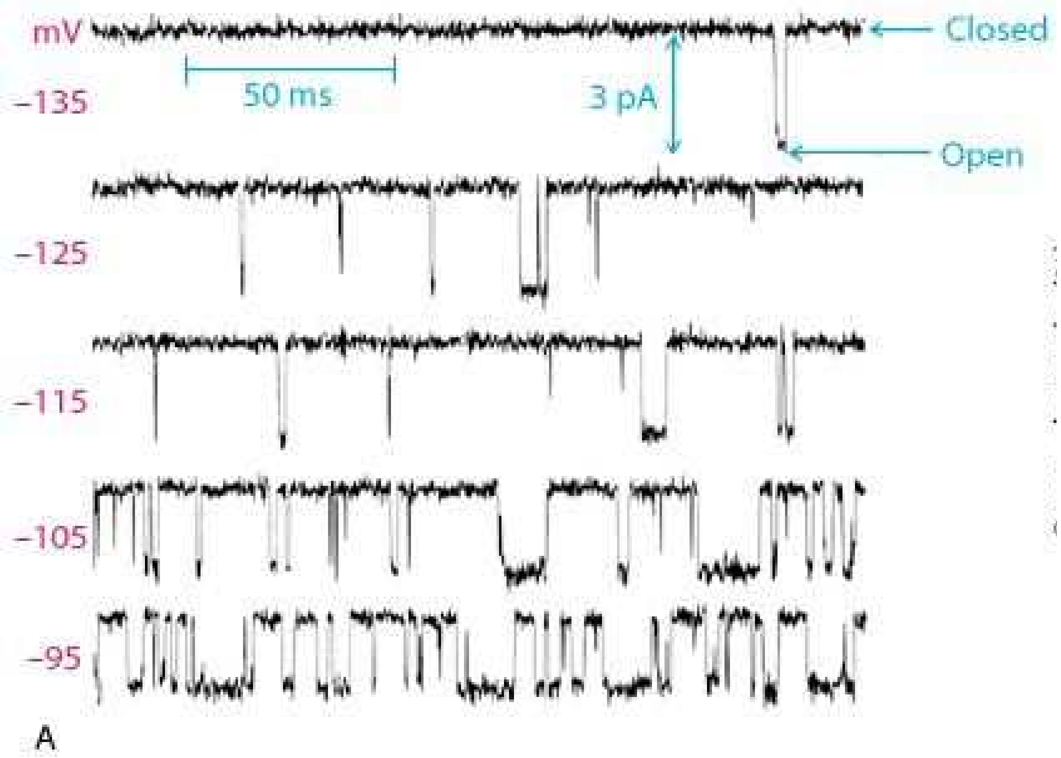


Figure 7.2c Physical Biology of the Cell (© Garland Science 2009)

Na reconstructed channels,
from Hartshorne et al. PNAS**82**,240(1985)



PUMPS

Particle flux is against gradient

power in > power out

$$\frac{\Delta G_{ATP}}{t_{ATP}} > J\mu$$

$$\frac{\rho_1}{\rho_0} = 10 \rightarrow \mu = k_B T \ln 10 = 2.3 k_B T/particle$$

$$t_{ATP} = 1ms$$

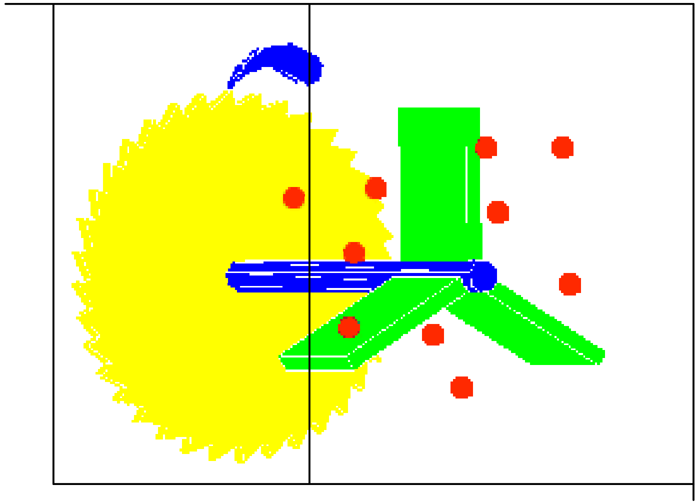
$$J < 10^3\, ions/s = 0.16\, 10^{-3}\, pA$$

WHAT ELSE ?

Put a ratchet in your model!

Overdamped Langevin equation
Two states ratchet potential
ATP dependent transitions
Energy constrains

Feynman, 1963



$T_1 < T_2$

2. MOTORS

Bacterial Flagellar Motor (BFM)

Chien-Jung, et al. Bioph. J. 93, 294 (2007)

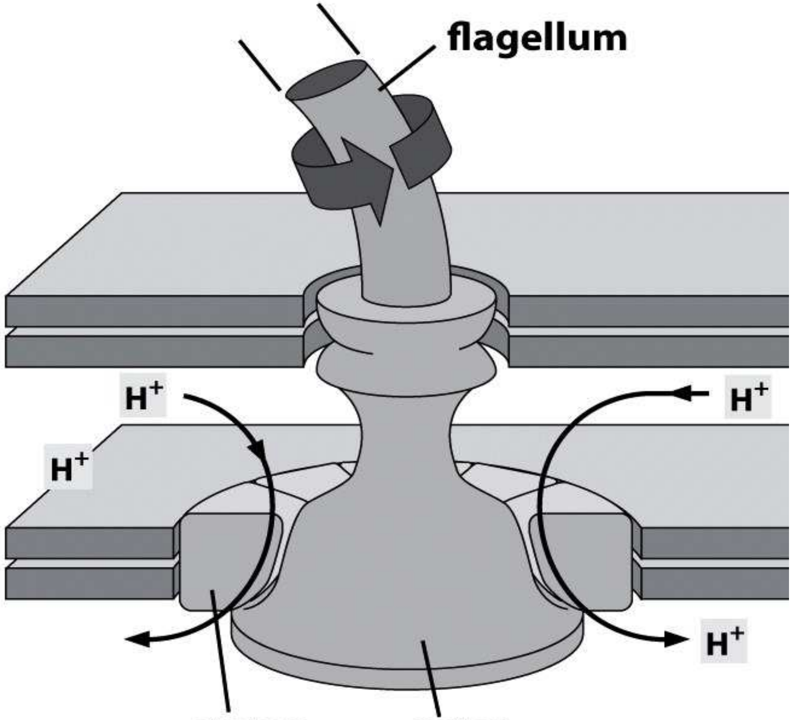
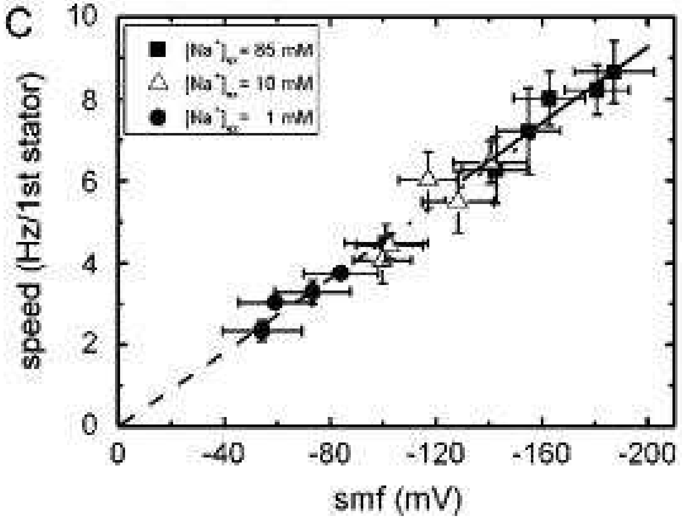


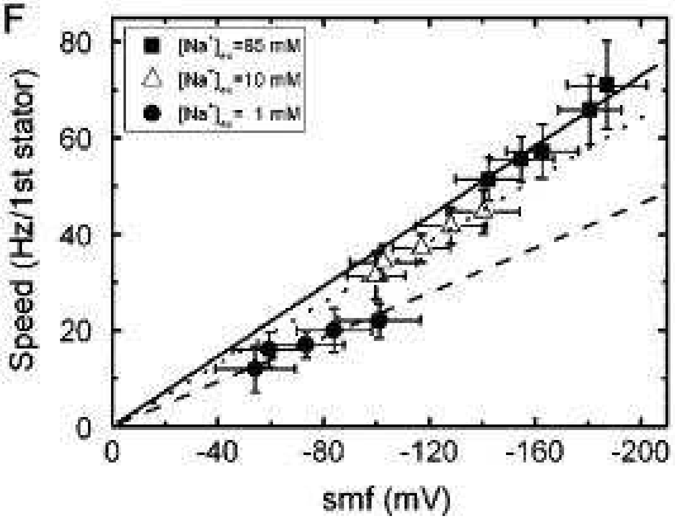
Figure 16.13a Physical Biology of the Cell (© Garland Science 2009)

$$smf = -\Delta\phi + k_B T \ln \frac{[Na]_{in}}{[Na]_{ex}}$$

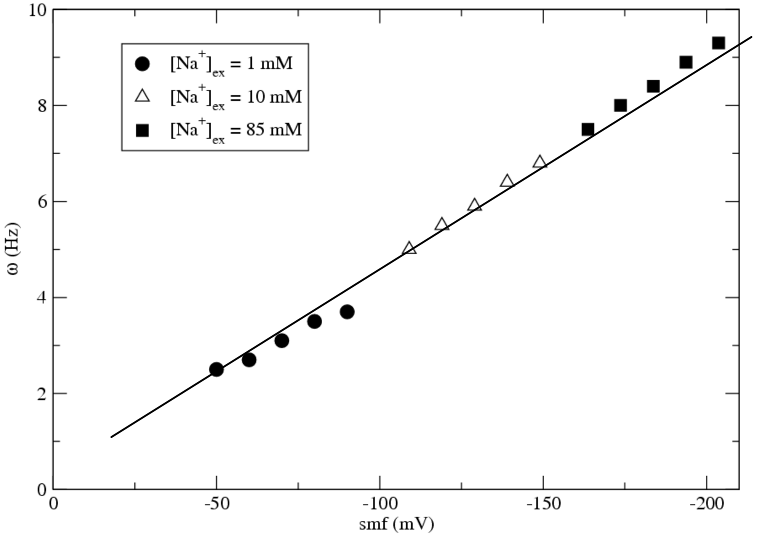
$$\omega = \frac{1}{t_0\left(1+\frac{k}{[Na]}\right)+\frac{\gamma_0+\gamma_L}{-smf}}$$

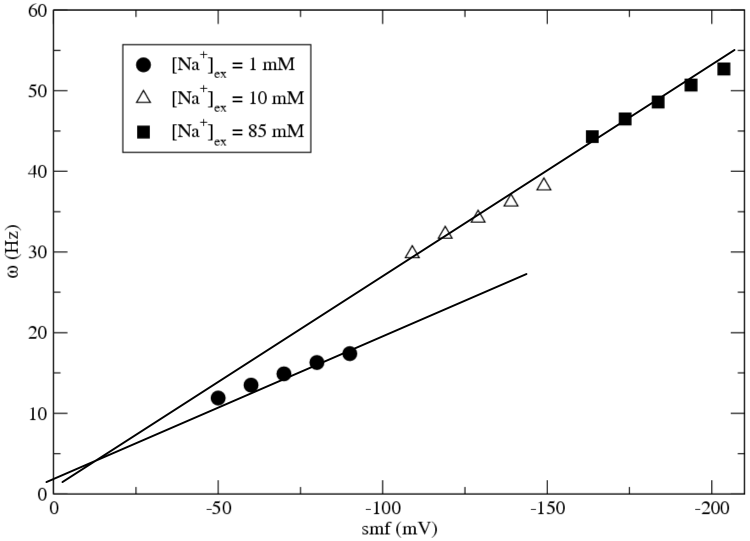
Experiment $1\mu m$ $0.35\mu m$





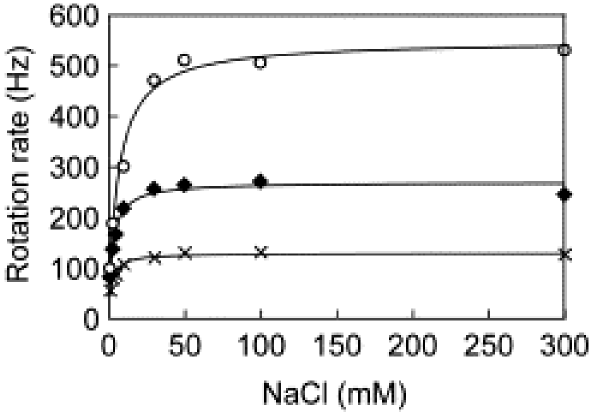
Theory

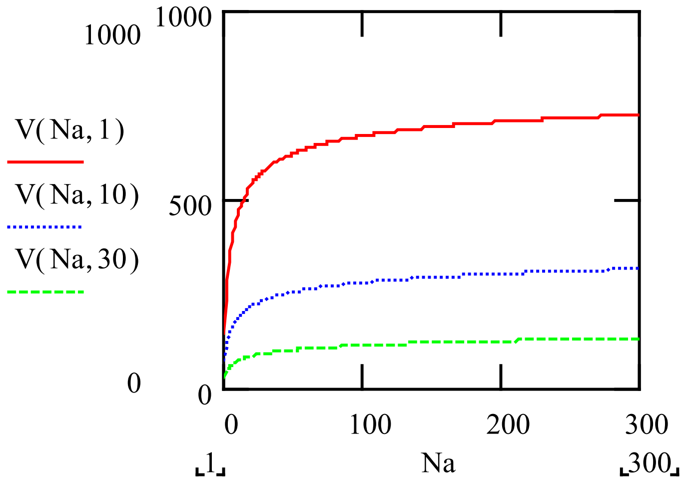


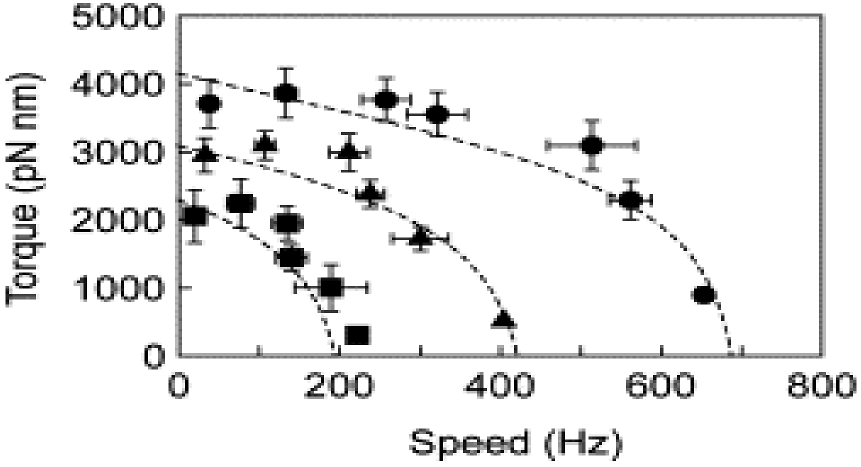


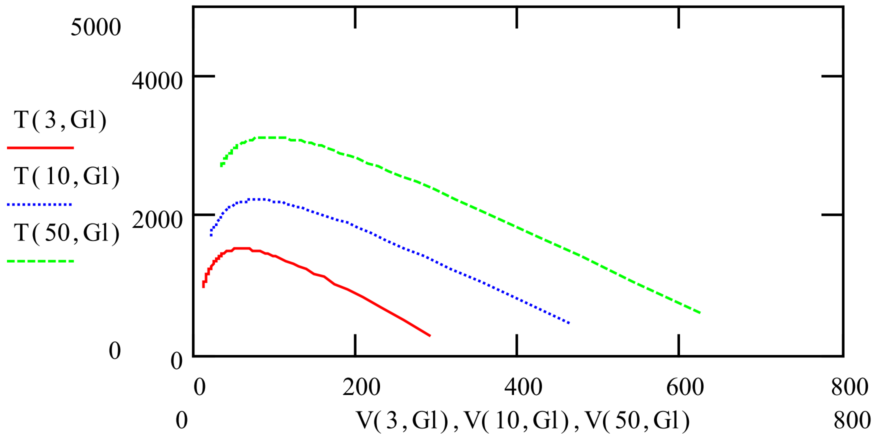
Sowa et al, JMB **327**, 1043(2003)

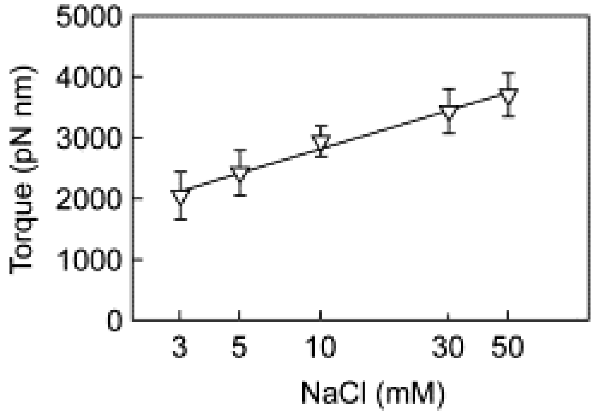
$$\omega = \frac{L_0}{t_0\left(1+\frac{k}{[Na]}\right)+\frac{(\gamma_0+\gamma_L)L_0^2}{-smf-b*R}}$$

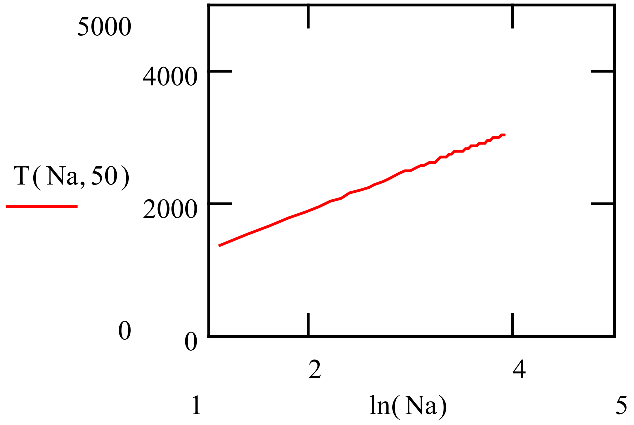





5000
4000
3000
2000
1000
0
Torque (pN nm)
0
200
400
600
800
Speed (Hz)

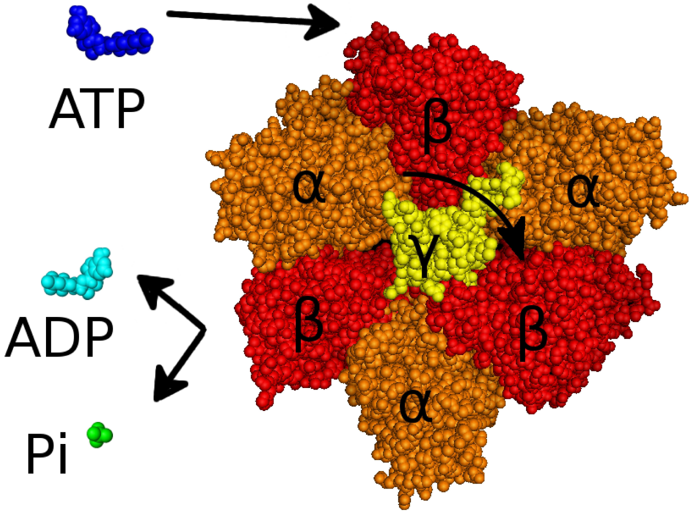

5000
4000
2000
0
0
T(3 , Gl)
T(10 , Gl)
T(50 , Gl)
0
200
400
600
800
0
V(3 , Gl) , V(10 , Gl) , V(50 , Gl)
800

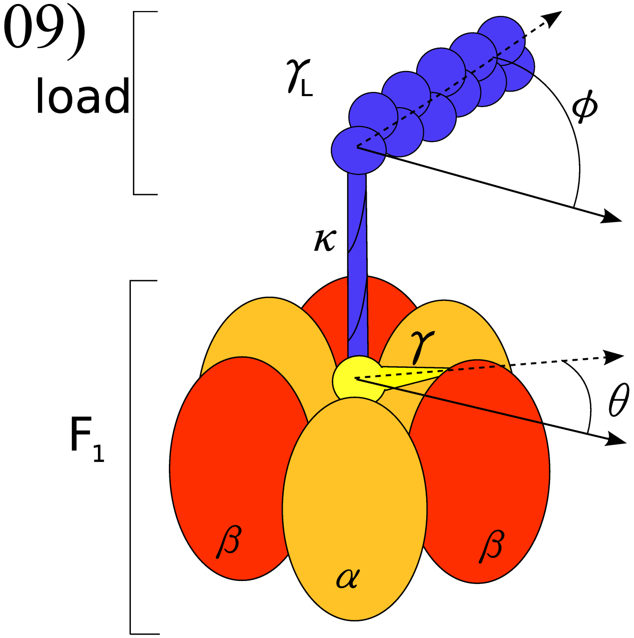

5000
4000
3000
2000
1000
0
Torque (pN nm)
3
5
10
30
50
NaCl (mM)


5000
4000
2000
0
0
T(Na, 50)
2
4
1
ln(Na)
5

F1-ATPase

Theoretical analysis of the F1-ATPase experimental data, R. Perez-Carrasco & JMS, preprint (2009)

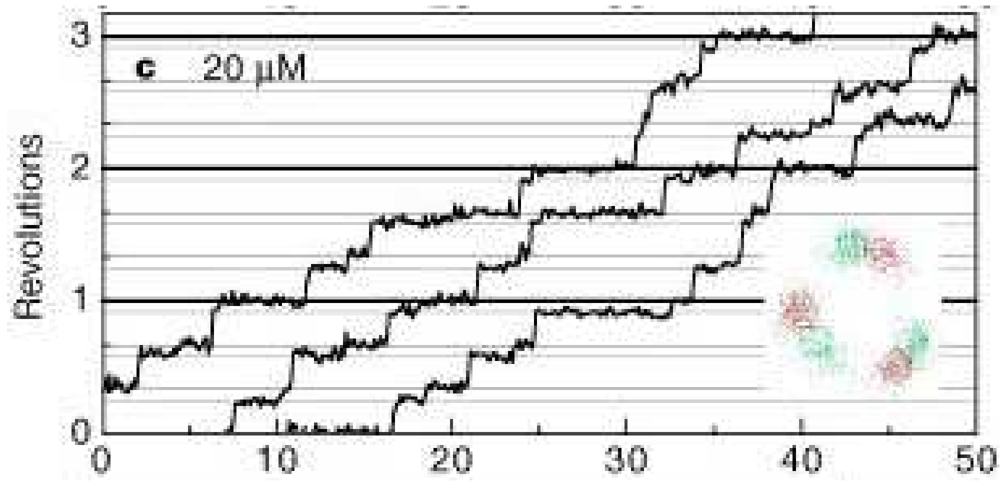




Observable: load angle $\phi(t) \longrightarrow \omega$

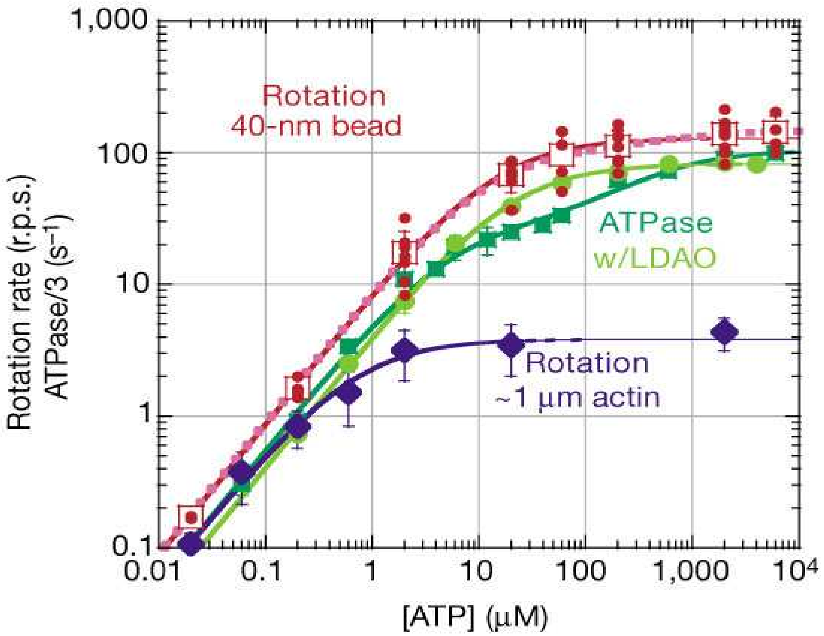
Parameters: load friction γ_L and [ATP]

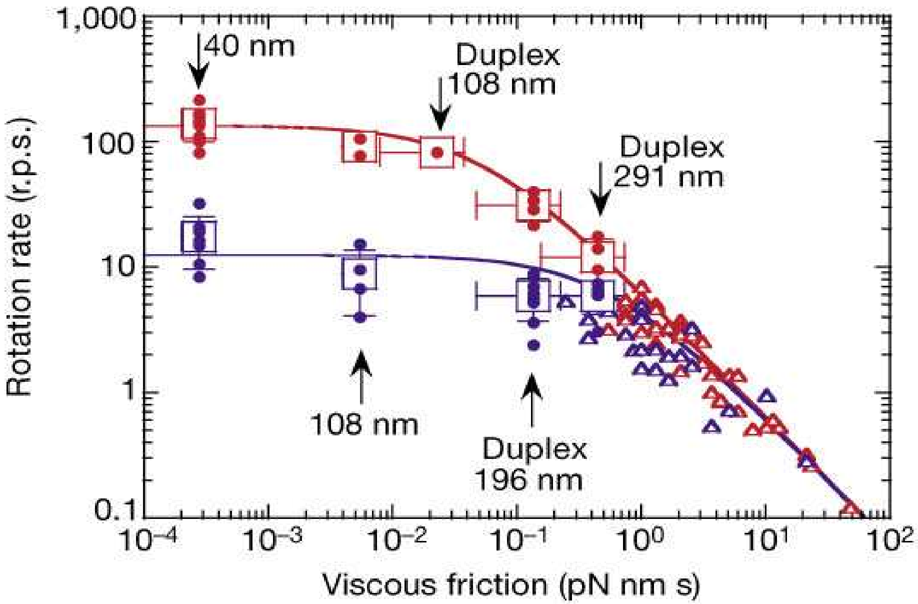
Experimental data from: Yasuda et al., Nature **410**, 898 (2001)



Substeps: 90º+30º

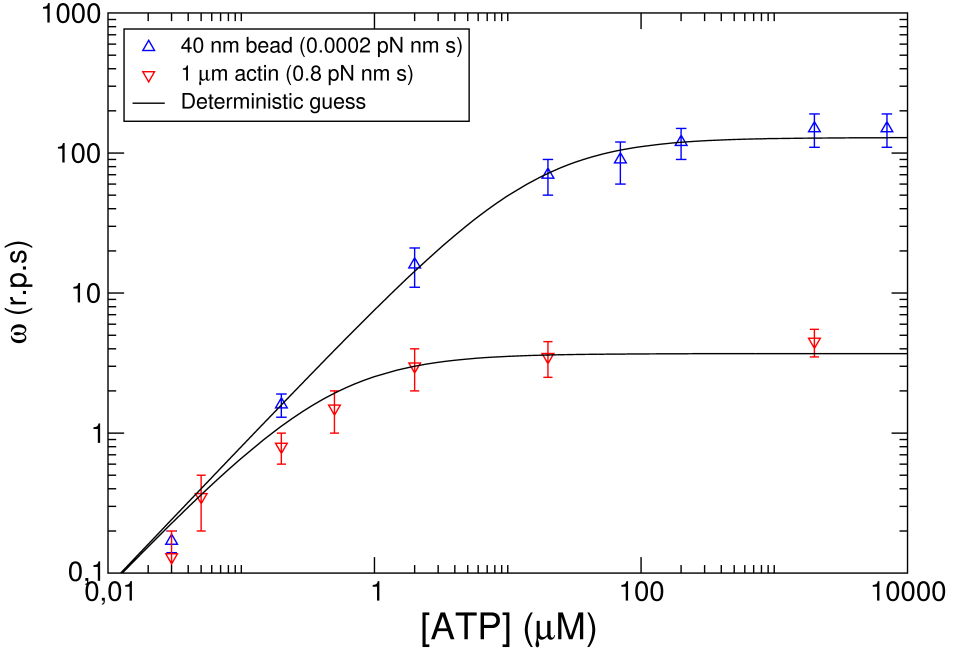
ms

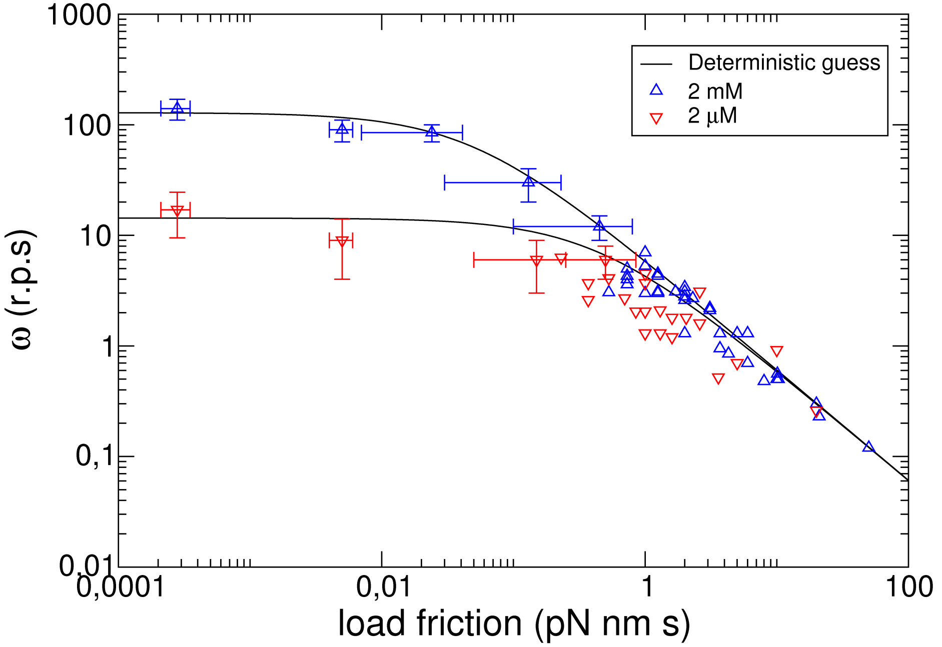




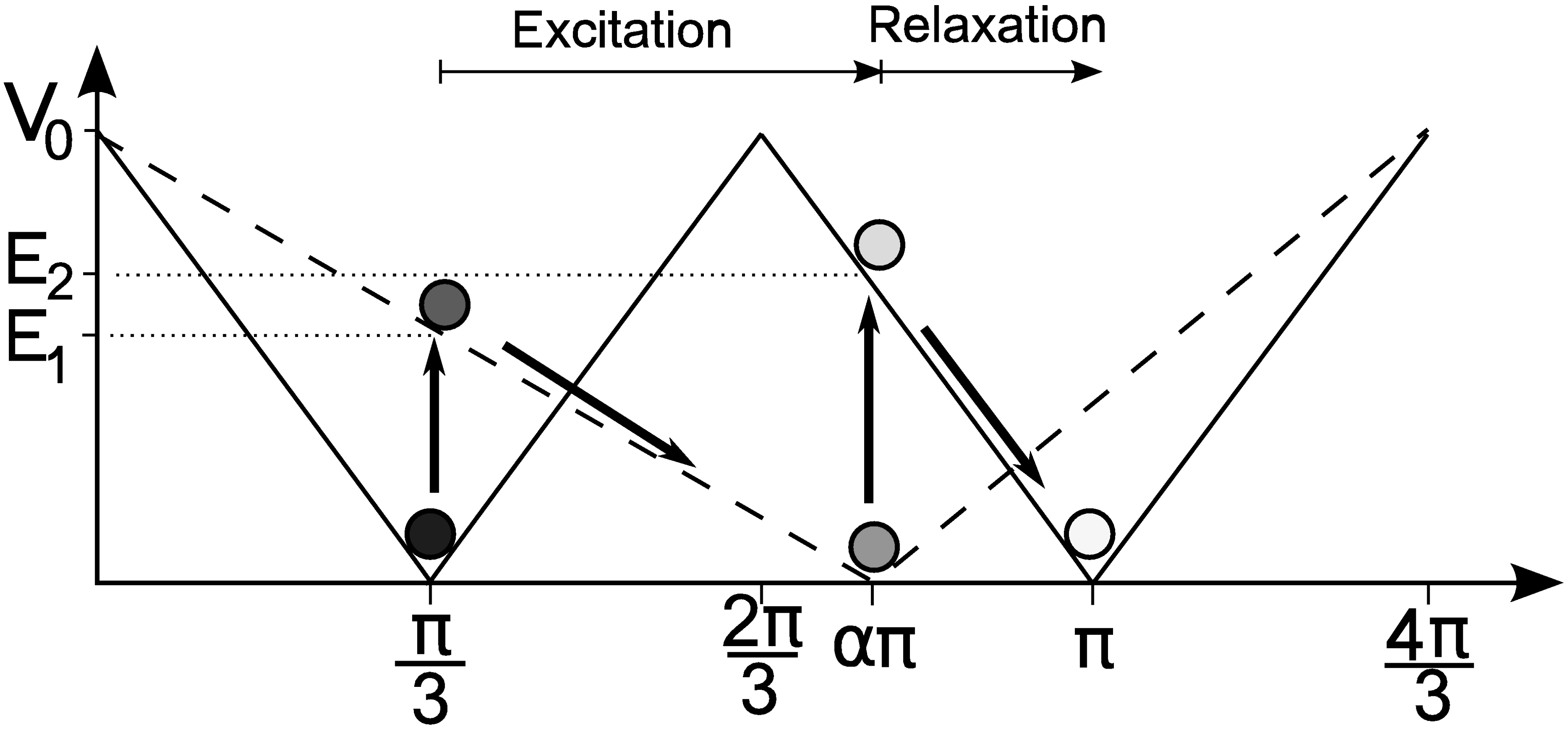
First approach

$$\omega = \frac{\theta_0}{t_0\left(1+\frac{k}{[ATP]}\right)+\frac{\theta_0^2(\gamma_0+\gamma_L)}{\Delta G_{ATP}}}$$





Last approach: The ratchet potential model



Parameters: V_0, E_1, E_2, and α.

Conditions:

$$E_1 = V_0(1 - \frac{1}{3\alpha})$$
$$E_2 = 3V_0(1 - \alpha)$$
$$E_1 + E_2 = \Delta G_{ATP}$$

α

Optimization: ω small → t maximum

$$\alpha = 0.805 \rightarrow \theta_1 = 84.85^0 \qquad \theta_2 = 35.15^0$$

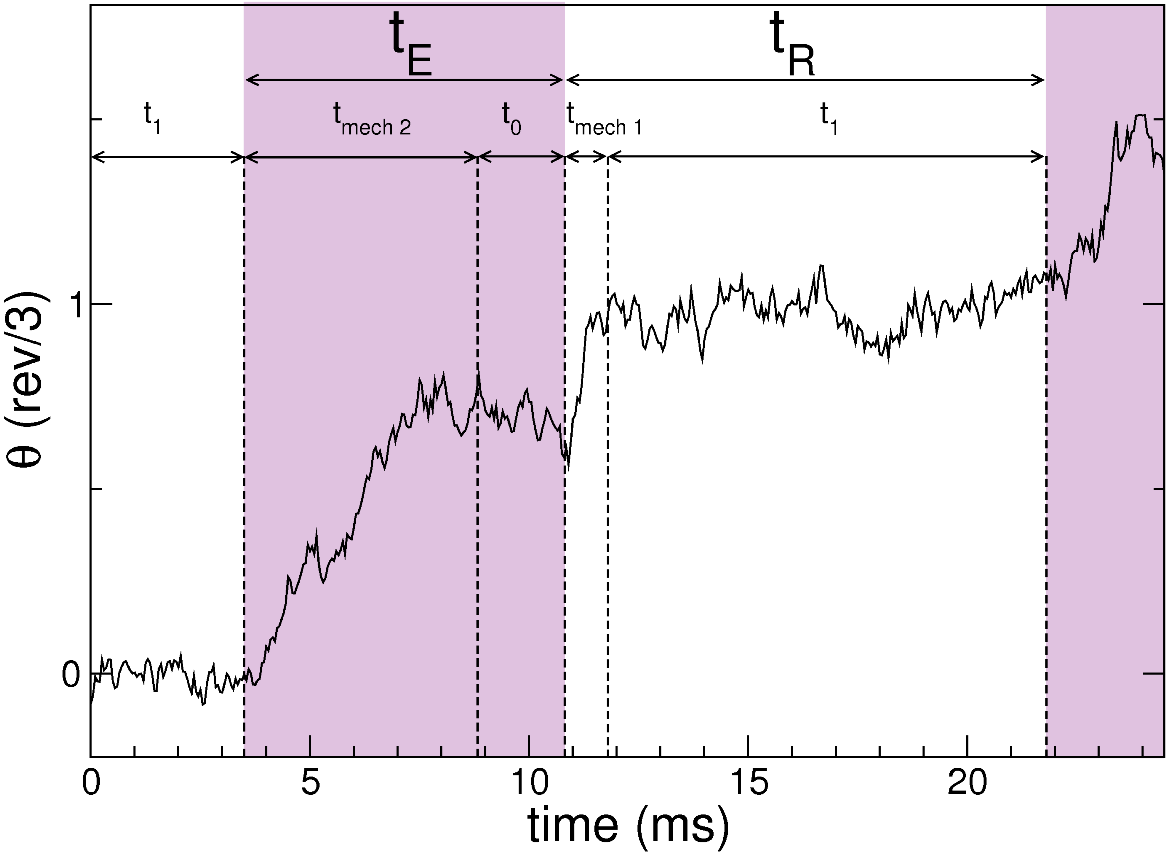
Torques

$$\tau_1 = 27\, pN.nm, \tau_2 = 65\, pN.nm, \bar{\tau} = 40\, pN.nm$$

Dynamical equations

$$\gamma\dot{\theta} = -V(\theta, [ATP]) + k(\phi - \theta) + \xi(t)$$

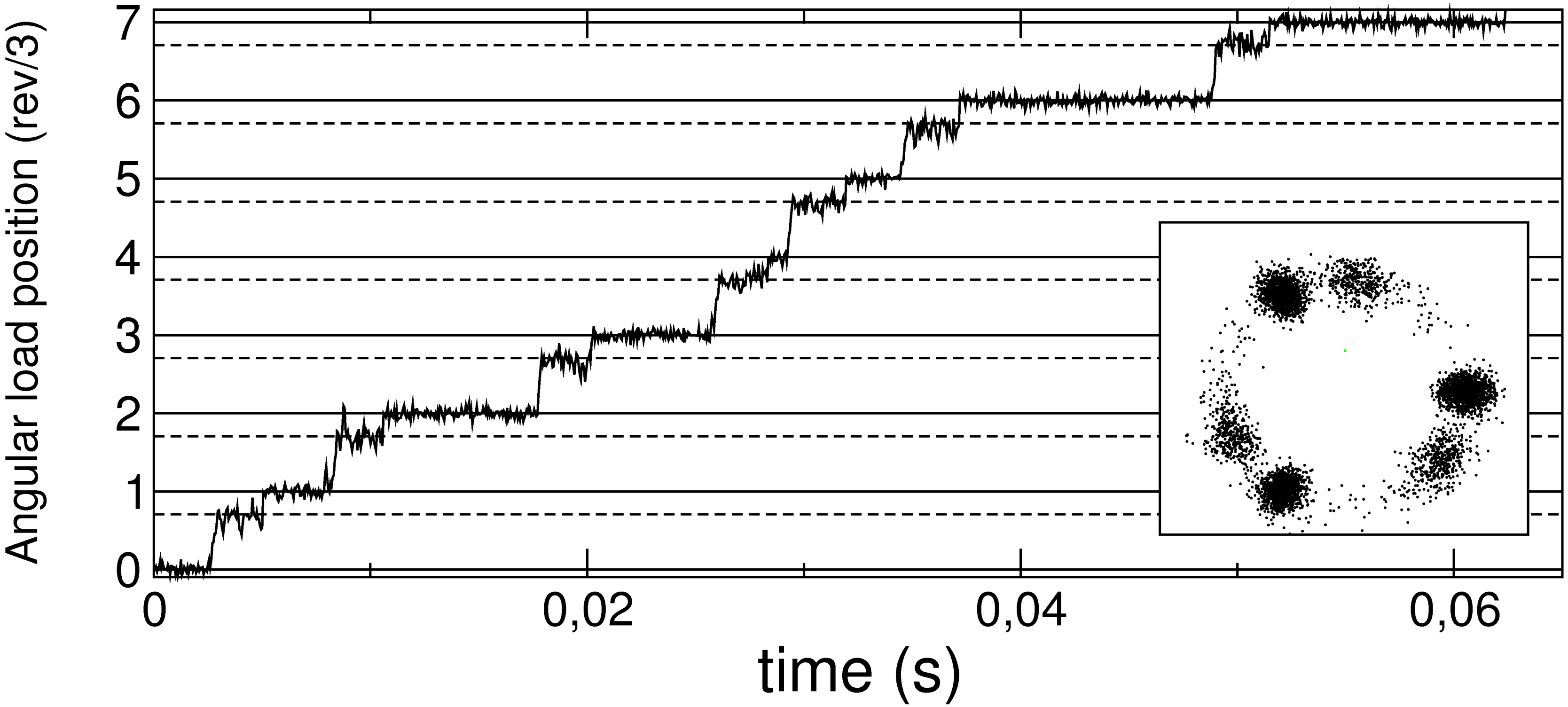
$$\gamma_L\dot{\phi} = -k(\phi - \theta) + \xi_L(t)$$

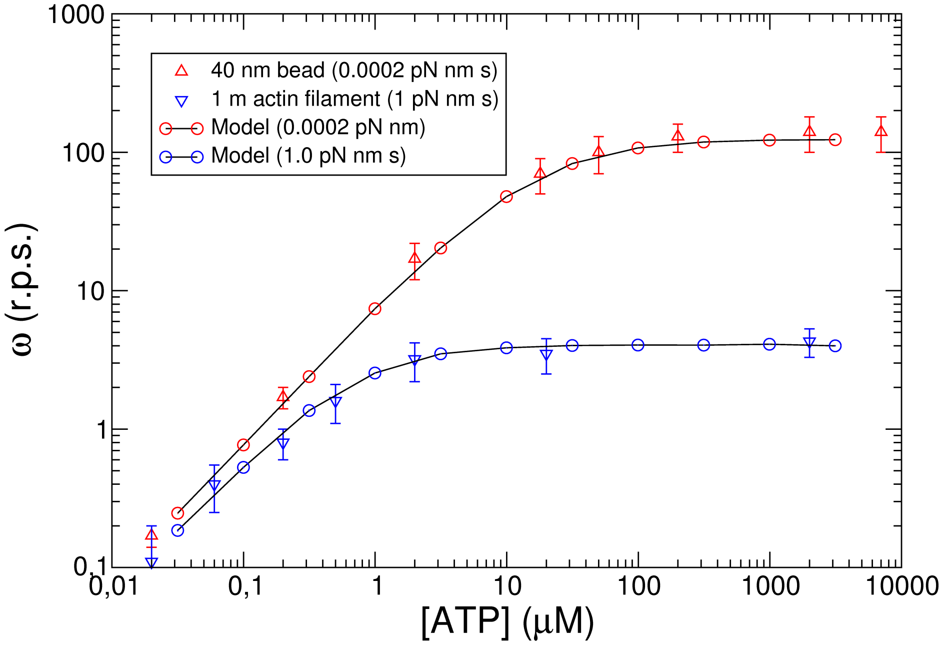


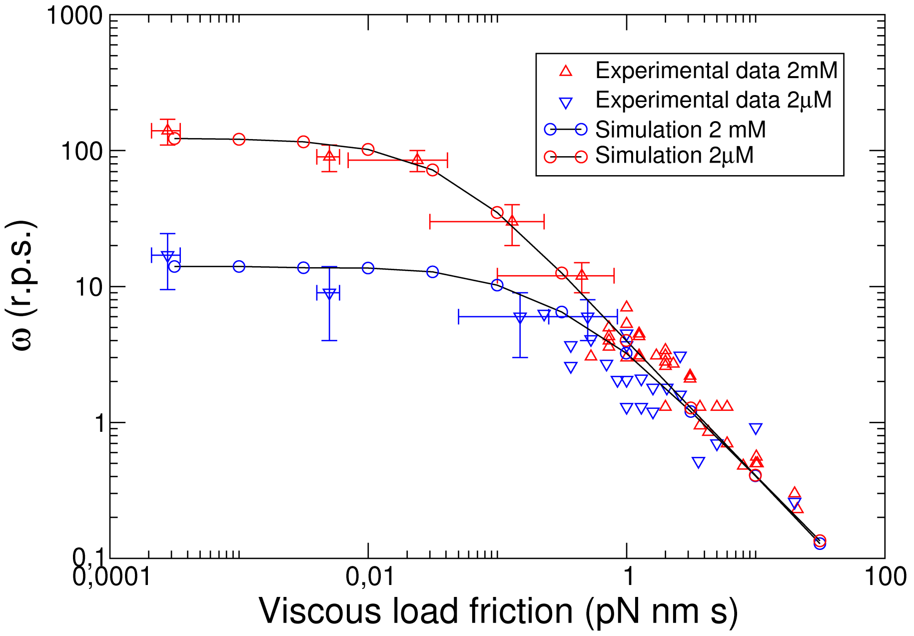
$$t_R = t_{mech\,1} + t_1, \qquad t_E = t_{mech\,2} + t_0$$

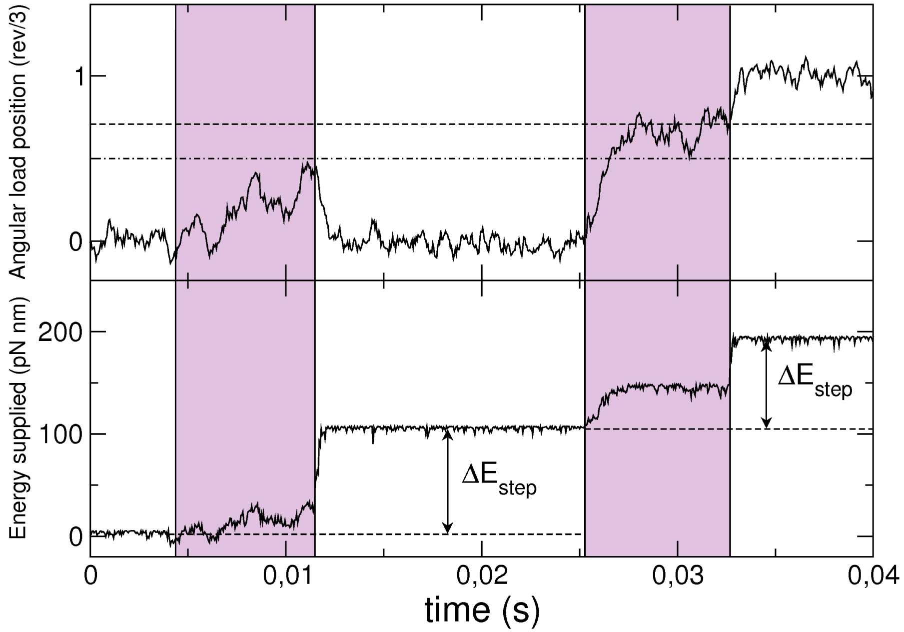
$$P(t_1) = \frac{1}{t_{ATP}} e^{-t_1/t_{ATP}}$$

$$t_{ATP} = t_0 \frac{k}{[ATP]}$$









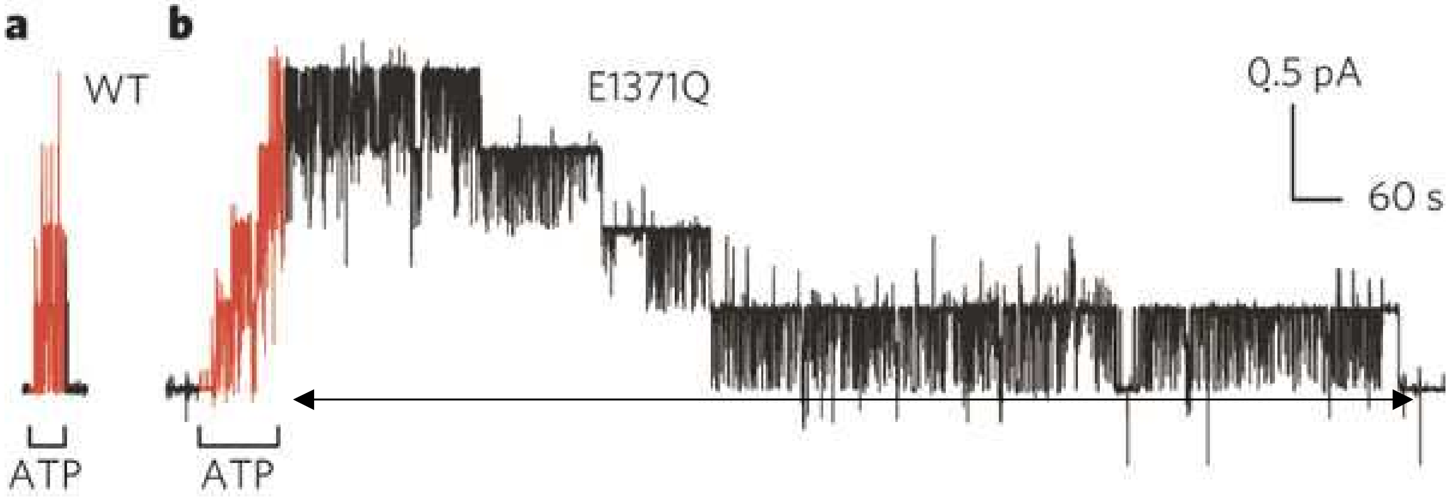
Input energy coupling ratio

3. CHANNELS

CFTR chloride channel (cystic fibrosis)

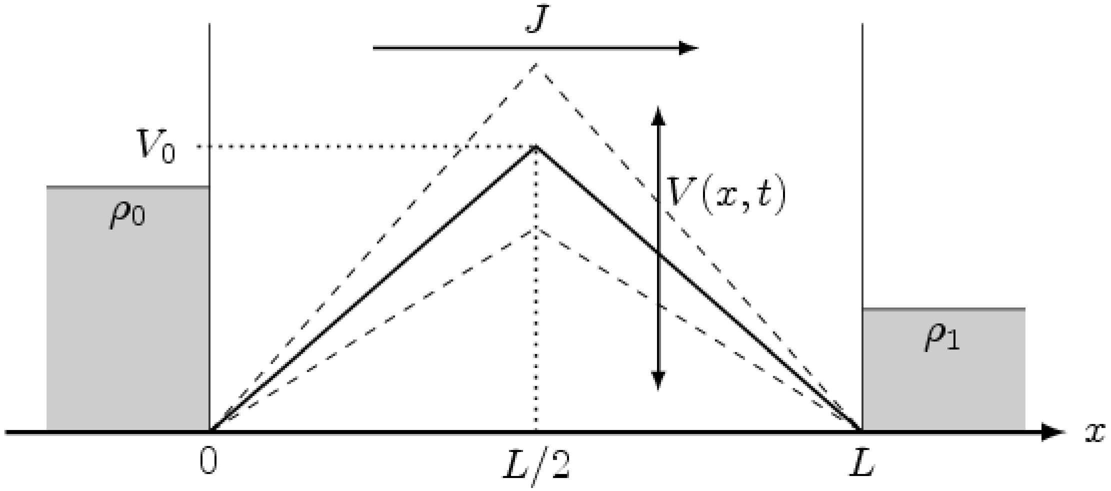
Gadsby, et al, NATURE **440**,477(2006)

Experimental data: channels need energy input



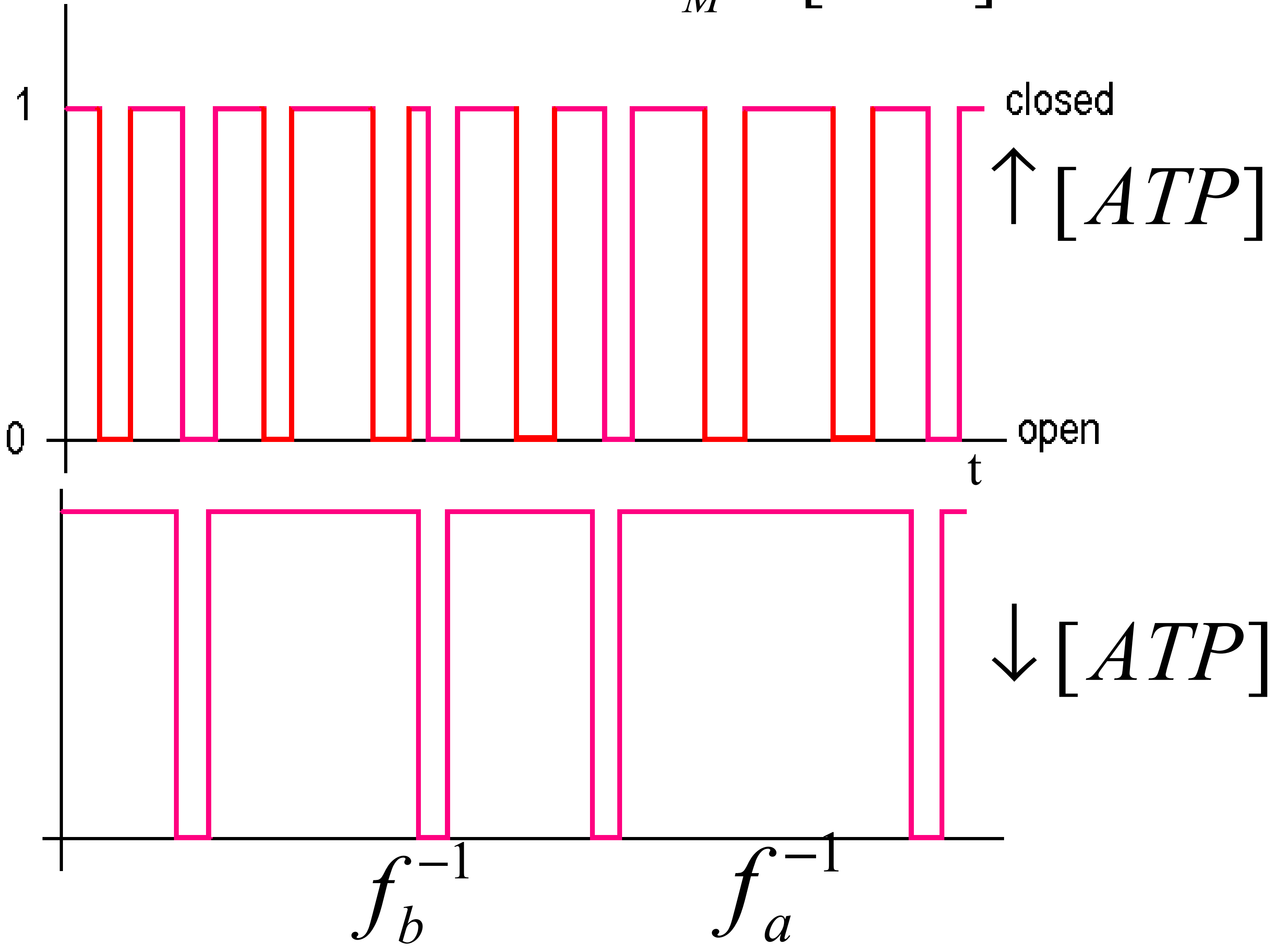
ATP-binding OPEN
ATP-hydrolisis CLOSED

Simple model



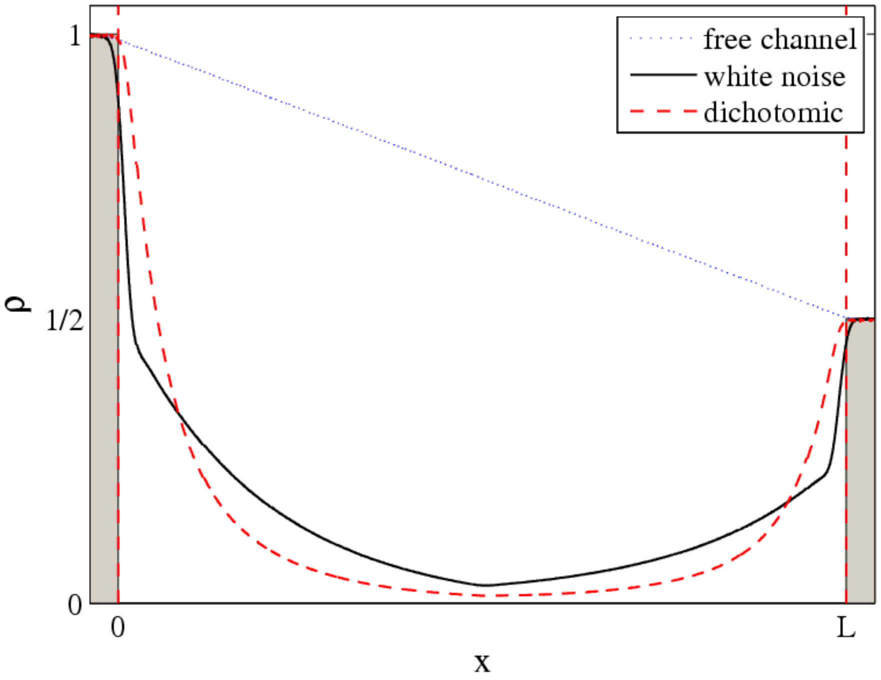
$$\gamma \dot{x} = -V(x,[ATP]) + \xi(t)$$

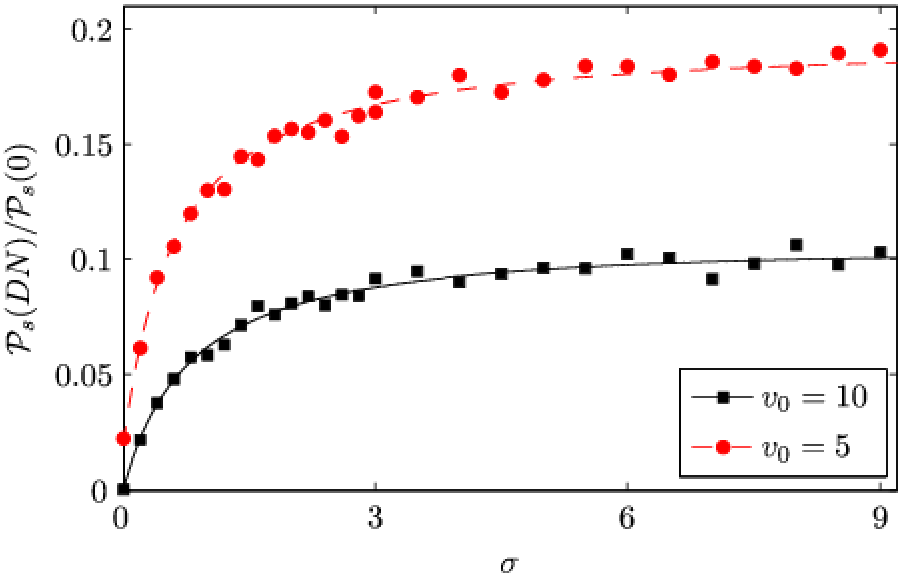
$$f_b = \omega_0 \qquad f_a = \frac{\omega_0[ATP]}{k_M + [ATP]}$$



Theoretical study of a membrane channel gated by ATP

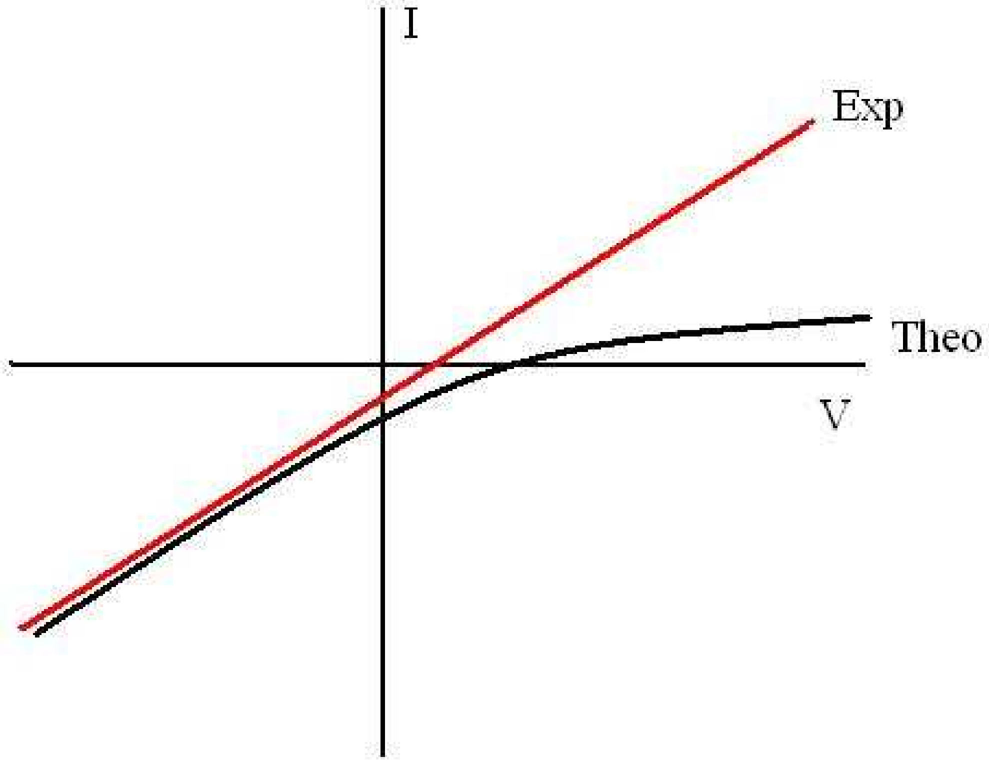
J.G. Orlandi & JMS, Eur.Phys.J. (2009)





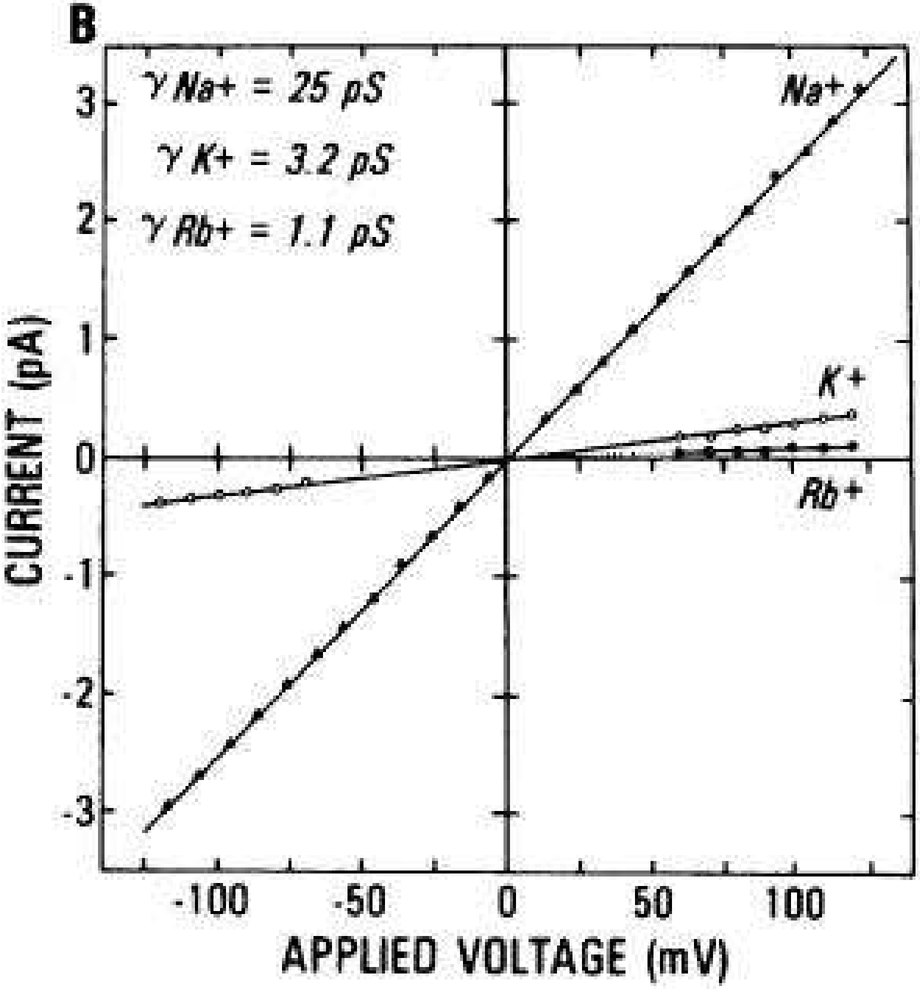
...but...experimental data for CLC Cl channel...

Accardi et al. J. Gen. Physiol., 123, 109(2004)



Improvement ?:   structured potential inside the channel

Selectivity

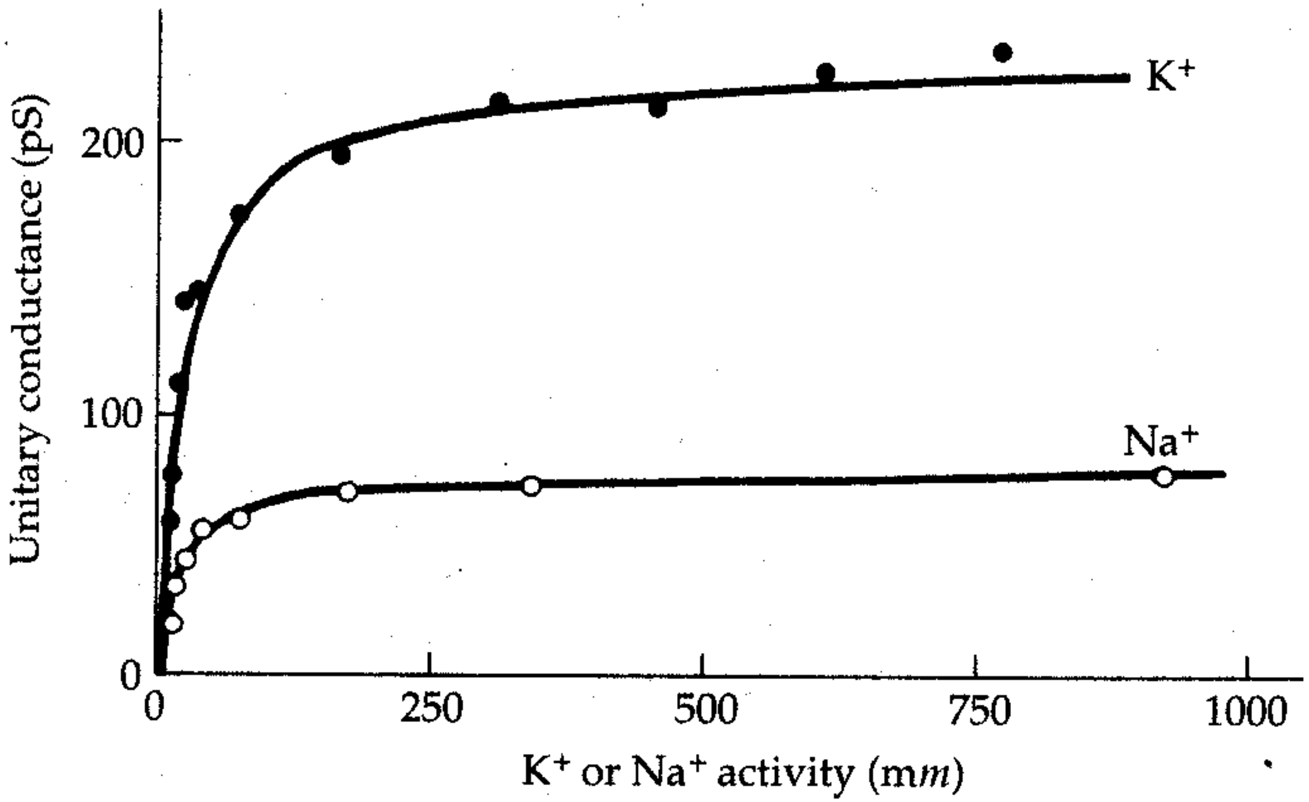


from Hartshorne et al. PNAS**82**,240(1985)

SATURATION

$$J = \frac{J_{max}[S]}{K_s + [S]}$$

(B) SARCOPLASMIC RETICULUM CHANNEL



Na anomalous conductance?

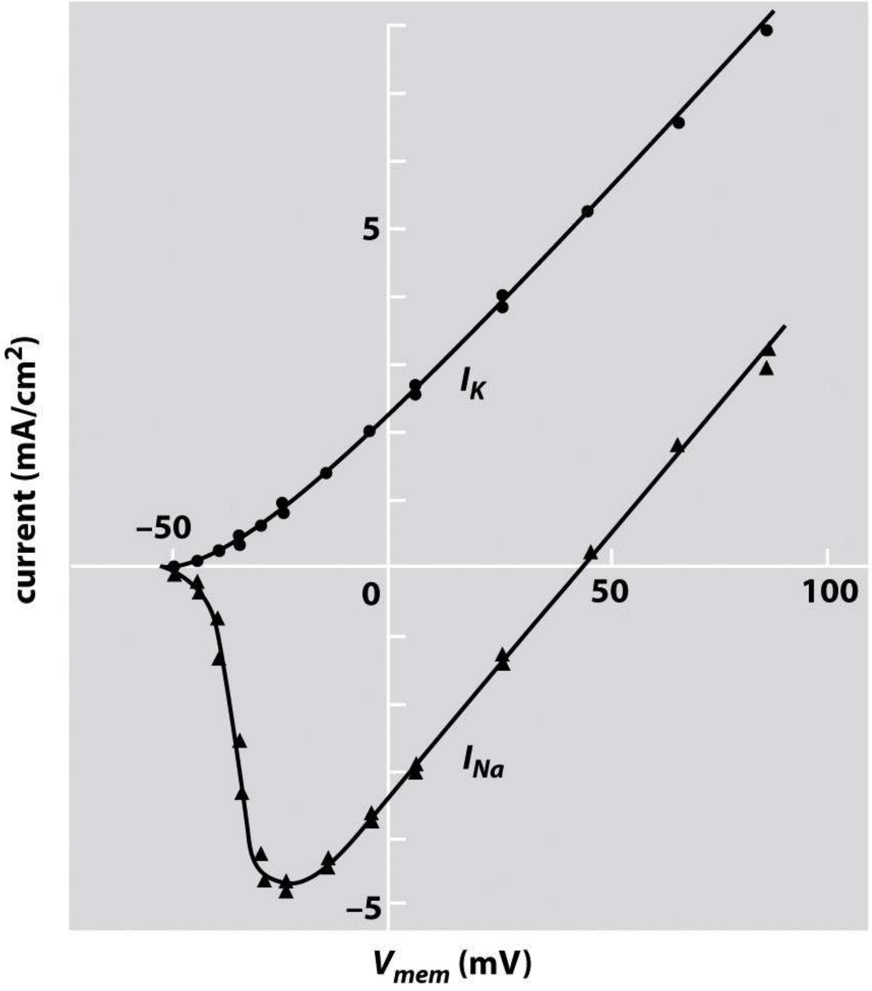
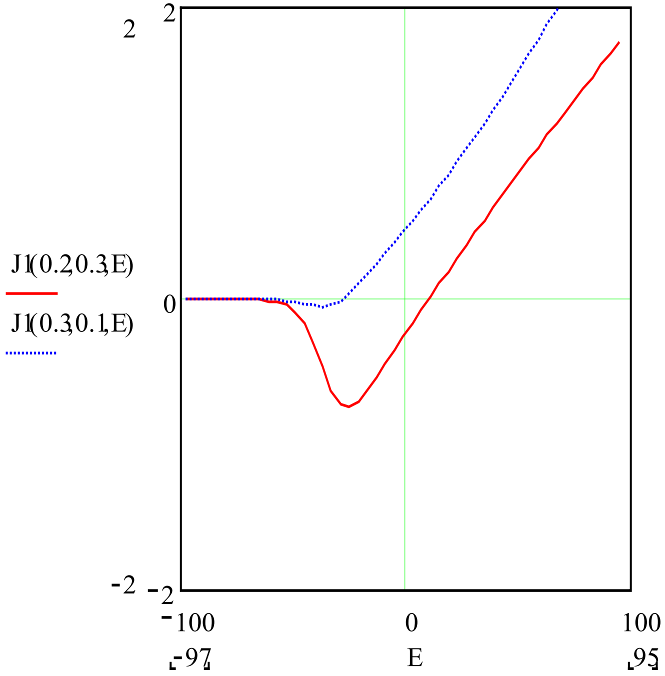


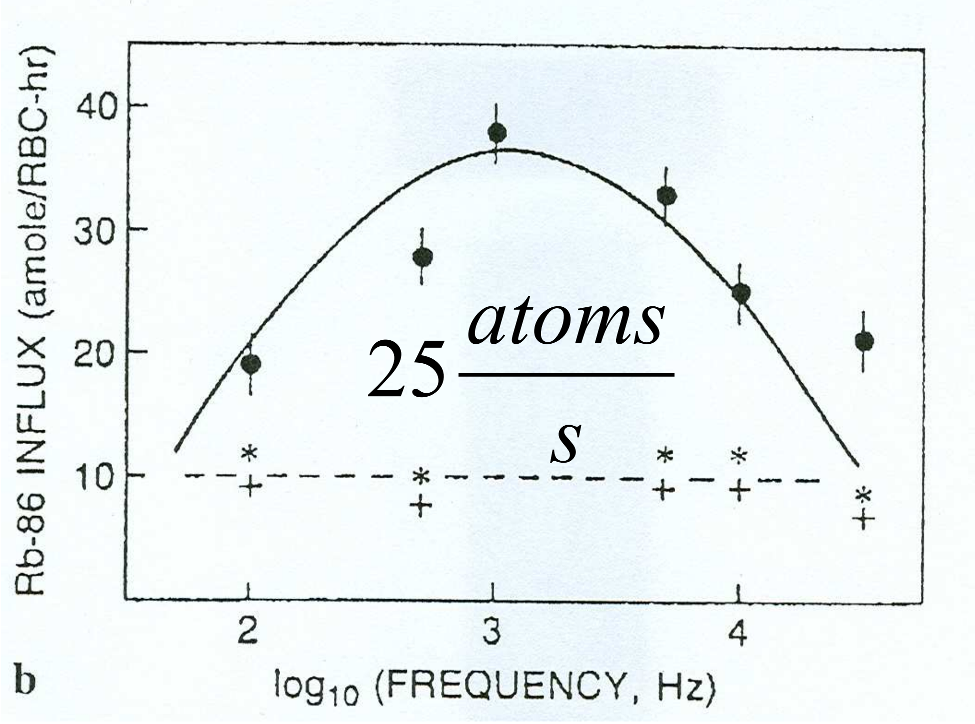
Figure 17.12 Physical Biology of the Cell (© Garland Science 2009)



4. PUMPS

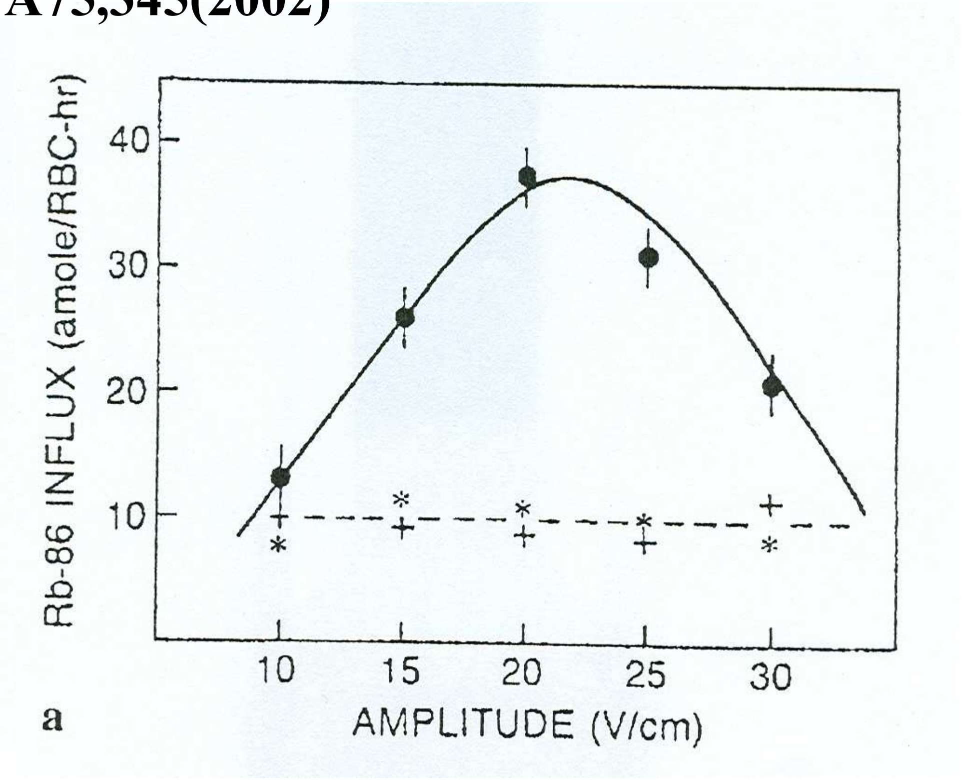
Na-K pump

Tsong & Xie, Appl.Phys. A75,345(2002)

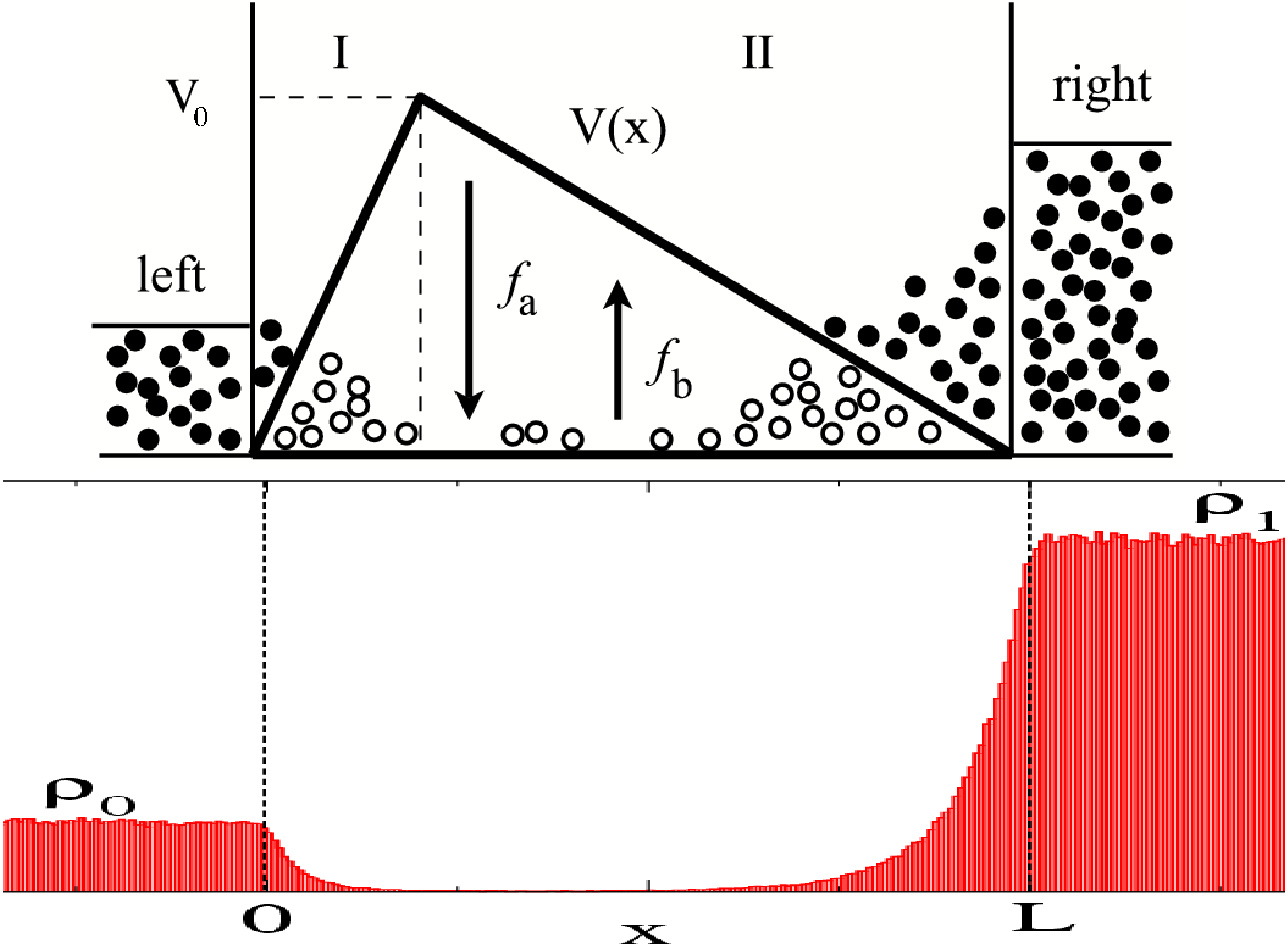


$$25\frac{atoms}{s}$$

b



a


I
II
V_0
V(x)
right
left
f_a
f_b
ρ_1
ρ_0
0
x
L

$$\left[d_z^3 - 2F_i d_z^2 - (f_a + f_b) d_z + 2 f_b F_i\right] \rho_i(z) = J(f_a + f_b)$$

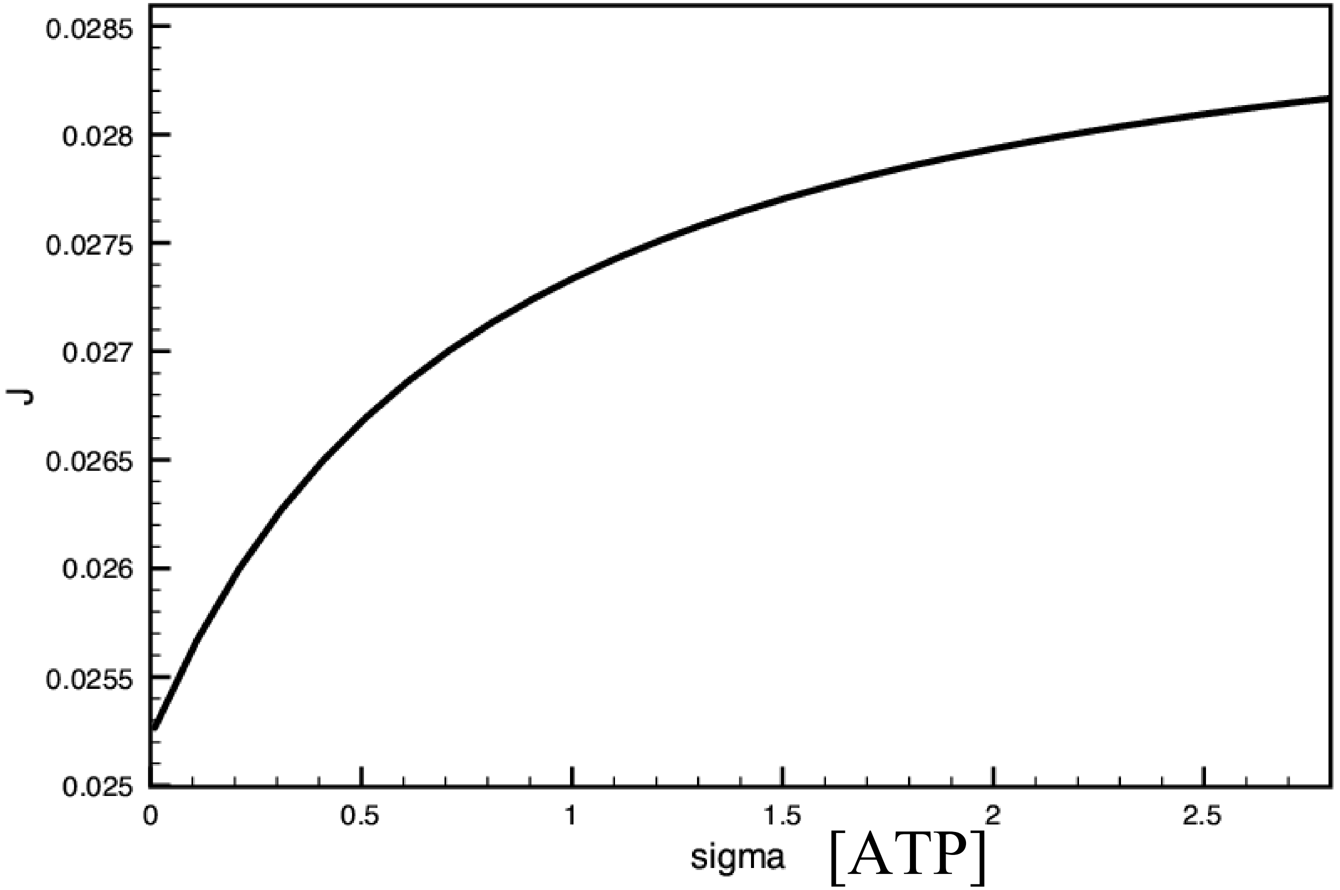
Seven unknown constants with seven boundary conditions

if $J = 0$ → $\dfrac{\rho_{\max}}{\rho_0}$

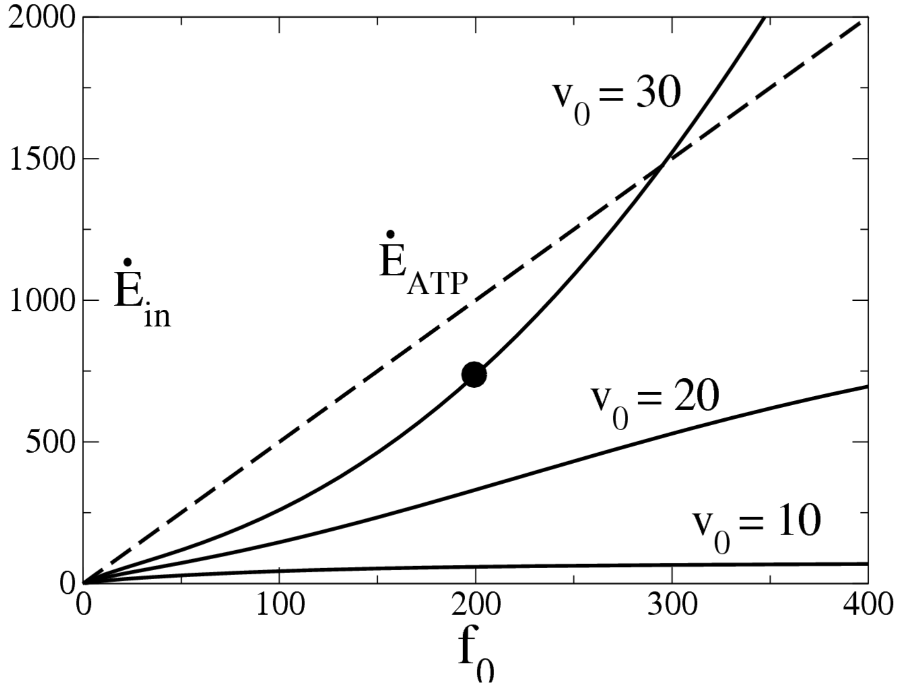
If $\rho_1 < \rho_{\max}$ → $J \neq 0$

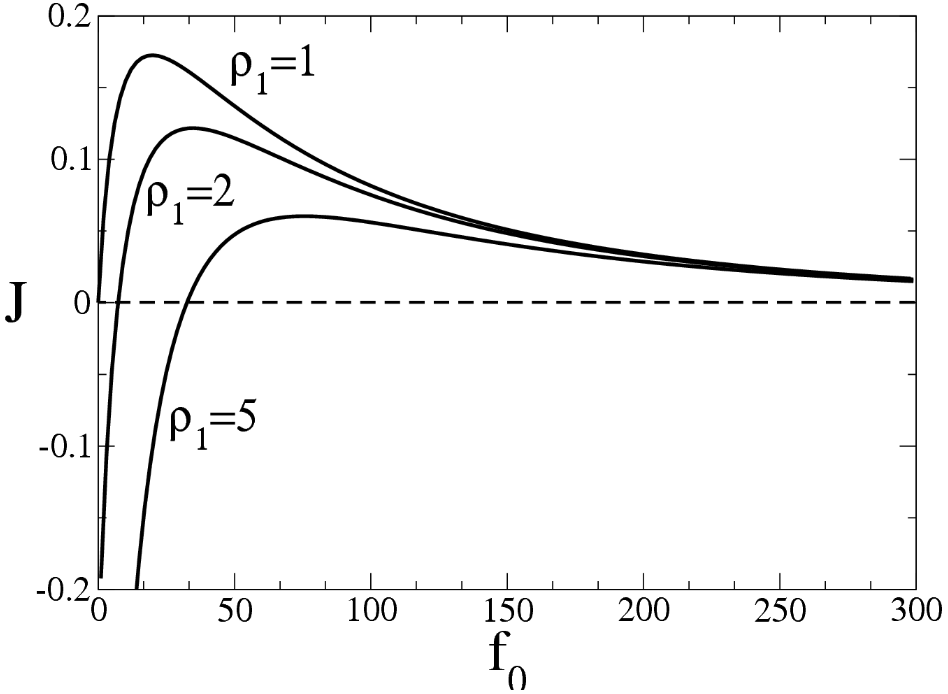
Parameters: $V_0, \delta, L, S, \gamma, \rho_0, \omega_0, \sigma([ATP]), \ldots$

$$\omega_0 = 300\,, \frac{\rho_1}{\rho_0} = 10$$



[ATP]


2000
1500
1000
500
0
$\dot{E}_{in}$
$\dot{E}_{ATP}$
$v_0 = 30$
$v_0 = 20$
$v_0 = 10$
0
100
200
300
400
f_0


0.2
0.1
0
-0.1
-0.2
J
ρ_1=1
ρ_1=2
ρ_1=5
0
50
100
150
200
250
300
f_0

2. CONCLUSSIONS

Work in progress

V gated channels
Binding sites
Selectivity
Saturation against substrate (M-M)
Rotatory pumps (mechanical model)
Motors fueled by gradient and... more: ΔG_{ATP} , τ_{ext}

Important points

-Complex systems
-Different levels of description
-Variety of models
-Experimental data available but not enough

-Biological parameter values

THANKS

Muneyuki et al, Bioph.J. 92, 1806 (2007)