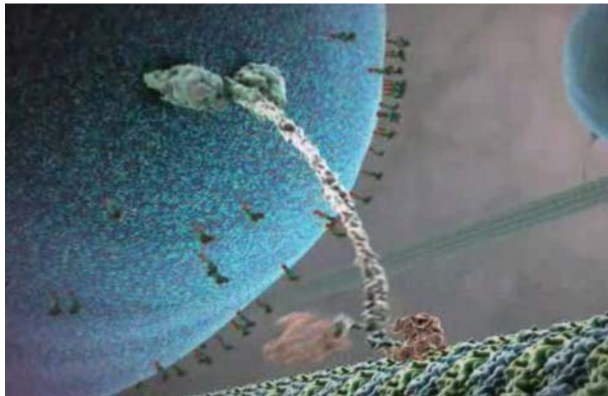
UNIVERSITÄT
LEIPZIG

Lecture 8

# Soft Matter Physics

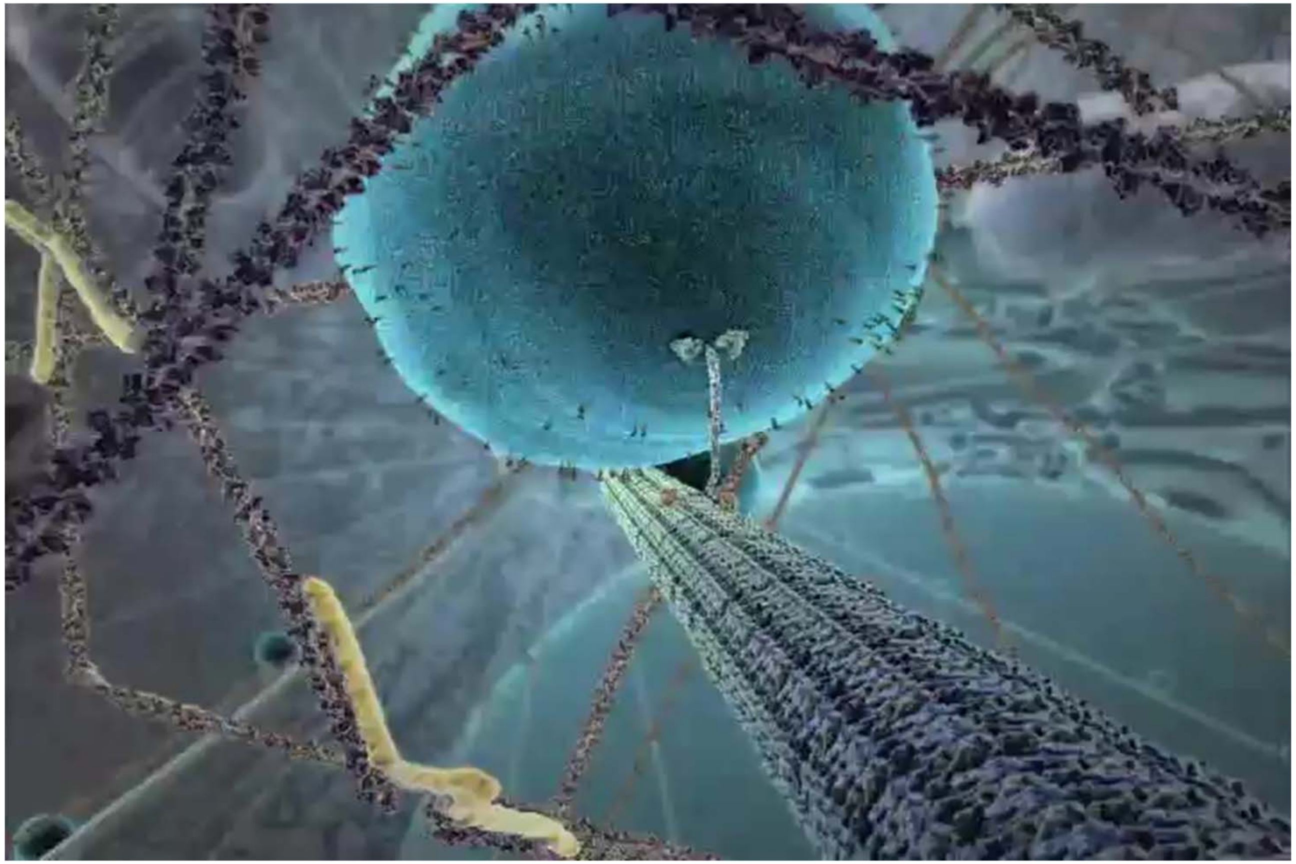
## Dynamics of molecular motors

- Molecular machines
- Brownian motors
- Driven walks to describe motor dynamics
- Force dependence of molecular motors



Ralf Seidel | Peter Debye Institute for Soft Matter Physics | Molecular Biophysics Group

# Cargo transport by a kinesin motor



# Velocity-Force relations of molecular motors

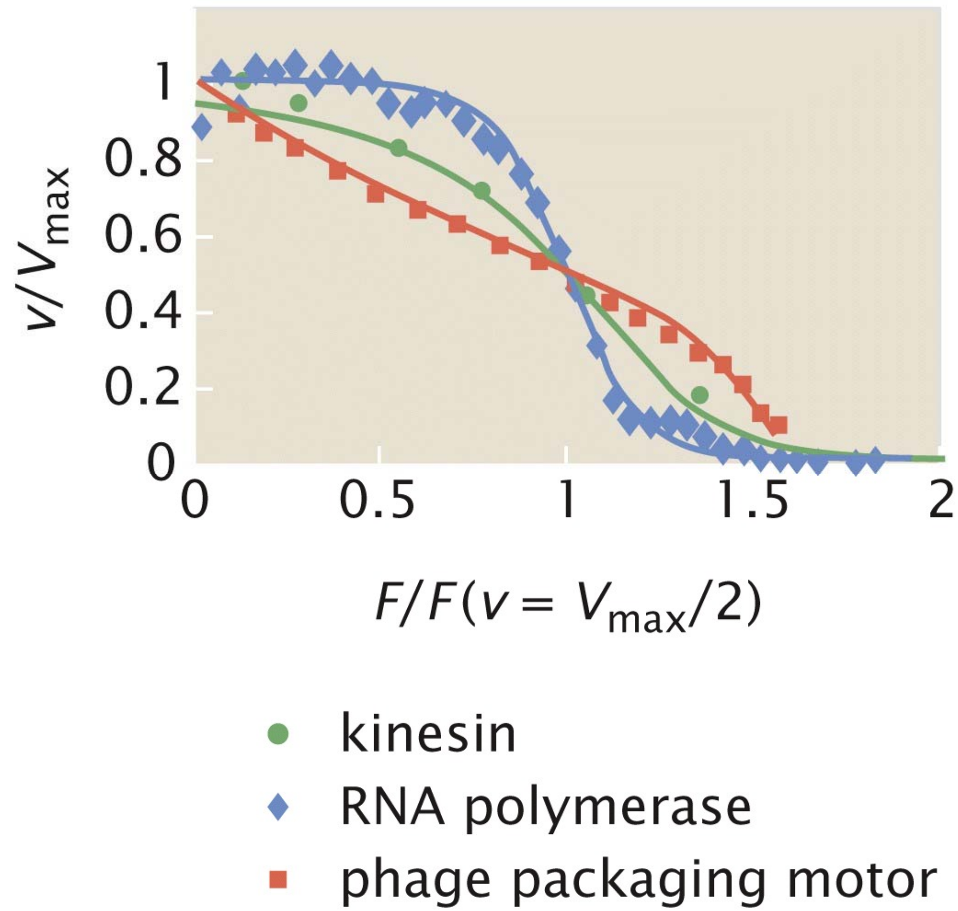



Figure 16.1 Physical Biology of the Cell, 2ed. (© Garland Science 2013)

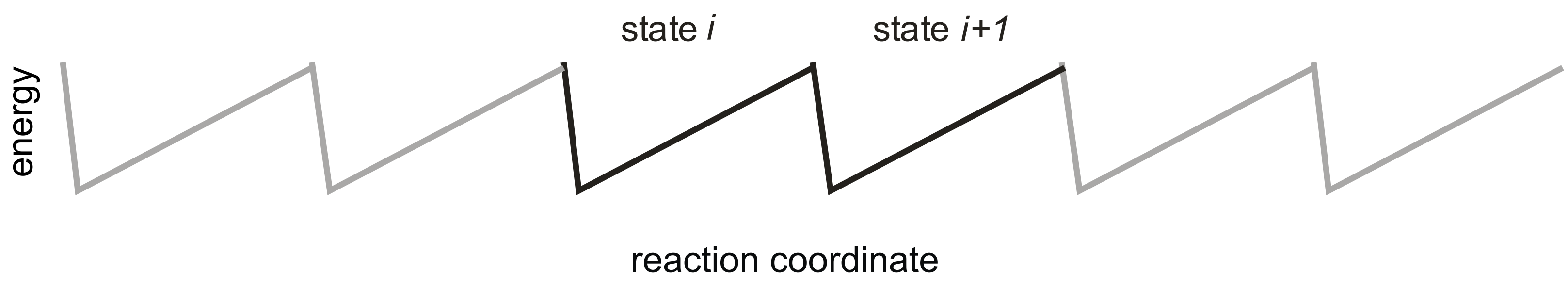
Can one understand mechanisms of biological machines from studying their power generation characteristics?

# What is a machine ?

System that converts energy

energy source -> useful energy form (e.g. mechanical work)
-> not simple energy dissipation

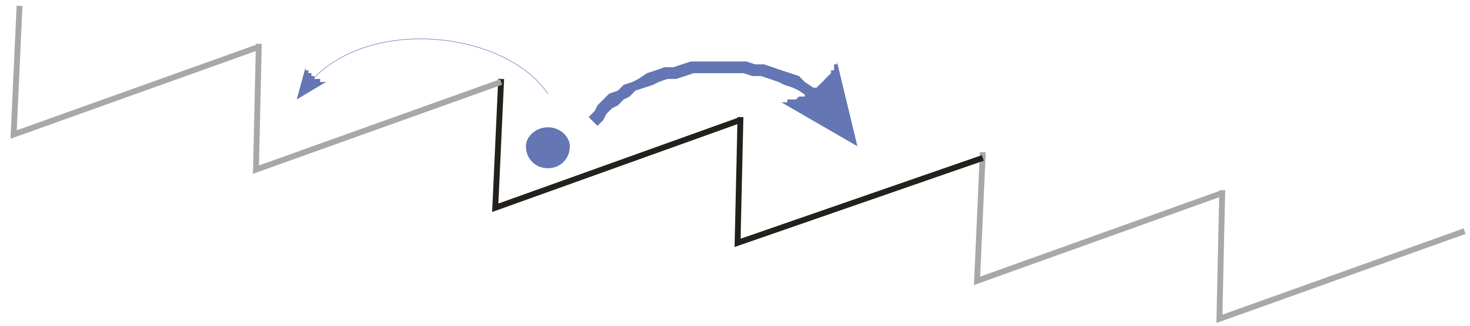
consists of different energy states




**We need non-equilibrium conditions such that the machine can do work!**

# How to introduce non-equilibrium conditions (1)

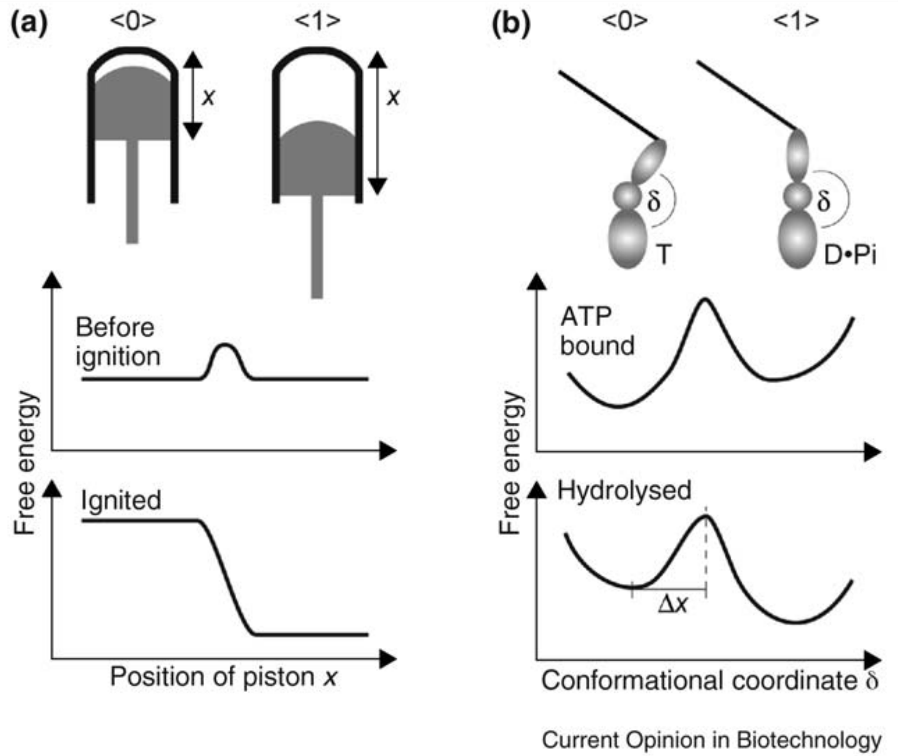
Tilt energy landscape



Spatial bias in energy landscape, e.g. potential energy difference, chemical gradient

# How to introduce non-equilibrium conditions (2)

**Cyclic machines:** Modulate energy landscape in time




Hugel & Lumme
Curr Opin Biotech 2010

Sequential modulation: ATP hydrolysis more than simple ligand binding

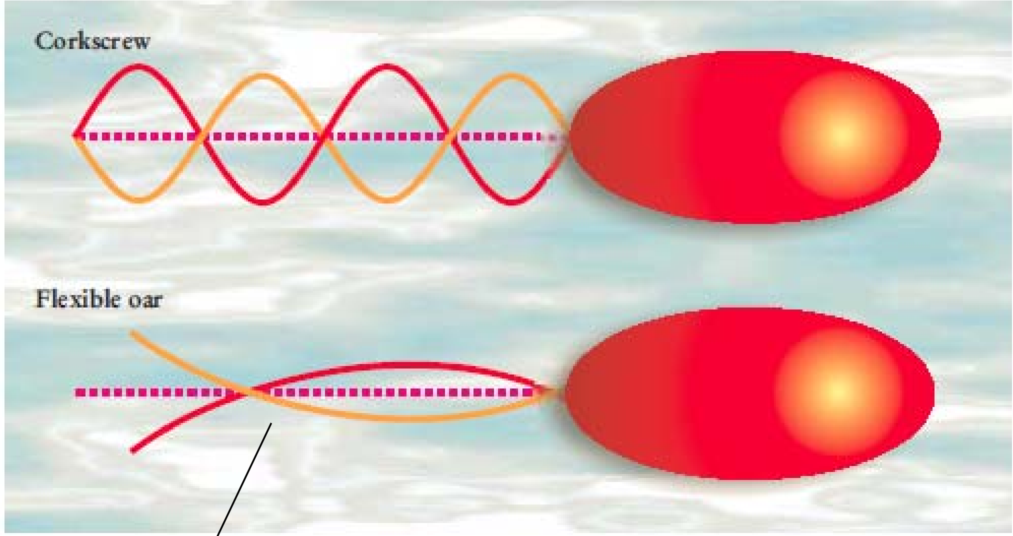
Used in engineering and by molecular machines

# Differences of microscopic machines (compared to macro)

**Low Reynolds number swimmers**

adiabatic mechanism
("all" energy is converted into work)

non-adiabatic mechanism
(dissipation, diffusion part of mechanism)




$v \sim f$

$v \sim f^2$

(Hänggi et al.)

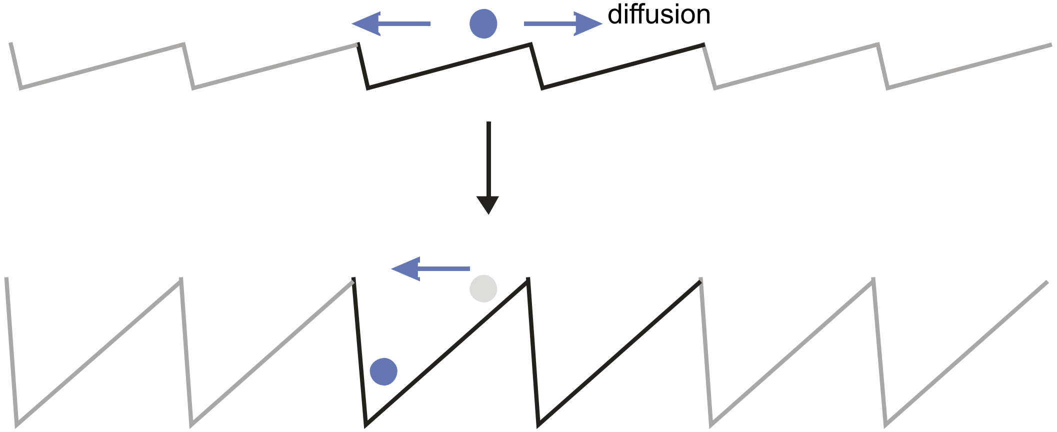
breaking time reversibility requires oar bending from viscous drag

**Dissipation and thus a non-adiabatic mechanism can be central for the function**

# Simple realization of Brownian motor

**Machines where diffusion is central for the function are called Brownian motors**

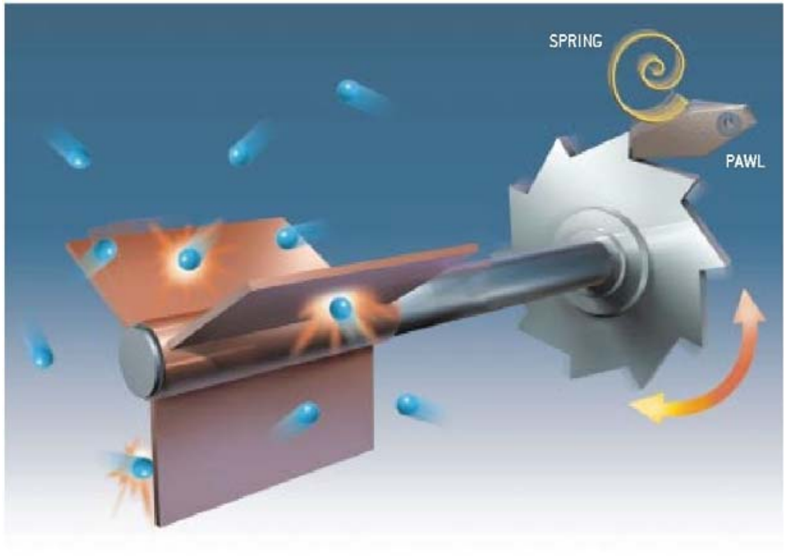
Modulation of asymmetric energy landscape in time




Random or periodic modulation of barrier height

Temporal bias in energy landscape, e.g. ATP hydrolysis

# Brownian motor: Non-equilibrium required!

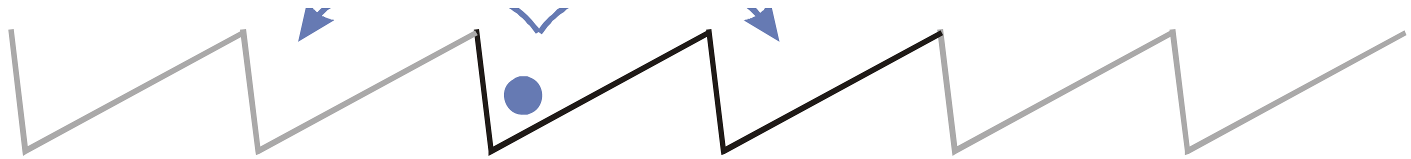



**Thermal fluctuations/diffusion matter!**

**Brownian ratchet**
Perpetual motion of $2^{nd}$ kind!
Breaks $2^{nd}$ law of TD not $1^{st}$ law!

Does not produce work!

Equal movement in both directions



**A static asymmetric unbiased periodic potential does not produce net motion**

- Molecular machines need to be operated away from thermal equilibrium! (external fields, chemical energy, modulations in time)
- But: Under appropriate nonequilibrium conditions structural anisotropy can sustain directed motion (wheel can turn only in direction if driven)

# Cytoskeletal motors: Two motor domains

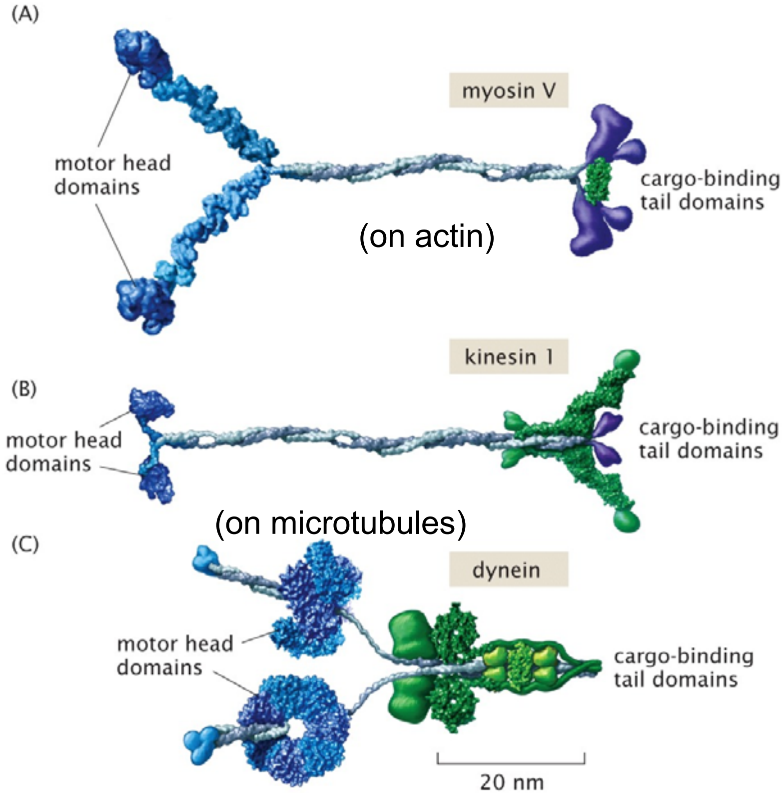



Figure 16.2 Physical Biology of the Cell, 2ed. (© Garland Science 2013)

(on microtubules)

motor domains: hydrolysis of ATP coupled to (amplified) large-scale conformational change
coordination between both motors required!!!

Maximum force of microtubule motors

$$F_{\max} = \frac{\text{free energy of ATP hydrolysis}}{\text{step size}} \approx \frac{20\, k_B T}{8\,\text{nm}} \approx 10\,\text{pN},$$

Dir. transport of membrane-bound organelles by kinesins and dyneins

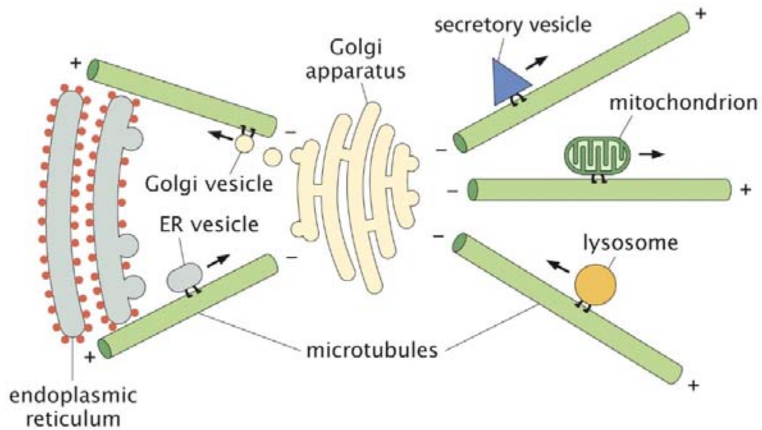



Figure 16.3 Physical Biology of the Cell, 2ed. (© Garland Science 2013)

Each single motor is processive!

# Idealized structure of muscle

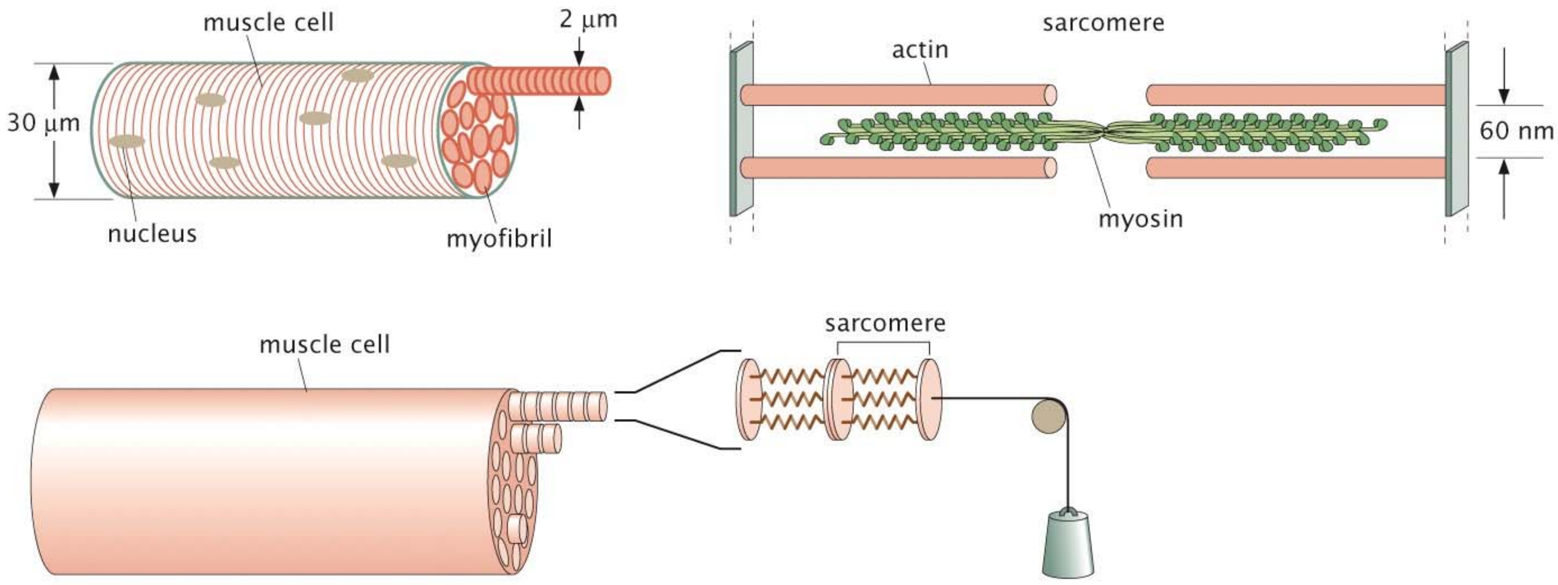


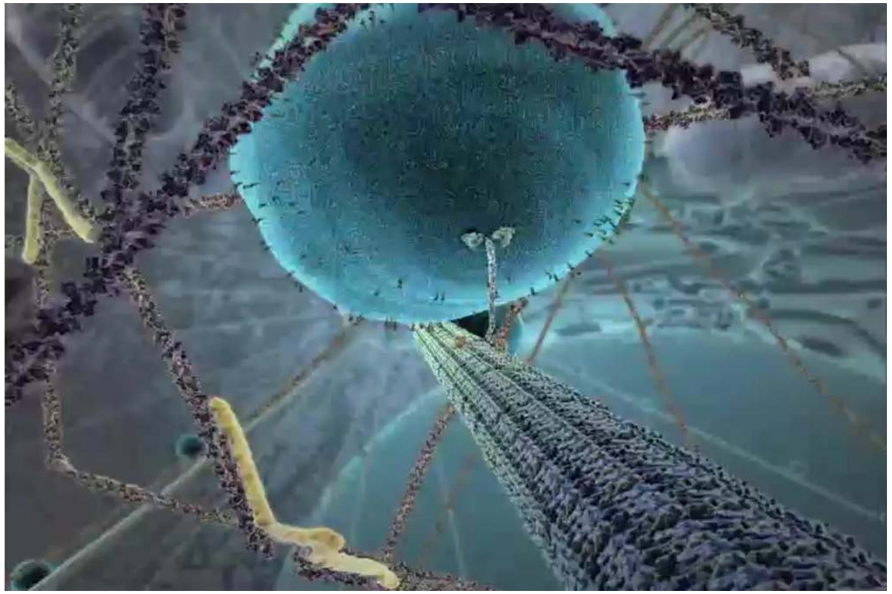

Figure 16.8 Physical Biology of the Cell, 2ed. (© Garland Science 2013)

Skeletal muscle myosin: two motor domains but only one active
→ unprocessive motion
motor spends most of the time unbound from actin
→ gives little „pushes“ (many motors act simultaneously)
→ energy required to just hold things!

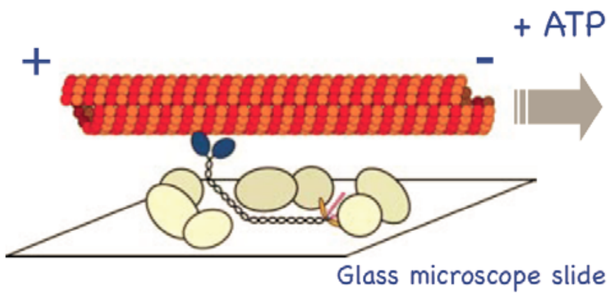
# Studying molecular motors: Gliding assay

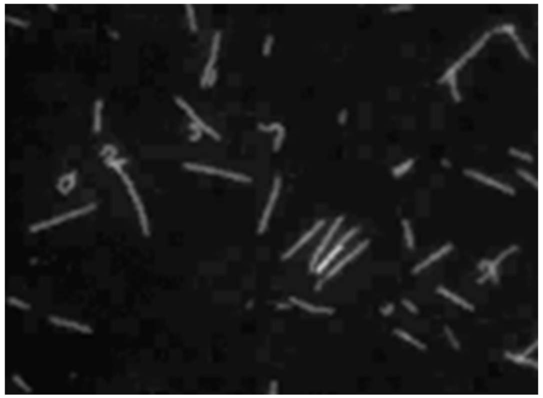
e.g. kinesin 1 on microtubules

Involved in vesicle transport
along microtubules



In vitro gliding assay
(up side down assay)
no info on processivity



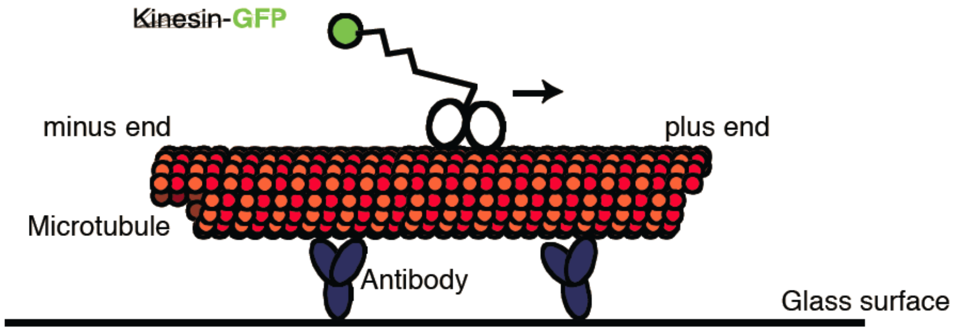



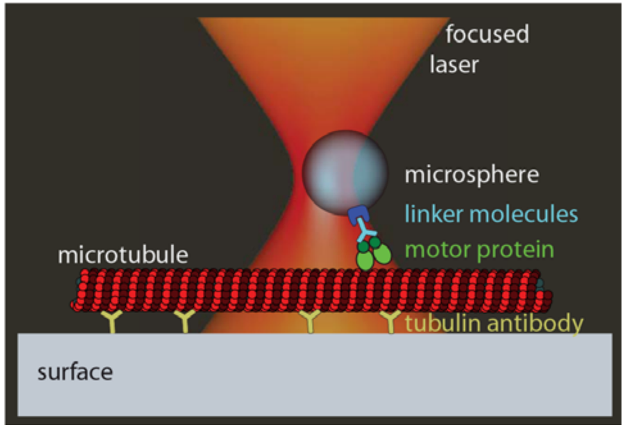
Stefan Diez lab, TU Dresden

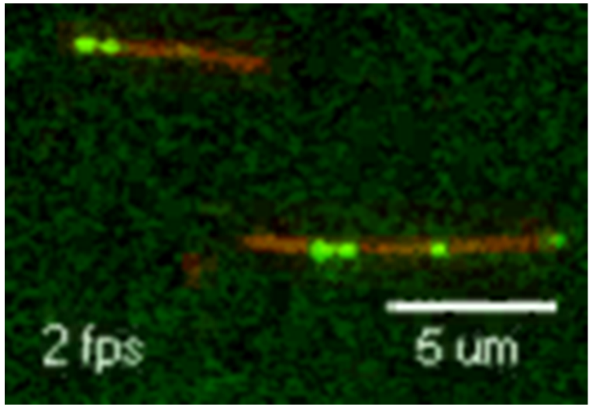
# Studying molecular motors: Stepping assay

e.g. kinesin 1 on microtubules

Gives direct insight into processivity



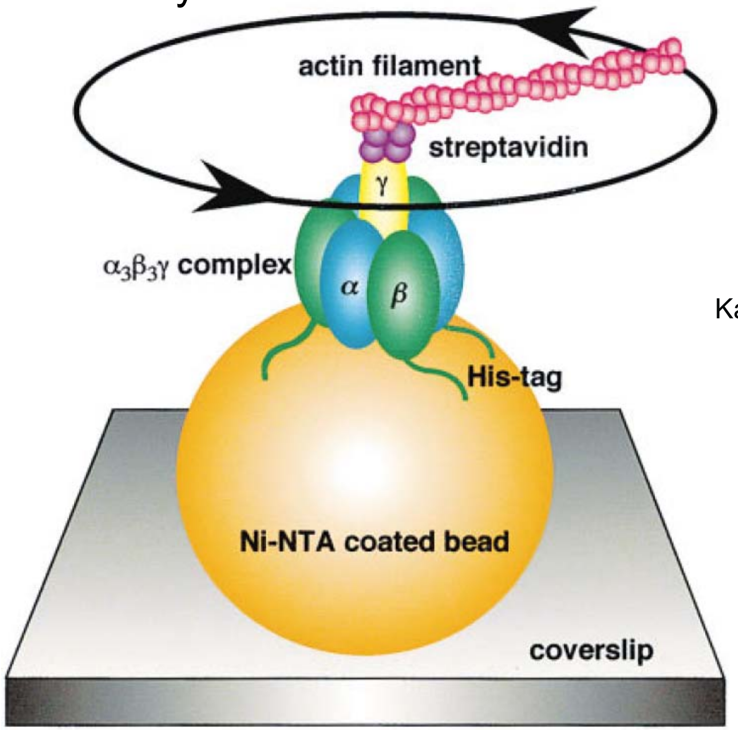

Stefan Diez lab, TU Dresden

also used in force-based experiments

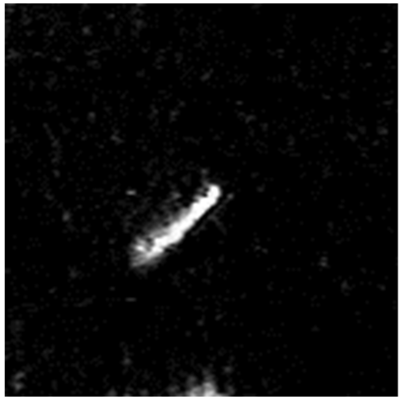
# Rotary motors: F-type ATP synthases

ATP synthases are reversible, combination of proton-driven rotor and ATP synthease

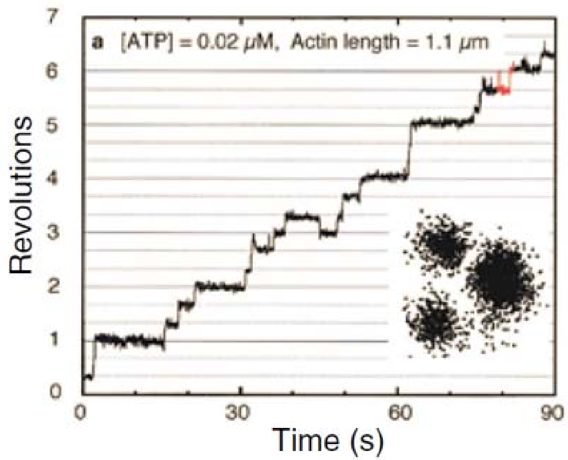
Only ATP subunit




very high energy efficiencies



Kazuhiko Kinosita, Jr.



# Scheme of position-state models to describe motors

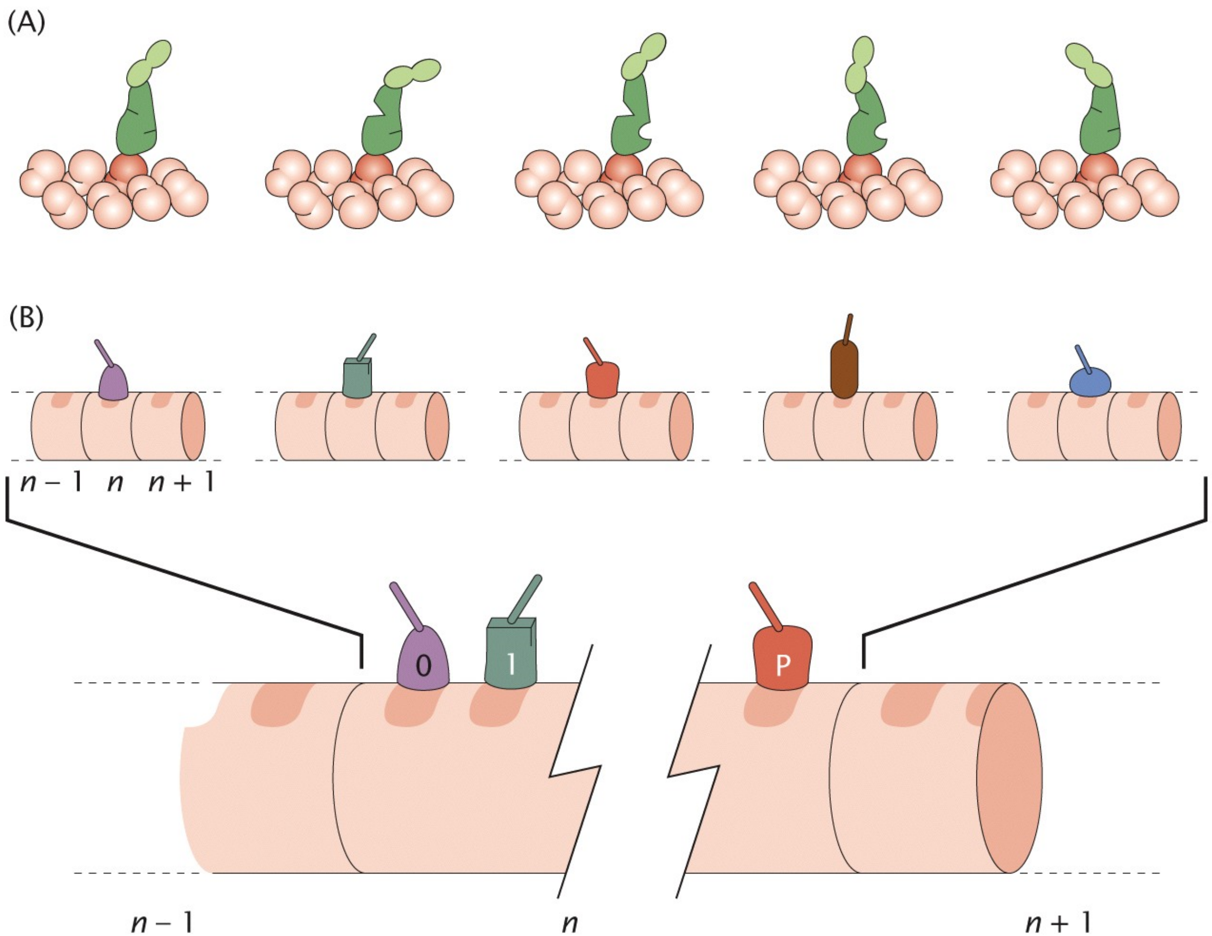



Figure 16.20 Physical Biology of the Cell, 2ed. (© Garland Science 2013)

# One-state model

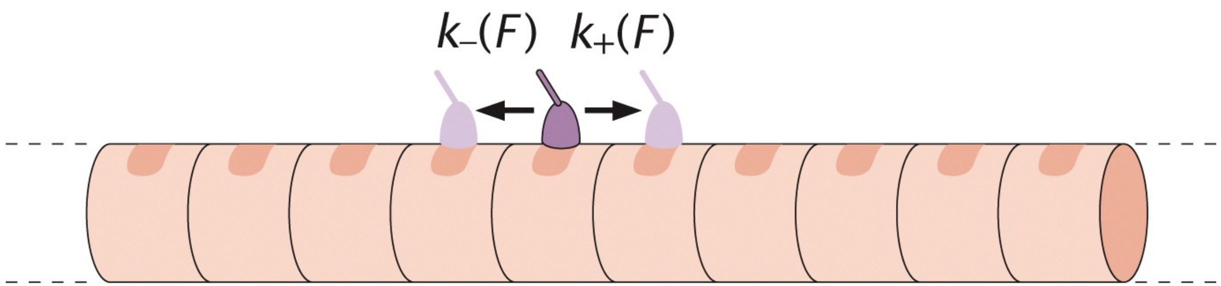



Figure 16.21 Physical Biology of the Cell, 2ed. (© Garland Science 2013)

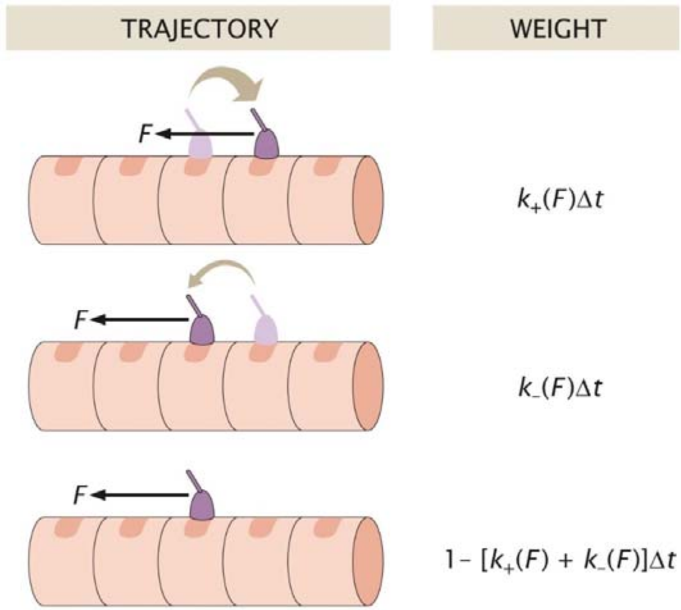



Figure 16.22 Physical Biology of the Cell, 2ed. (© Garland Science 2013)

# One-state model: position probability over time

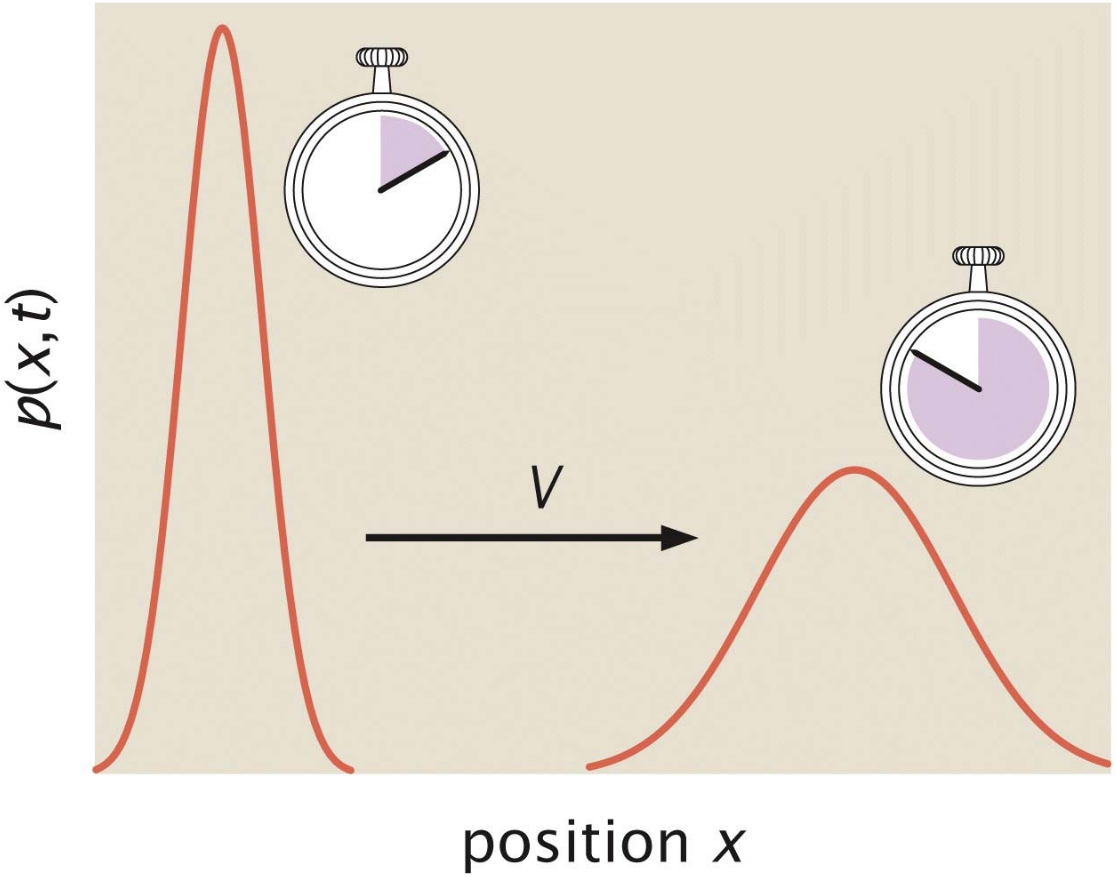
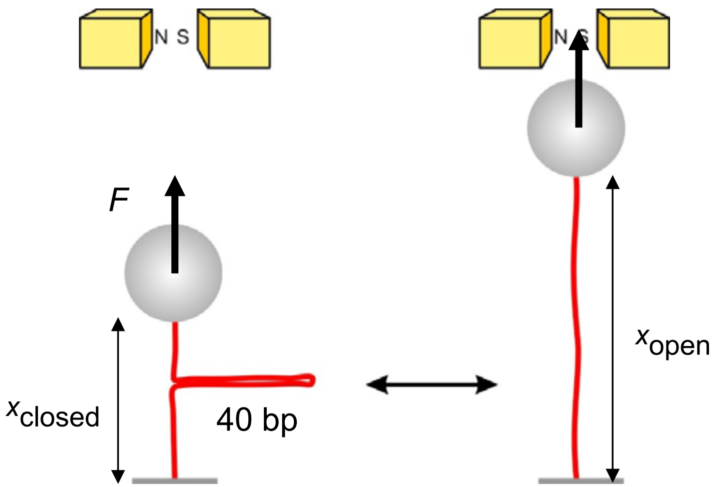
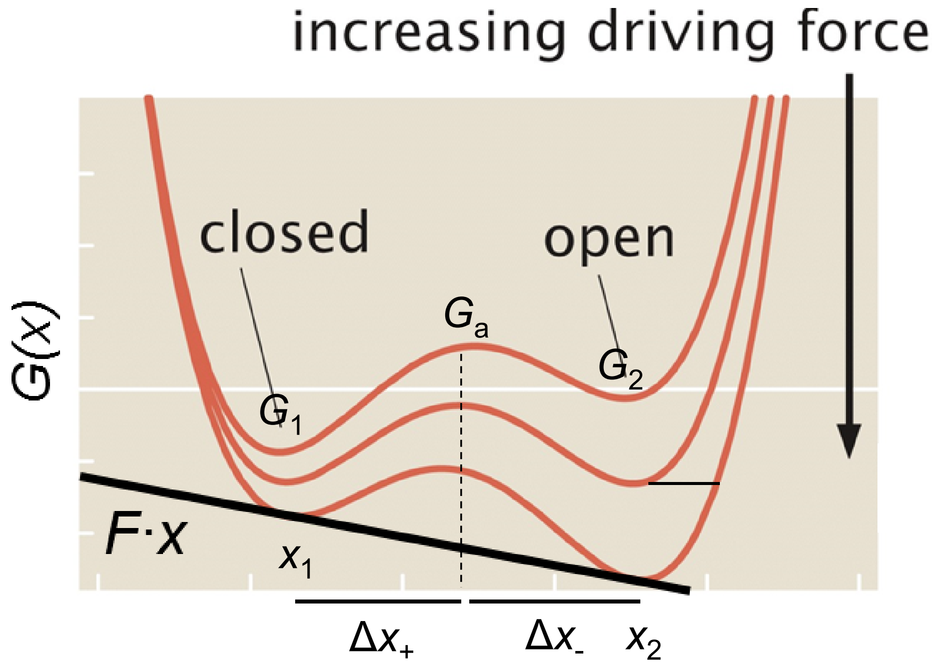



Figure 16.23 Physical Biology of the Cell, 2ed. (© Garland Science 2013)

# Remember: Force dependence of reaction rates

For external driving force, e.g. a mechanical force



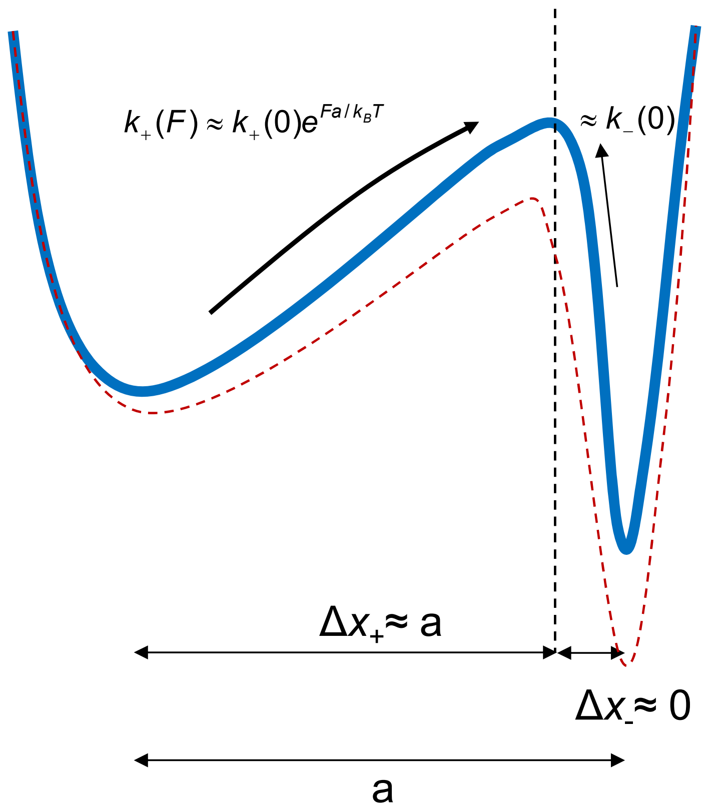




$$k_+(F) = k_+(0)e^{F\Delta x_+/k_BT}$$
$$k_-(F) = k_-(0)e^{-F\Delta x_-/k_BT}$$

# Force-dependence on forward or backward step

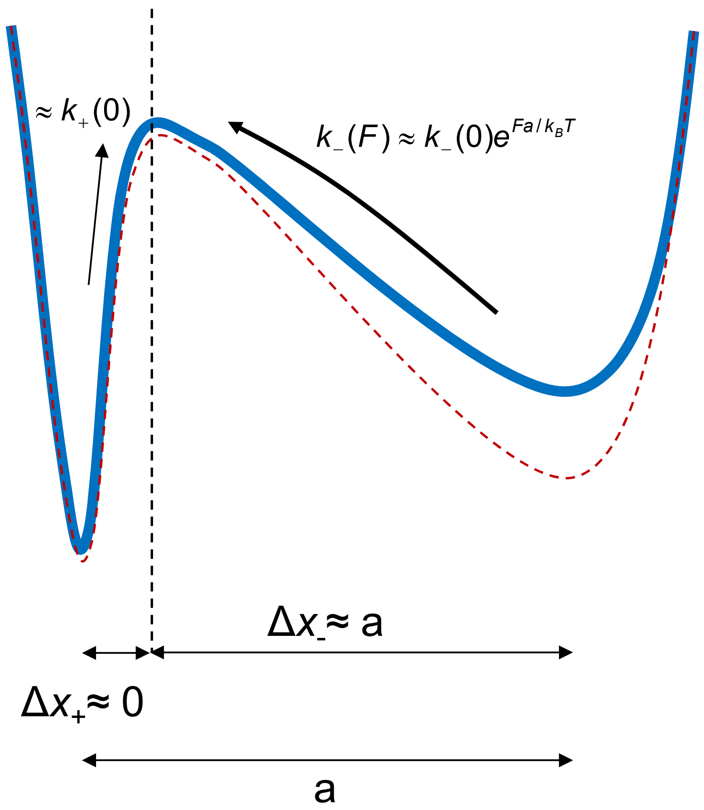
## Force-dependence on forward step

Long diffusion into forward position
**Force helps a lot**




## Force-dependence on backward step

Small distance to reach forward position
**Force assists only little**

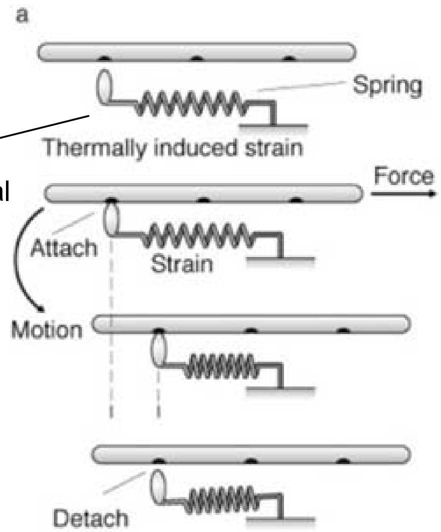


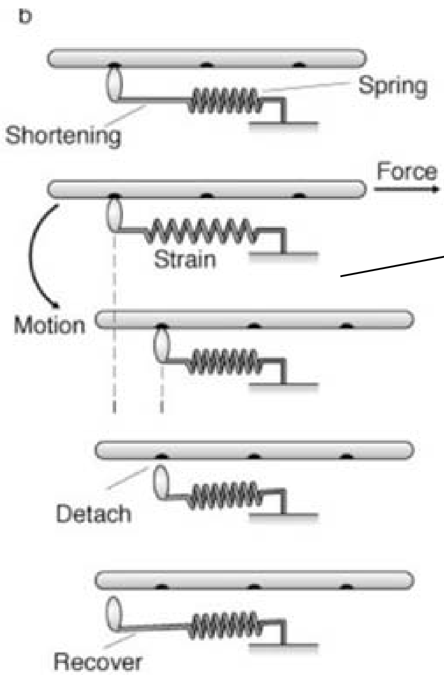
# Thermal ratchet versus power stroke (the two extremes)

**Thermal ratchet**
(Brownian motor)

**Power stroke**
(adiabatic mechanism)

Large diffusive conformational change into new state. Energy source blocks backward diffusion




Small internal energy-source driven conformational change produces strained state that relaxes into new equilibrium state

Huxley 1957
Cordova et al. 1992

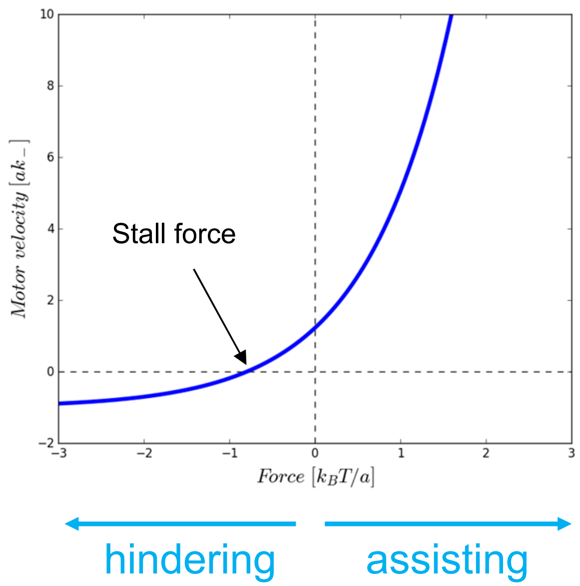
Eiesenberg & Hill 1978
Parmeqqiani et al. 1999

**Force dependence mainly at forward step**

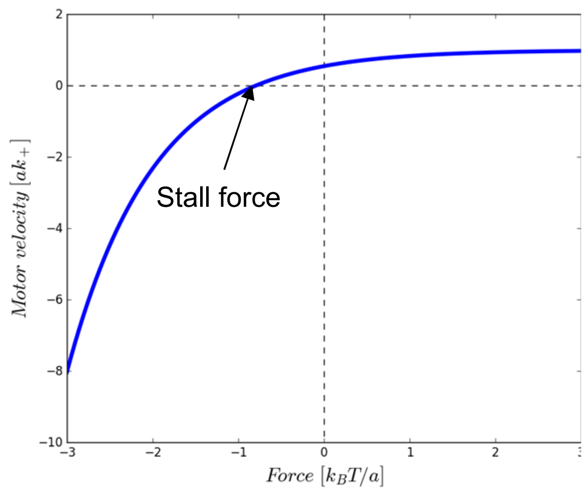
**Force dependence mainly at backward step**

# Force-dependence of motor velocity (one-step model)

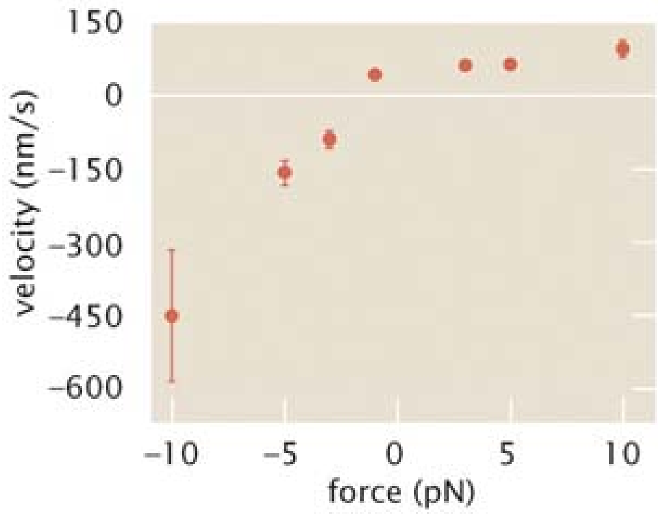
only forward step force-dependent



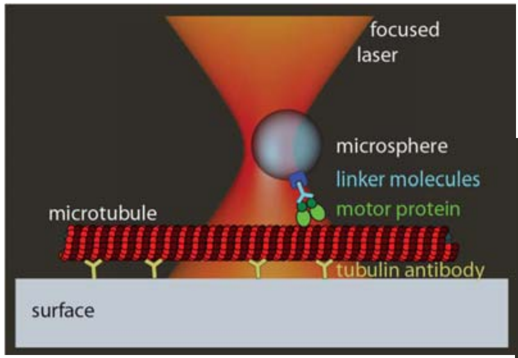

only backward step force-dependent



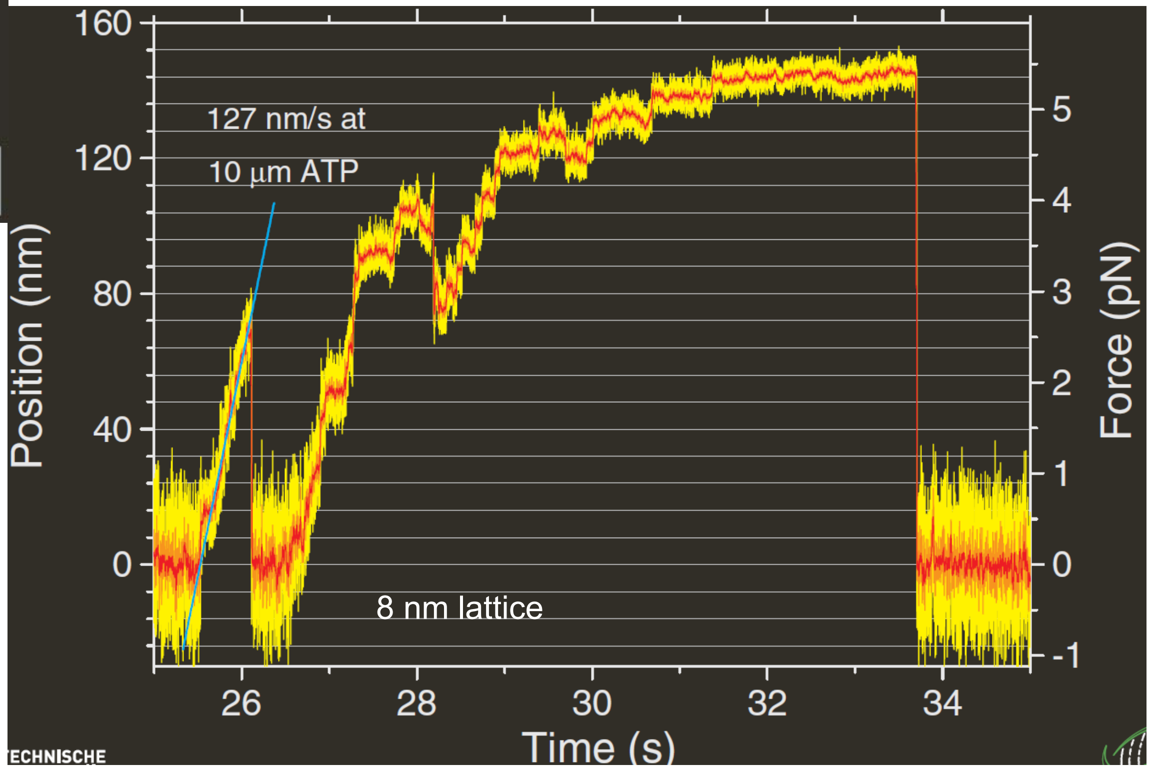

Myosin V data



# How to measure the force of motor proteins



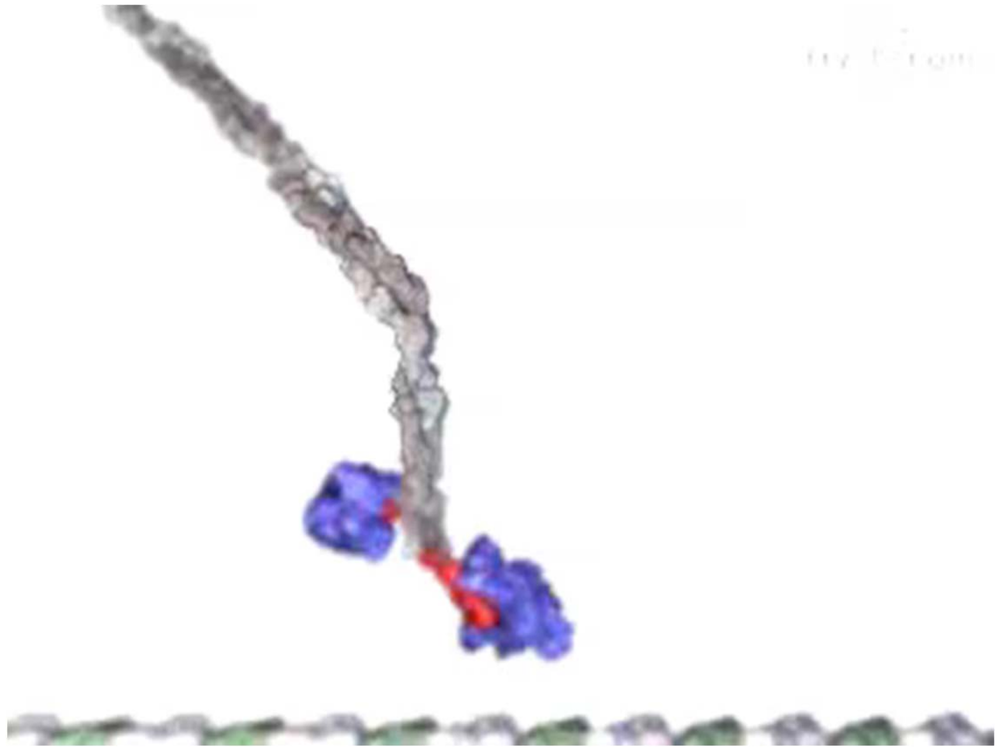

Force-velocity curve




E. Schäffer, Uni Tübingen

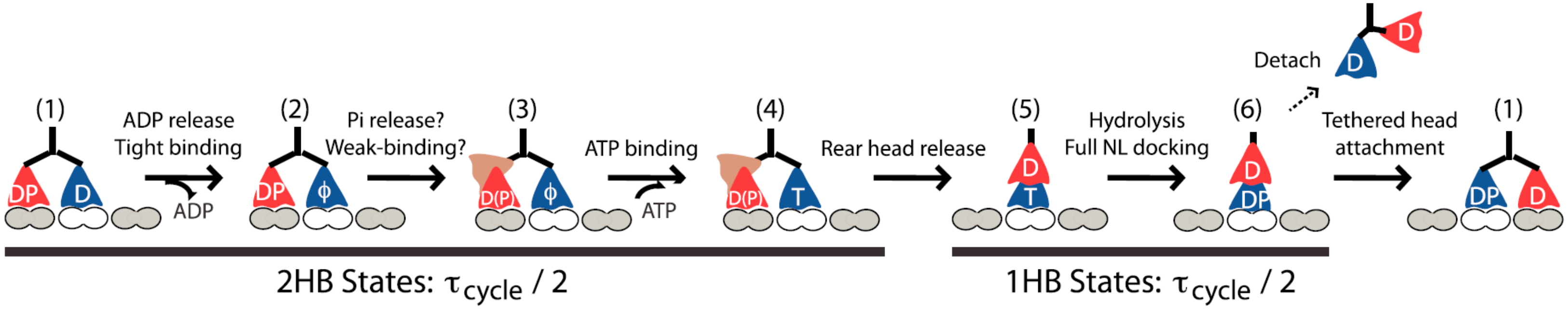
Stall force measurements reveal that kinesin has an energy efficiency of about 50%

# Kinesin stepping



1 ATP per single 8 nm step

# The latest kinesin stepping model




Strongly bound states: empty (Φ) & ATP
Weakly bound states: ADP (D) & ADP+$P_i$

Coordination:
ATP binding causes conformational change in front head (so-called power stroke), such that it pulls on rear head (step 4)
+ stress on rear head causes fast rear head detachment

Movement:
Hydrolysis in front head (step 6) promotes preferential „rear“ head binding in front of front head