Multiscale Motility of Molecular Motors

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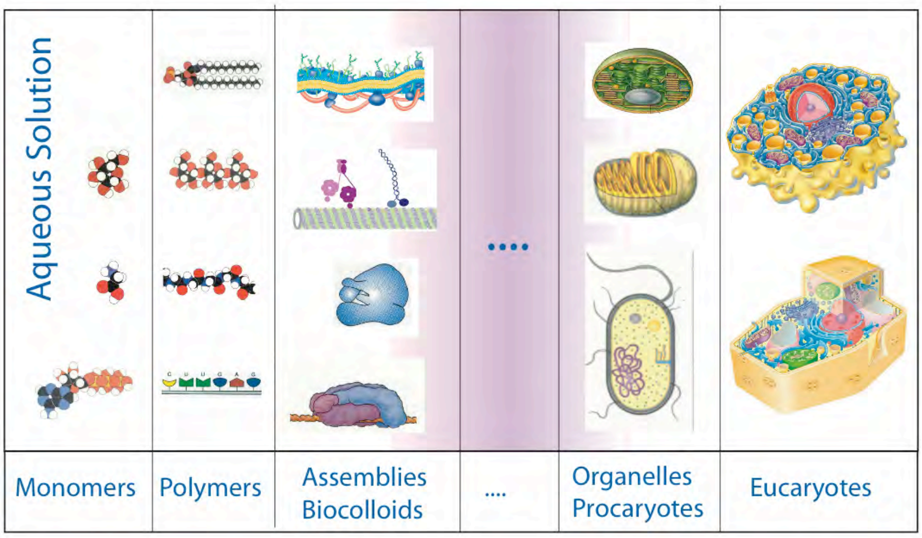
MPI of Colloids and Interfaces, Theory & Bio-Systems

- Introduction
- Single Molecular Motors
- Cargo Transport by Motor Teams
- Motor Traffic

Bio-Nano: From Molecules to Cells

Hierarchy of Structures, Bottom-Up:

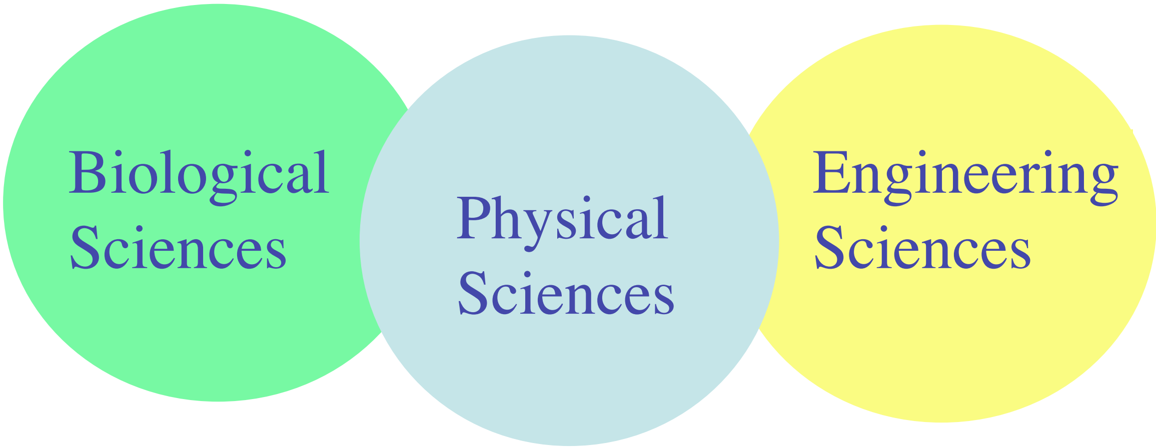


... Matter

Transition regime
Complexity gap

Life ...

Different Disciplines



Discovery Understanding Construction

3

Understanding

Crucial insight provided by physical sciences:

~1840 "Energy is conserved in organisms"

Helmholtz, du Bois-Reymond, Mayer

-> End of vitalism

~1940 "Genes are molecules"

Bohr, Delbrück, Schrödinger

Watson + Crick

-> Beginning of Molecular Biology

Construction on Small Scales

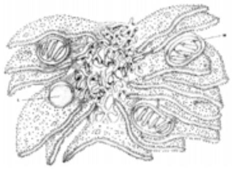
Milestones of Human Engineering:

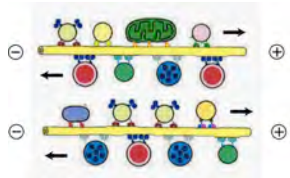
- Architecture

- Energy Conversion

- Information Processing

Bio-Nano:



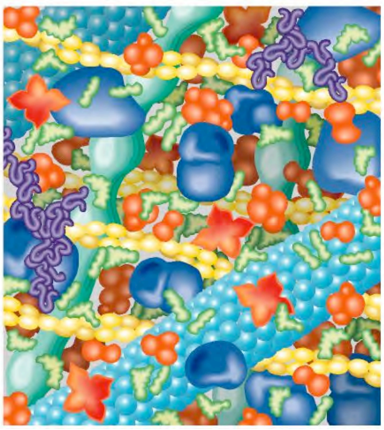


Feynman: ‘Plenty of Room at the Bottom’

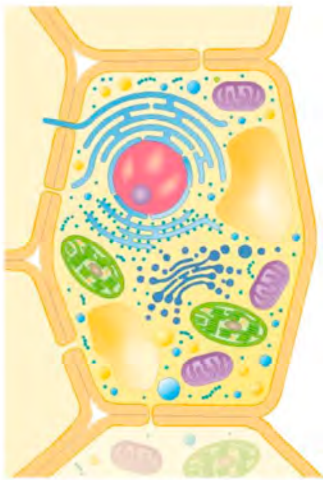
Order and Disorder

- Polymer Level:
  Native / Denatured
- Assembly Level:
  Spatial Architecture

2 nm



25 nm



25 µm

Selforganization

- Assembly of Building Blocks "by themselves"
- Instructions only from local environment

(1) Selforganization via Molecular Interactions

Structure formation close to equilibrium

Examples: Protein folding, Membrane assembly, ...

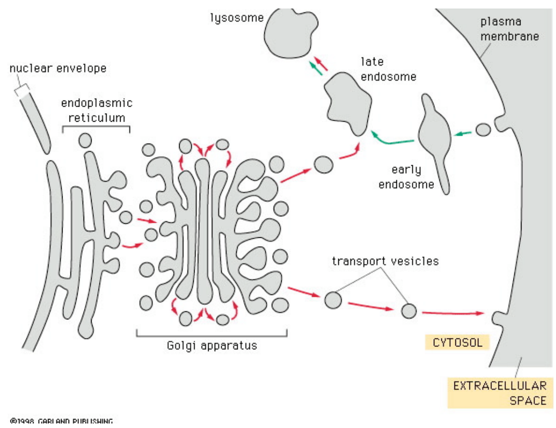
(2) Selforganization via Energy Conversion

Structure formation far from equilibrium

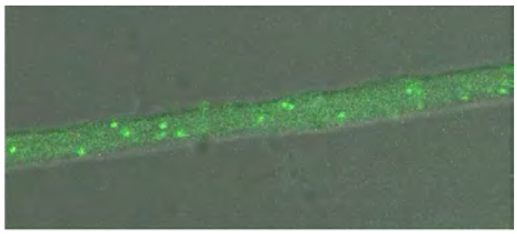
Examples: Molecular motors, Filament assembly, ...

'Order by Motion'

- Intracellular patterns of organelles and vesicles:



- Spatio-temporal order created by mol motors:



Example: Axon

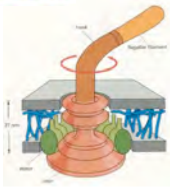
Smith et al, *PNAS.* ***98*** (2001)

Molecular Machines

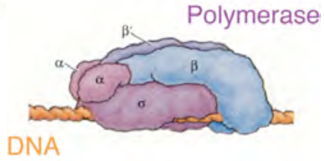
"Every Motion has its Motor"

Protein Oligomers + Substrate:

- Nano-Motors:
  - Stepping motors: Kinesin, Dynein, ...
  - Rotary motors: Bacterial flagellae

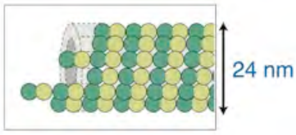


- Nano-Pumps: Na-K-Pump, ...
- Nano-Assemblers: Polymerases, ...



Assembly of Many Proteins:

- Growing filaments

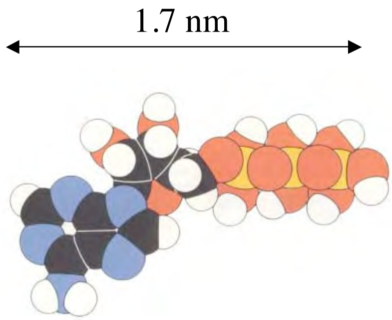


Chemomechanical Coupling

- Molecular machines:

  Conversion of chemical energy
  into mechanical work

- Universal chemical energy source
  provided by ATP:



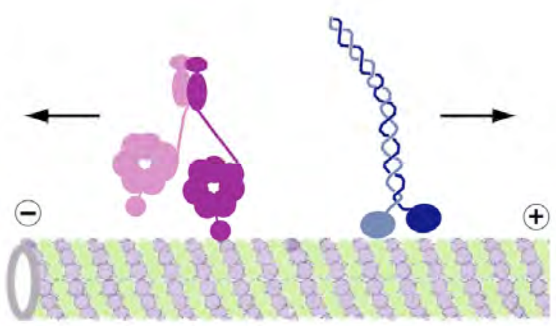
- Hydrolysis of ATP: ATP -> ADP + P
- Synthesis of ATP: ADP + P -> ATP

Nucleotides
ATP, ADP, P

"Human body hydrolyses and synthesizes 60 kg of ATP per day!"

Stepping Motors

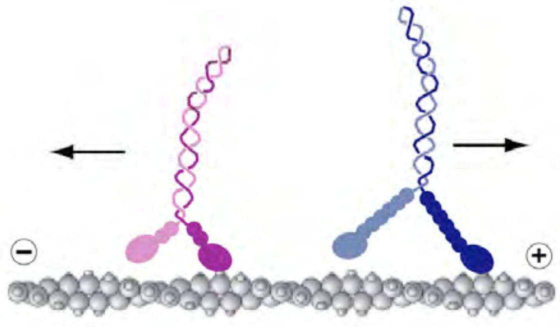
• Filament = Microtubule



Dyneins to minus end

Kinesins to plus end

• Filament = F-Actin



Myosin VI to minus end

Myosin V to plus end

- Filaments are polar: Plus- und Minus-Ends (no charges)
- No load: Each motor steps into a prefered direction
- Each motor has two heads that hydrolyze ATP
- Each motor makes discrete steps with fixed step size

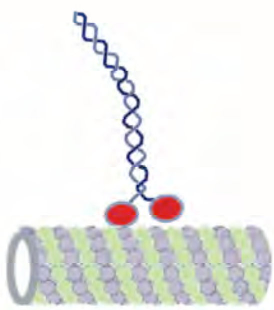
Multiscale Motility

ATP Binding

Nucleotide Binding Pocket ~ 1 nm

10^{-3} s

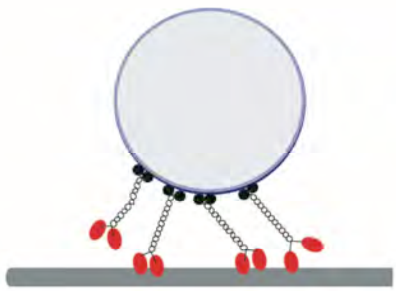
Mechanical Steps



Single head moves by 16 nm

10^{-6} s

Transport

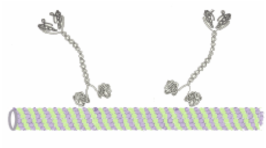


Cargo transport over cm or m !

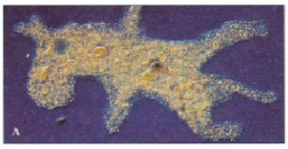
10^4 - 10^6 s

Hierarchy of Time Scales ≠ Hierarchy of Length Scales

Hierarchy of Force Generation



- Single stepping motor generates 10^{-12} Newton = PicoNewton



- Teams of stepping motors + bundles of filaments generate 10^{-9} Newton = NanoNewton

- Muscles of human body generate 10^{2} Newton

Cooperative action of many motors generates forces between 10^{-12} und 10^{2} Newton:

Single Motors

- Single motor experiments
- Chemomechanical coupling
- Network of motor cycles
- Run length and unbinding rate

Motor Teams

- Uni-directional cargo transport
- Bi-directional cargo transport
- Enhanced transport

Motor Densities

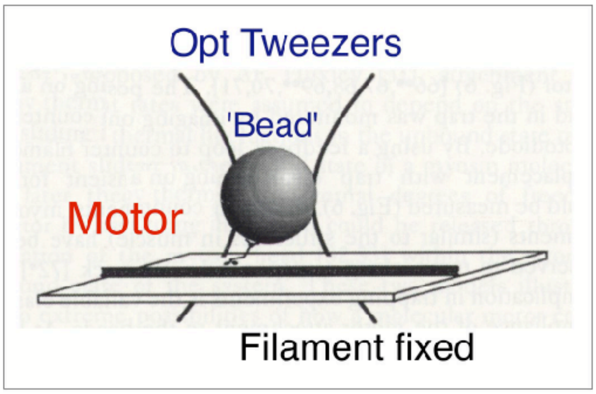
- Traffic of motors and cargos

Discrete Steps (nm): Single motor experiments, Chemomechanical coupling, Network of motor cycles

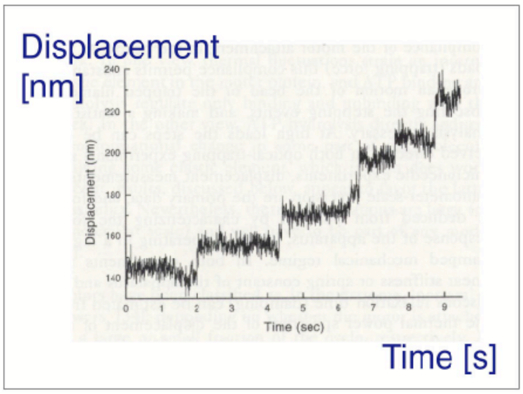
Large-Scale Transport (μm to m): Run length and unbinding rate through Traffic of motors and cargos

Experiment: Kinesin

- Bead Assay:



- Discrete Steps:



Svoboda ... Block, *Nature* ***365*** (1993)

- Kinesin's center-of-mass moves by 8 nm
- Each head moves by 16 nm (hand-over-hand motion)
- Hydrolysis of one ATP per step (tight coupling)

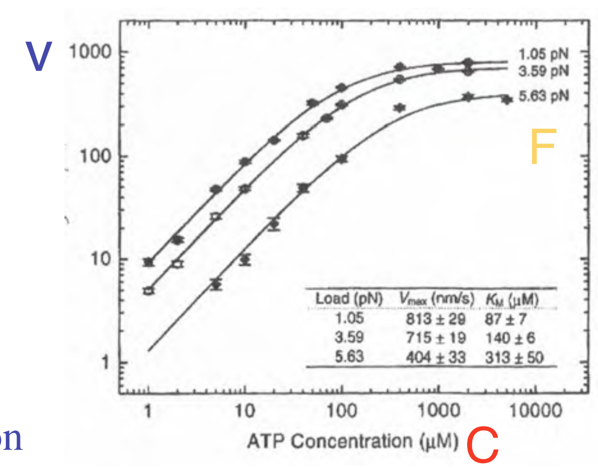
[ATP] Dependence of Velocity

- Velocity v as a function of concentration [ATP] = C and external force F

$$v(C,F) \simeq v_{sat}(F) \frac{C}{C_*(F) + C}$$

'Michaelis-Menten Relation'

Visscher et al, *Nature* ***400*** (1999)



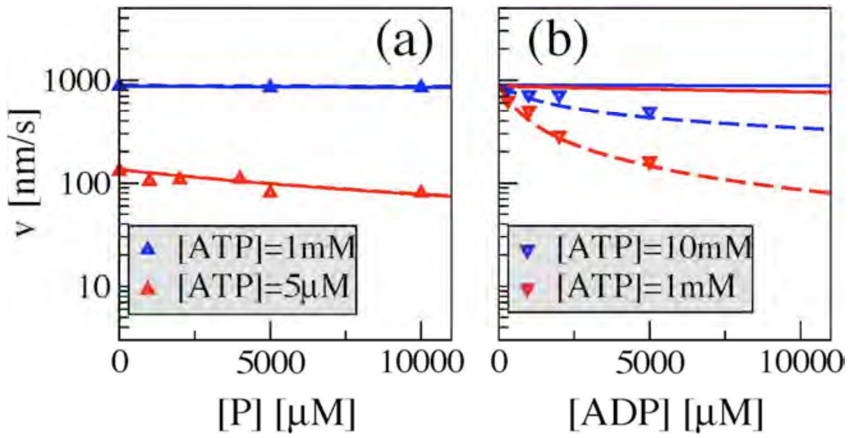
- Simple functional dependence on two variables C and F
- Predicted by a large class of motor models

RL, *PRL*. ***85*** (2000)

[ADP] and [P] Dependence

Schief … Howard, *PNAS* ***101*** (2004)

- Motor velocity decreases slowly with increasing [P]
- Motor velocity decreases strongly with increasing [ADP]

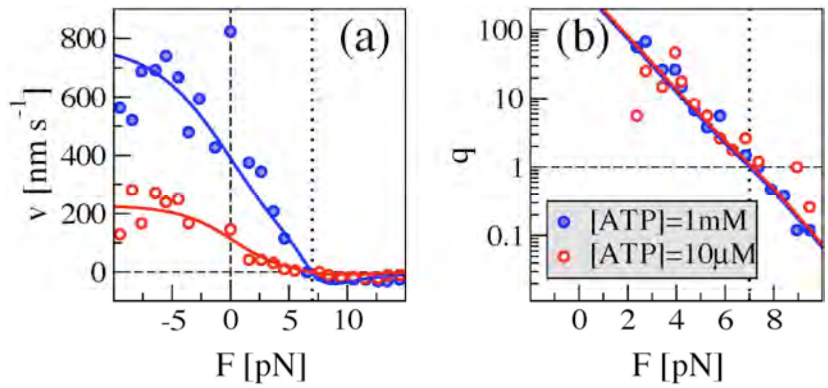


Load Force Dependence

Nishiyama ... Yanagida, *Nat. Cell Biol.* ***4*** (2002)

Carter and Cross, *Nature* ***435*** (2005)

Resisting Load Force F > 0



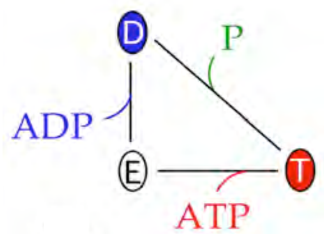
- Kinesin generates force of about 7 pN = stall force F_s
- Kinesin makes processive backwards steps
- Mechanical steps are very fast (faster than 15 μs)

Theory: Single Motor Head

- Single head of kinesin with one nucleotide binding pocket (NBP): empty, occupied by ATP or ADP

NBP

- Chemical network with three motor states:

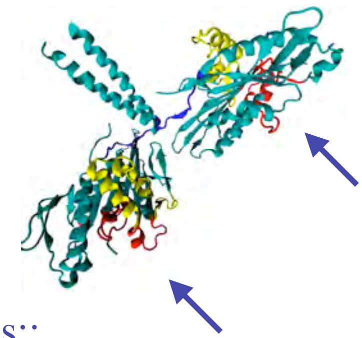


empty (E)
occupied by ATP (T)
occupied by ADP (D)

- Each edge = two directed edges = forward + backward transition
- One motor cycle = two directed cycles or dicycles

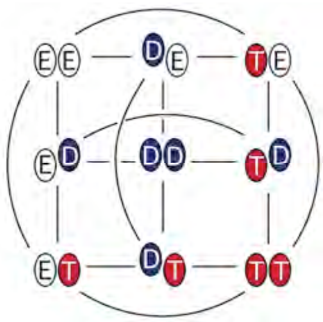
Two Motor Heads

- Two motor heads with two NBPs each of which can be E, T, or D



Liepelt and RL, *EPL 77* (2007)

- Chemical network with 9 motor states::



3^2 = 9 states EE, DE, ...

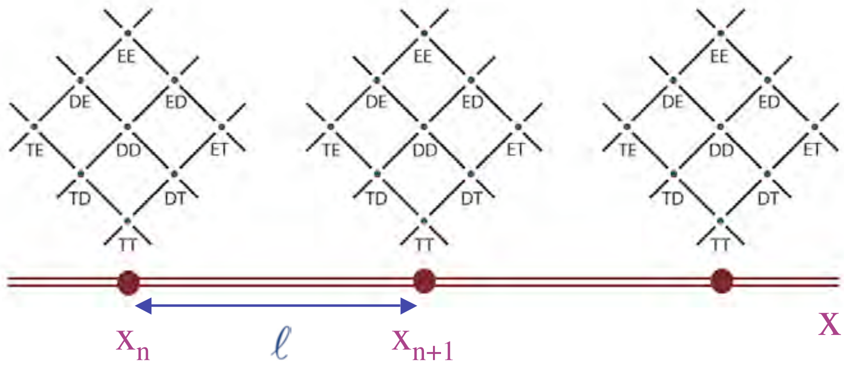
18 edges, 36 chemical transitions

More than 200 cycles !

Chemomechanical Networks

- Mechanical transitions = Spatial displacement x along filament
- Discrete step size ℓ defines lattice of motor positions:

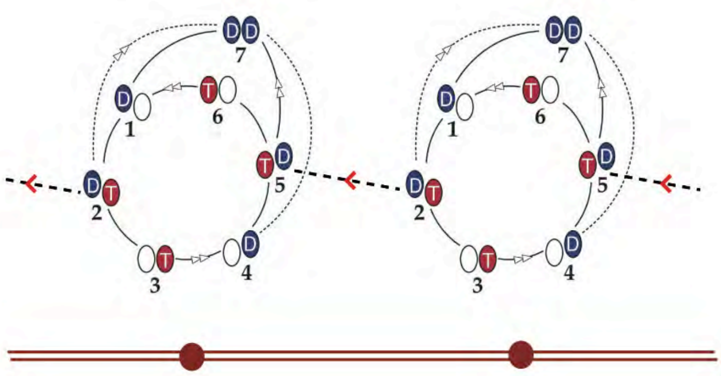
RL et al , *J. Stat. Phys.* ***135*** (2009)



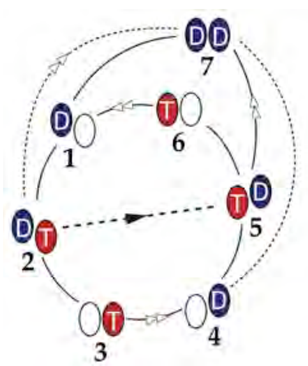
- Mechanical transitions from
  chemical state at site x_n to chemical state at site x_{n+1}
- Specific motor governed by certain sub-network

CM Networks for Kinesin

- Sub-network with seven motor states
- Mechanical stepping from DE to ED = broken edge



Extended network



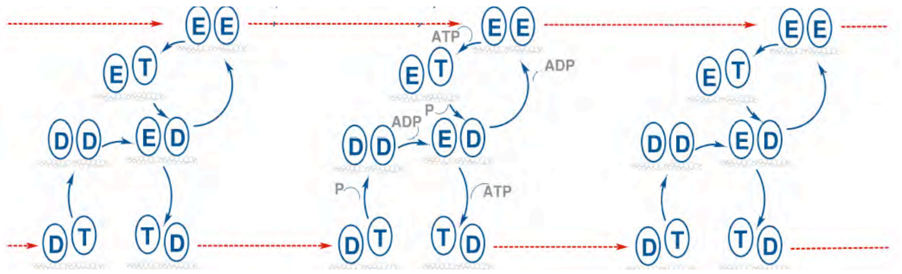
Compact network

Liepelt and RL , *Phys. Rev. Lett.* ***98*** (2007)

CM Network for Myosin V

V. Bierbaum

- Sub-network with six motor states
- Two types of mechanical steps (red lines):



Motor Dynamics

- Markov process on on CM network with motor states i
- Master equation for probabilities P_i :

$$d P_i / dt = - \sum_j \; [P_i \, \omega_{ij} - P_j \, \omega_{ji}]$$

Transition rates ω_{ij}

- Local excess fluxes $\Delta J_{ij} = P_i \, \omega_{ij} - P_j \, \omega_{ji}$ for steady state determine motor properties as measured in single mol exp
- Example 1: Motor velocity v $= \sum^{f}_{lij>} \ell_{ij} \; \Delta J_{ij}$
- Example 2: Hydrolysis rate $h = \sum^{h}_{lij>} \; \Delta J_{ij}$
- Operation modes, efficiency, ..,

Liepelt and RL , *Phys. Rev. E.* ***79*** (2009)

Classification of Motor Cycles

- Each directed cycle C_ν^d , balance condition:

$$k_B T \ \ln(\Xi_\nu^d) \ = \ \mu(C_\nu^d) - W(C_\nu^d)$$

Transition rates — Chemical energy — Mechanical work

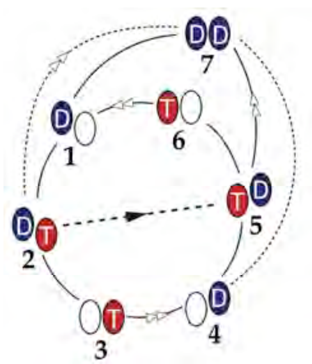
RL, Liepelt: *J. Stat. Phys.* ***130*** (2008)

Classification of cycles:

- Detailed balance: $\mu(C_\nu^d) = 0$ and $W(C_\nu^d) = 0$
- Mech nonequilibrium: $\mu(C_\nu^d) = 0$ and $W(C_\nu^d) \neq 0$
- Chem nonequilibium: $\mu(C_\nu^d) \neq 0$ and $W(C_\nu^d) = 0$
- Chemomech coupling: $\mu(C_\nu^d) \neq 0$ and $W(C_\nu^d) \neq 0$

Kinesin: Several Motor Cycles

Liepelt and RL , *Phys. Rev. Lett.* ***98*** (2007)



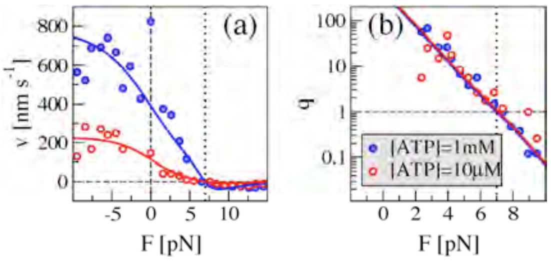
Three chemomechanical motor cycles

Dominat cycle depends on Concentration of ATP, ADP, and P

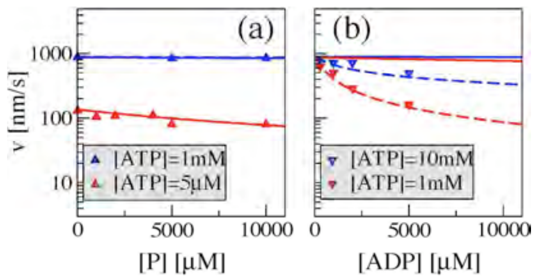
- Small ADP and P, small load force F: dicycle |25612>
- Small ADP and P, large load force F: dicycle |52345>
- Large ADP, small load force F: dicycle |25712>

Kinesin: Theory + Experiment

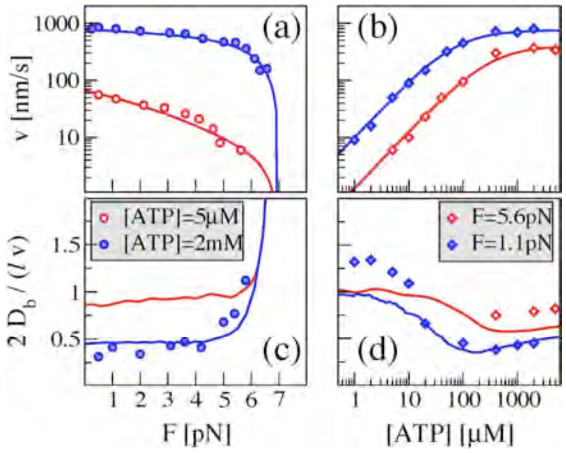
• Data of Carter, Cross (2005)



• Data of Schief et al (2004)



• Data of Visscher et al (1999)



• Data of Schnitzer et al (2000) on run length as a function of force and [ATP]

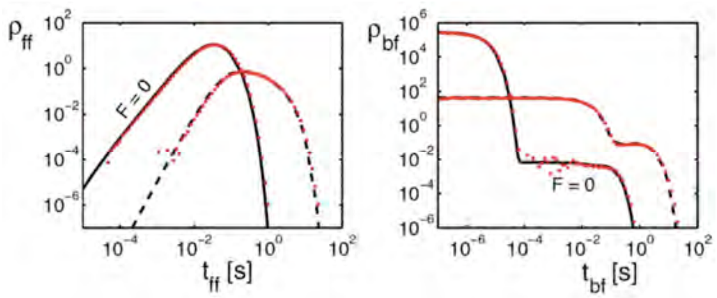
Stepping Process

Valleriani et al , *EPL* ***82*** (2008)

• Experiments can resolve forward and backward mechanical steps but cannot distinguish different chemical transitions

• Markov process on motor network generates mechanical stepping process that is non-Markovian

• Four different pairs ff, fb, bf, and bb of successive steps => four dwell time distributions



All dwell time distributions are non-exponential

Single Motors

- Single motor experiments
- Chemomechanical coupling
- Network of motor cycles

Discrete Steps (nm)

- Run length and unbinding rate

Motor Teams

- Uni-directional cargo transport
- Bi-directional cargo transport
- Enhanced transport

Motor Densities

- Traffic of motors and cargos

Large-Scale Transport (µm to m)

Thermal Noise and Run Length

- Molecular motor has finite binding energy
- Thermal noise leads to unbinding from filament

- Unbinding is a stochastic process:

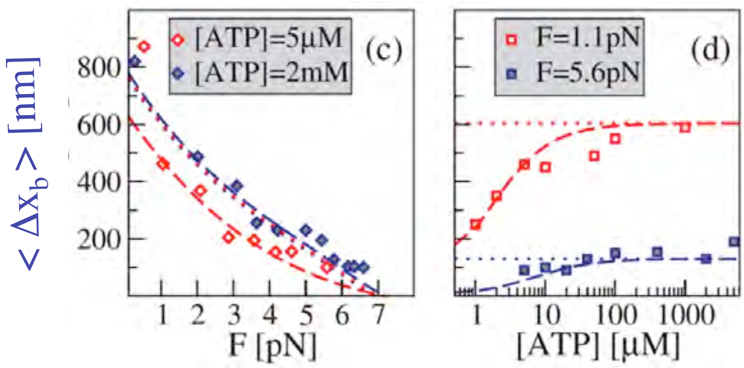
  at each step, unbinding probability ε

- Motor has finite run length (or walking distance)

  Single kinesin: about 100 steps or 800 nm

Unbinding Rate

- Average run length $< \Delta x_b >$:



- Unbinding rate $\varepsilon \sim 1/ < \Delta x_b >$
- Force dependence: $\varepsilon(F) \sim \exp(F/F_d)$

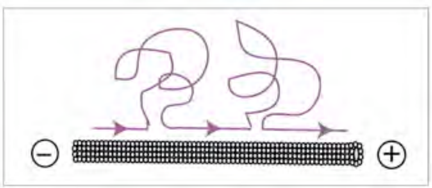
Detachment force F_d

RL et al , *J. Stat. Phys.* ***135*** (2009)

Composite Motor Walks

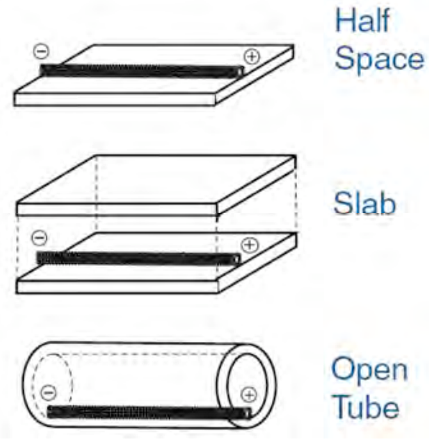
RL et al, *Phys. Rev. Lett.* ***87*** (2001)
Nieuwenhuizen et al, *EPL* ***58*** (2002)

- Single kinesin makes about 100 steps before it unbinds from filament



- Length scales >> run length: Alternating sequence of directed stepping and unbound diffusion

- Different compartments:



Single Motors

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Discrete Steps (nm)

Motor Teams

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- Enhanced transport

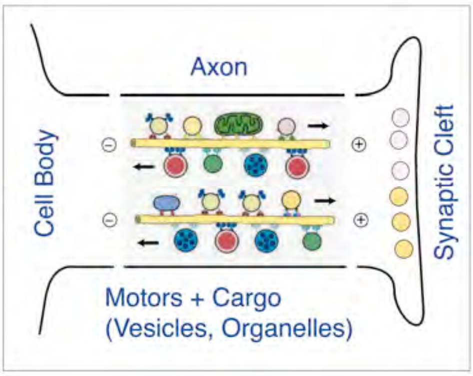
Motor Densities

- Traffic of motors and cargos

Large-Scale Transport (µm to m)

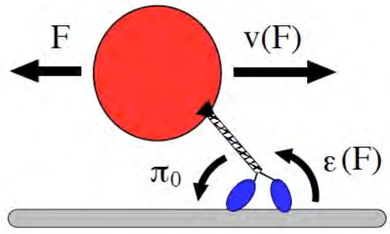
Intracellular Cargo Transport

- Example: Neuron, Axon, and Synapse



- Cargo transport by several motors:
  - Uni-directional transport by one motor team
  - Bi-directional transport by two motor teams
  - Enhanced transport by another motor team

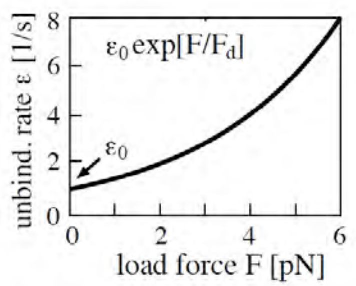
Single Motor Parameters



- Unbinding rate ε

  $\varepsilon(F) = \varepsilon_0 \; \exp(F / F_d)$

  Detachment force F_d

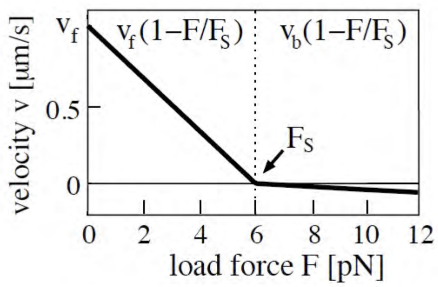


- Binding rate π_0
- Parametrization of velocity v:

  forward velocity v_f at zero load

  stall force F_s at which v vanishes

  backward velocity scale v_b



Cargo Transport by one Motor Team

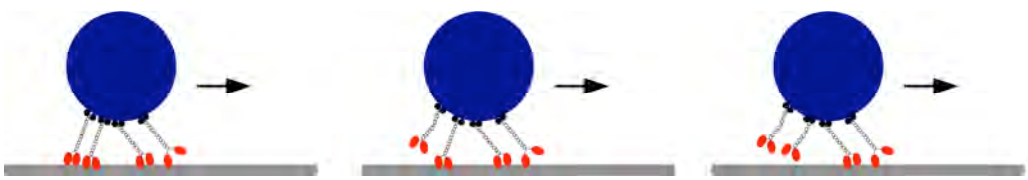
Klumpp, RL, *PNAS* ***102*** (2005)

- N identical motors firmly attached to cargo particle (vesicle, organelle)
- Thermal noise:

  Each motor unbinds and rebinds from filament

  => Number k≤ N of active motors is not fixed but fluctuates

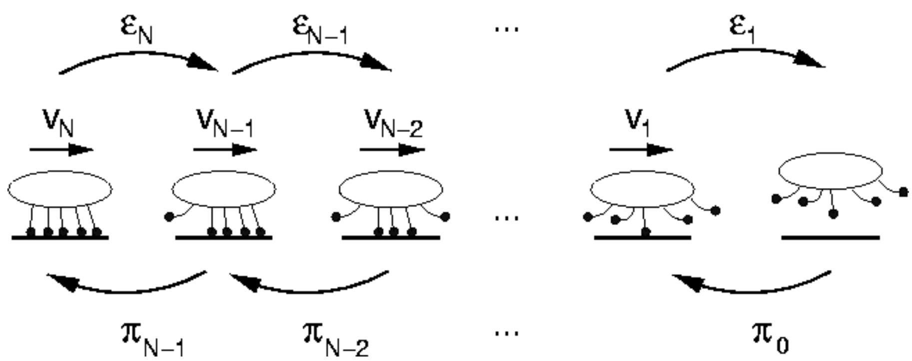
Ashkin et al. Nature 348 (1990)



Cargo States

- Up to N active motors:

|N> |N−1> |N−2>   … |1> |0>

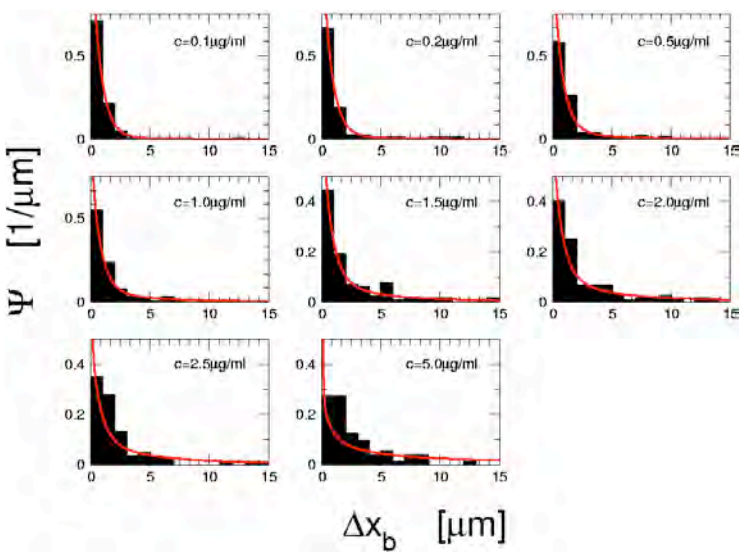


- State space: 1-dimensional lattice of cargo states
- Unbinding rates ε and binding rates π define Markov process

Run Length of Cargo

- Kinesin: Average run length $<\Delta x_b> \sim 5^N / N$ μm
  => N = 7 motors lead to run length of centimeters!

- Kinesin: Run length distribution



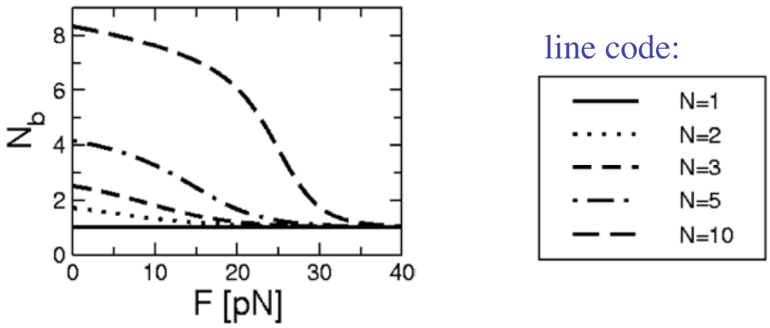
Comparison of Experiment and Theory

Beeg et al,
Biophys. J. ***94*** (2008)

External Load Force F

- Force F is shared by k ≤ N pulling motors
  => Each motor feels force F/k
- Unbinding rate ε ~ exp(F/k F_d)
  => Cascade of unbinding events
  k -> k−1 -> k−2 ...
- Average number of active motors:

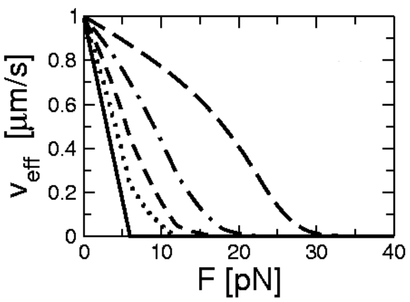
Effective, nonlinear motor-motor interaction



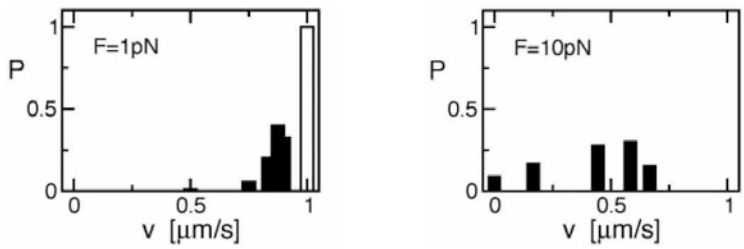
Klumpp, RL, *PNAS* **102** (2005)

Cargo Velocity v

- N = 1: Linear v(F) relation
  N > 1: Nonlinear v(F) relation
- For fixed F, velocity depends on N
- Velocity distribution in steady state:



Small F



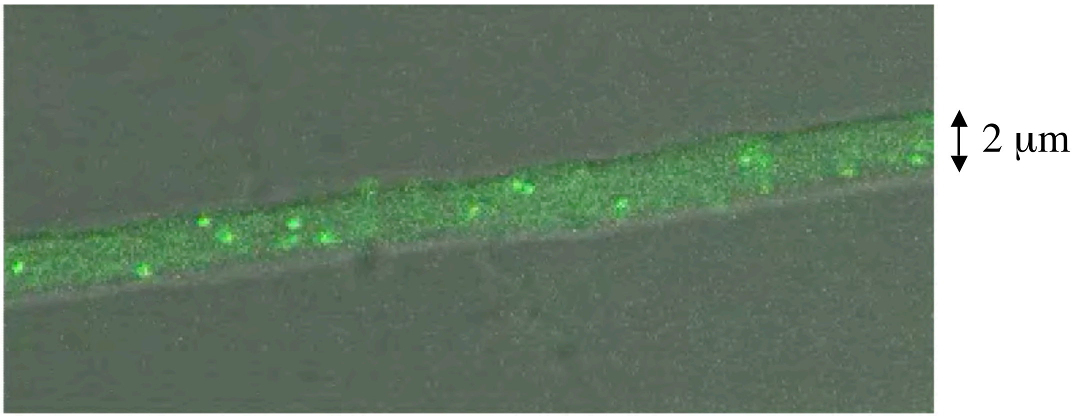
Large F

Distribution shifts towards smaller values, becomes broader and develops several peaks

Klumpp, RL, *PNAS* **102** (2005)

Axonal Cargo Transport

- Example: Transport of viruses in chick neurons
  Virus capsid labeled by GFP

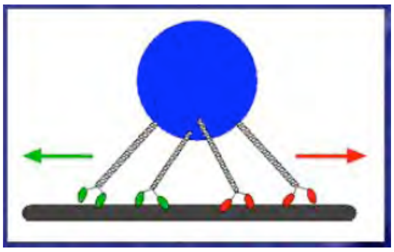


Smith et al, *PNAS*. ***98*** *(2001)*

Bi-Directional Transport

Müller et al, *PNAS* ***105*** (2008)
J. Stat. Phys. ***133*** (2008)

- Cargo with two antagonistic types of motors:



Green minus motors pull to the left
Red plus motors pull to the right

- Experimental observations reveal complex behavior:
  Different types of trajectories with and without pauses
  Changing one motor type affects both directions!

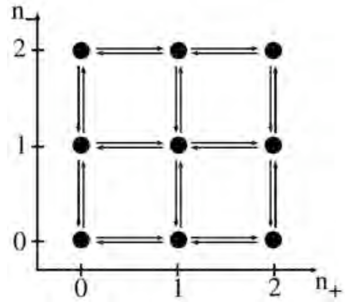
- Two proposals: Tug-of-war or coordination complex ?

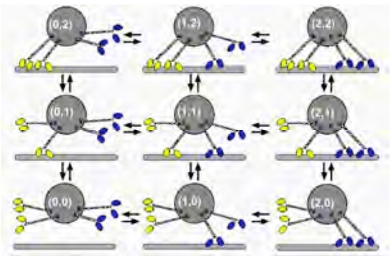
Stochastic Tug-of-War

- Thermal noise: # of minus and plus motors fluctuates in time
- Cargo states with (n_- , n_+) active motors, $n_- \leq N_-$ and $n_+ \leq N_+$

Example: (N_- , N_+) = (2,2)

2-dim lattice of cargo states





- Uni-directional transport for $N_- = 0$ or $N_+ = 0$
- All cargo states with $n_- > 0$ and $n_+ > 0$:

Plus motors pull on minus motors and vice versa

=> nontrivial force balance

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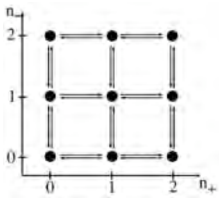
Tug-of-War: Steady States

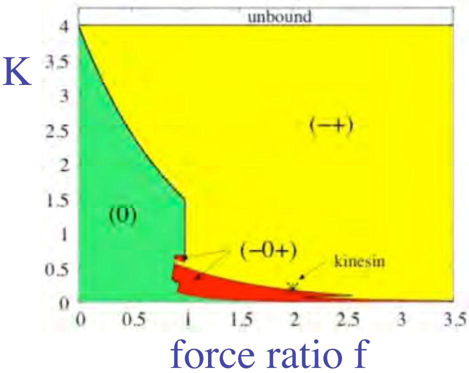


- St State distributions with 1, 2, or 3 maxima
- Four important parameters:

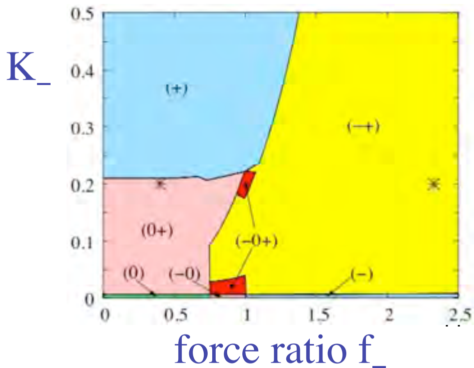
Motor numbers (N_- , N_+)

Desorption constant $K = \varepsilon / \pi$, Force ratio $f = F_s / F_d$

- $N_- = N_+ = 4$, symmetric motor + 'anti-motor'



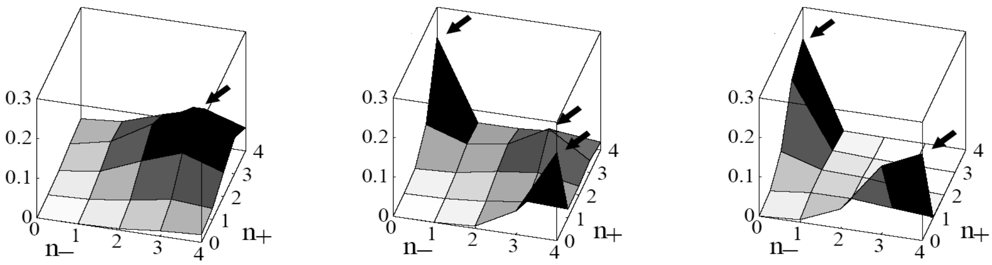
- $N_- = N_+ = 4$, asymmetric kinesin + variable dynein



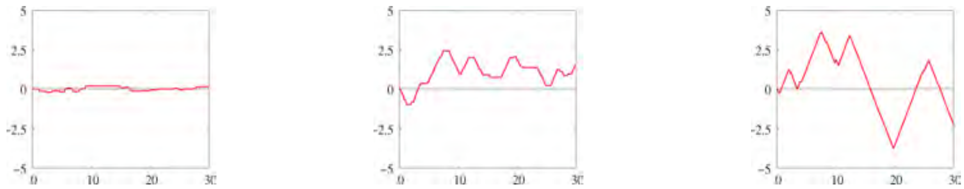
Müller et al, *PNAS* ***105*** (2008)

Example: 4 against 4 Motors

• Steady state distributions:



• Typical trajectories of cargo:



No motion	Bi-directional with pauses	Bi-directional without pauses

Müller et al, *PNAS* ***105*** (2008)

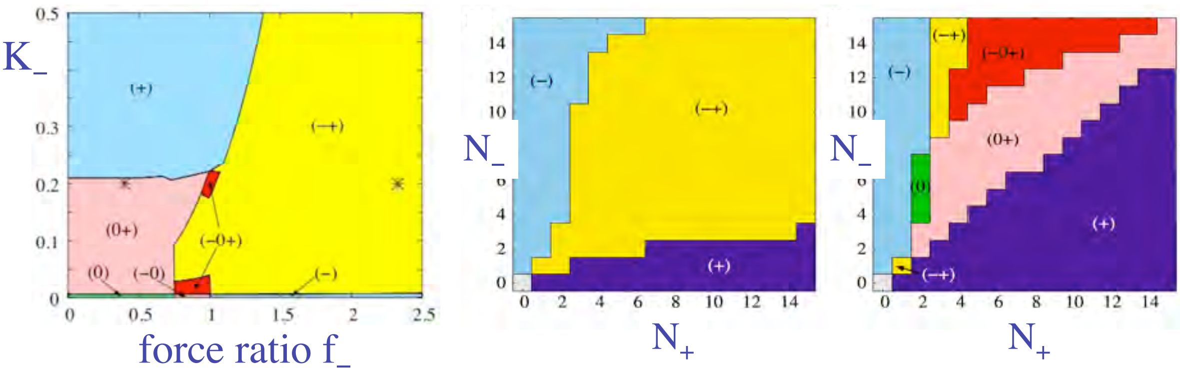
Motility Transitions

• Small changes in a single parameter: Cargo undergoes transition from one motility state to another one

• $N_- = N_+ = 4$, asymmetric kinesin + variable dynein

• N_+ kinesins + N_- strong dyneins

• N_+ kinesins + N_- weak dyneins



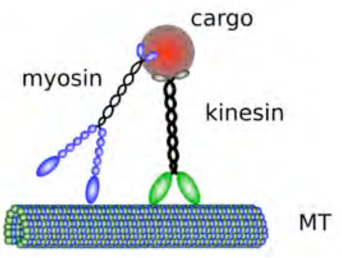
Müller et al, *J. Stat. Phys.* ***133*** (2008)

All experimental observations can be explained by small changes in single motor parameters !

Enhanced Transport

Berger et al, *EPL* ***87*** (2009)

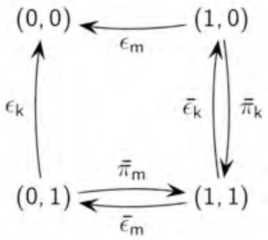
- Cargo + Kinesin + Myosin V



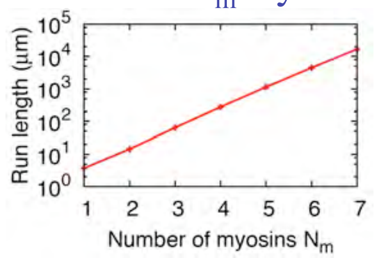
- Kinesin walks along MT
- Myosin V = diffusing anchor
- Increased rebinding of kinesin
  => Enhanced run length

Ali ... Trybus, *PNAS* ***105*** (2008)

- Cargo States for 1 kinesin + 1 myosin



- Generalization for 1 kinesin + N_m myosin

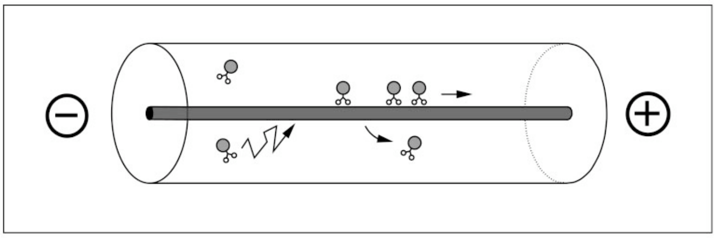


Single Motors	• Single motor experiments • Chemomechanical coupling • Network of motor cycles		Discrete Steps (nm)
	• Run length and unbinding rate		
Motor Teams	• Uni-directional cargo transport • Bi-directional cargo transport • Enhanced transport		Large-Scale Transport (μm to m)
Motor Densities	• Traffic of motors and cargos		

Example: Tube Geometries

RL et al, *Phys. Rev. Lett.* ***87*** (2001)
Klumpp, RL, *J. Stat. Phys.* ***113*** (2003)

- Axon-like tube compartment:



- Tube length >> run length:

  Motors (plus cargoes) completely unbind from filament, undergo unbiased diffusion, and eventually rebind to filament

- Repulsive motor-motor interactions: Simple exclusion processes
- Importance of boundary conditions

Traffic in a half open tube

Müller et al, *J. Phys. CM* ***17*** (2005)

- Axon-like boundary condition = half open tube

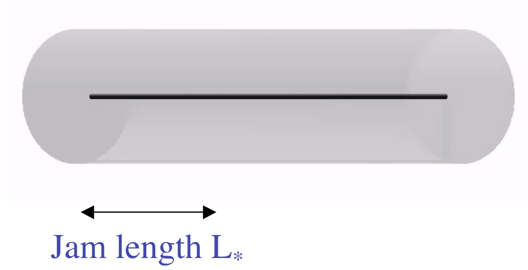
  left boundary open, reservoir of motors = ‘cell body’

  right boundary closed = ‘Synapse’

- (+) Motors (kinesins) moving to the right:

traffic jams

- (–) Motors (dyneins) moving to the left

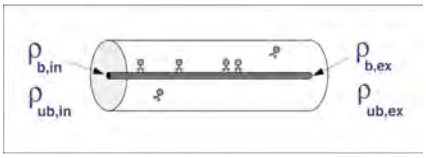


limited entry

More Traffic Phase Transitions

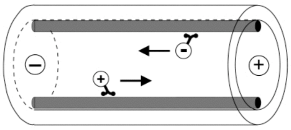
J. Stat. Phys. ***113*** *(2003)*

• Tube with two open boundaries:
TP transitions related to ASEP phases



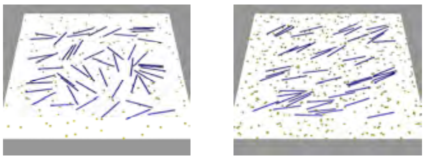
Europhys. Lett. ***66*** *(2004)*

• Traffic of two motor species in tubes:
Symmetry breaking TP transition

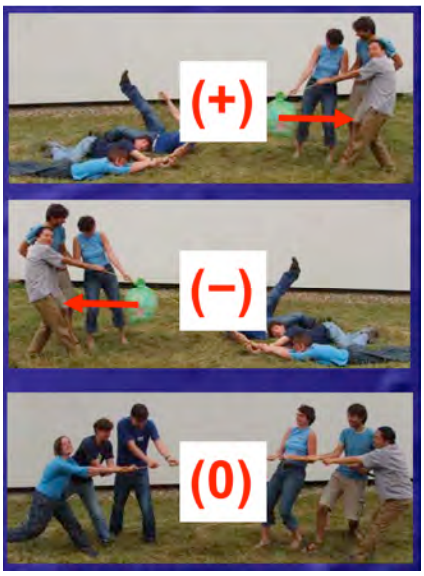


Phys. Rev. Lett. ***96*** *(2006)*

• Traffic of filaments along substrates:
Isotopic nematic TP transition



Coworkers



Stepping Motors, Theory:

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