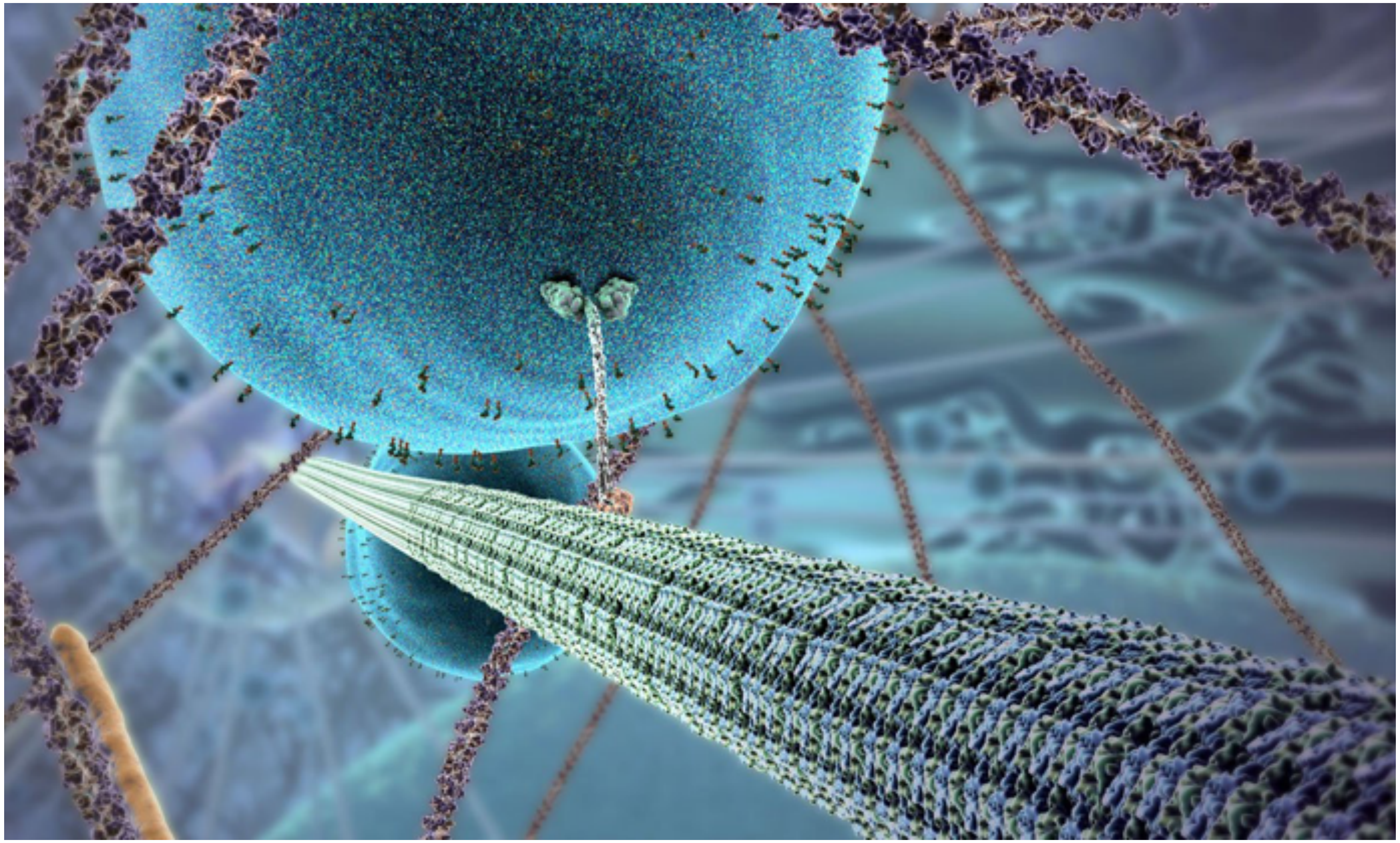
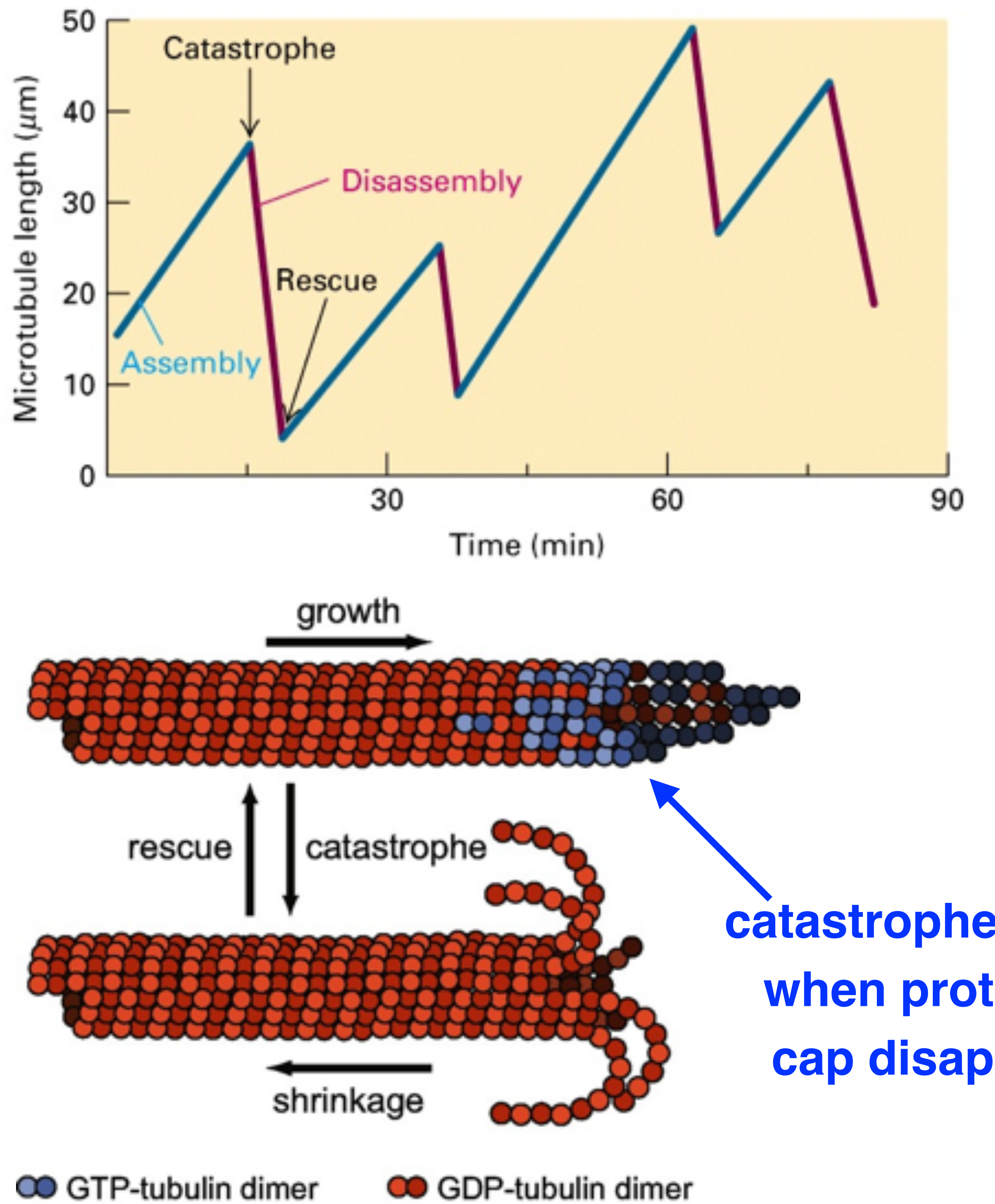
MAE 545: Lecture 7 (10/8)

Dynamics of microtubules and molecular motors

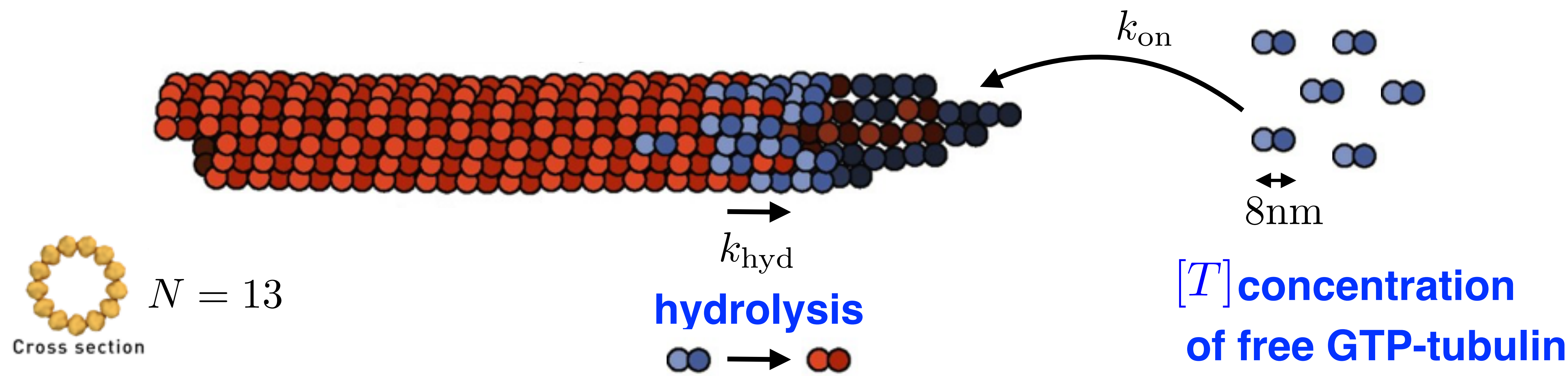


Microtubule dynamic instability

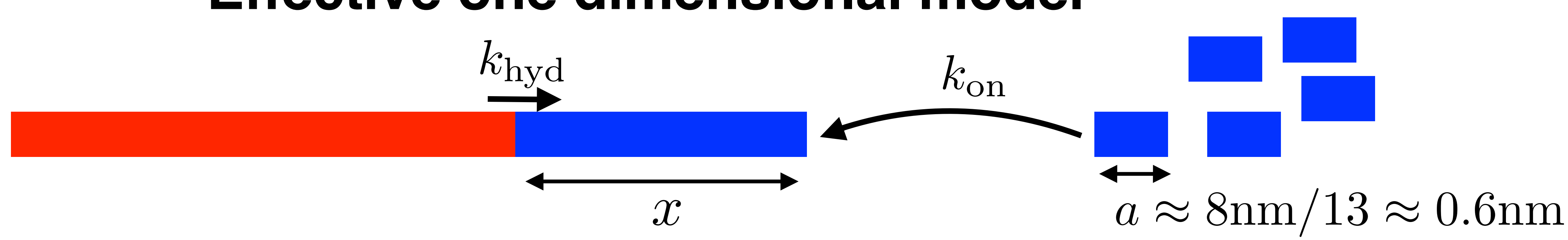


catastrophe occurs when protective cap disappears

Molecular model for growth of protective cap



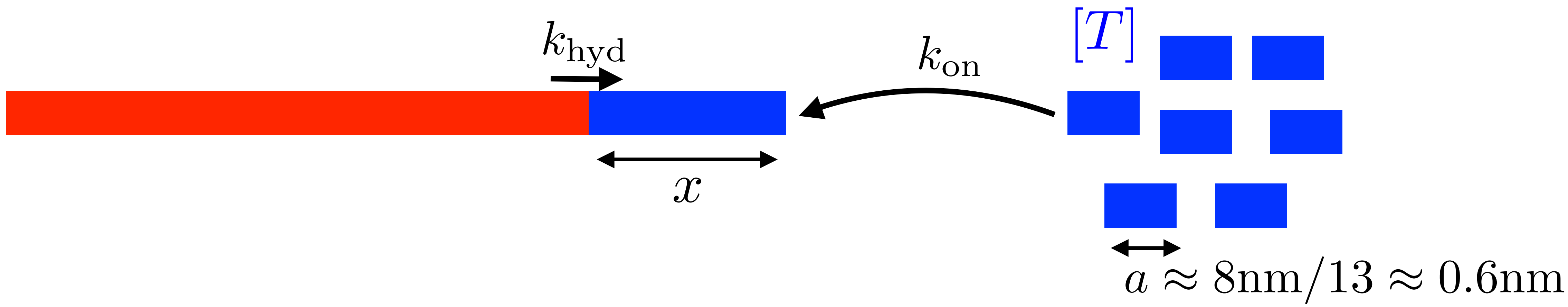
Effective one dimensional model



Master equation for the length *x* of protective cap

$$\frac{\partial p(x,t)}{\partial t} = k_{\mathrm{on}}[T]p(x-a,t) - k_{\mathrm{on}}[T]p(x,t) + k_{\mathrm{hyd}}p(x+a,t) - k_{\mathrm{hyd}}p(x,t)$$

Molecular model for growth of protective cap



Master equation for the length *x* of protective cap

$$\frac{\partial p(x,t)}{\partial t} = k_{\rm on}[T]p(x-a,t) - k_{\rm on}[T]p(x,t) + k_{\rm hyd}p(x+a,t) - k_{\rm hyd}p(x,t)$$

Continuum limit of the master equation

$$\boxed{\frac{\partial p(x,t)}{\partial t} = -v\frac{\partial p(x,t)}{\partial x} + D\frac{\partial^2 p(x,t)}{\partial x^2}}$$

growth speed of protective cap

$$v = ak_{\rm on}[T] - ak_{\rm hyd}$$

$$v = v_g - v_{\rm hyd}$$

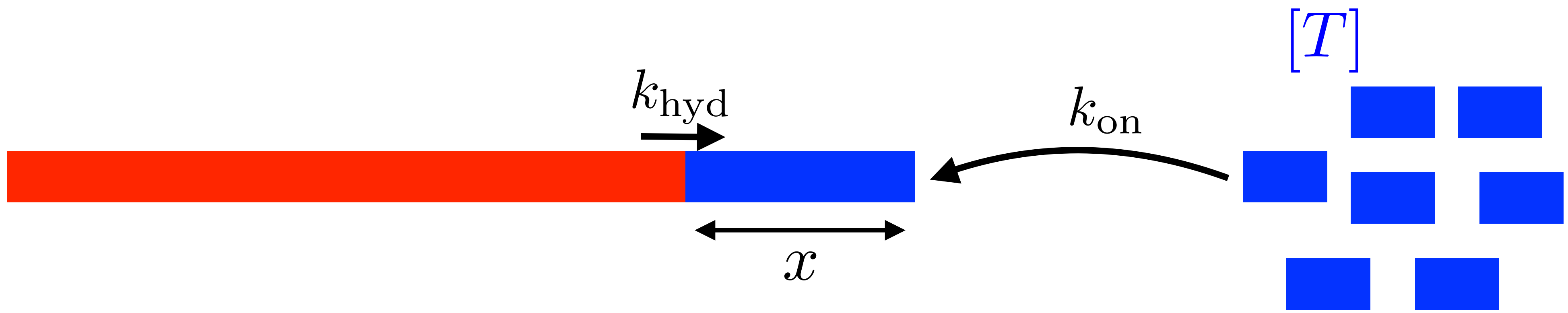
diffusion constant

$$D = \frac{a^2}{2}\left(k_{\rm on}[T] + k_{\rm hyd}\right)$$

Waiting time to the next catastrophe event

Gedanken experiment

Grow microtubules in a medium with large GTP-tubulin concentration [*T*] for time t_0



length of protective cap $x = vt_0 = a(k_{\mathrm{on}}[T] - k_{\mathrm{hyd}})t_0$

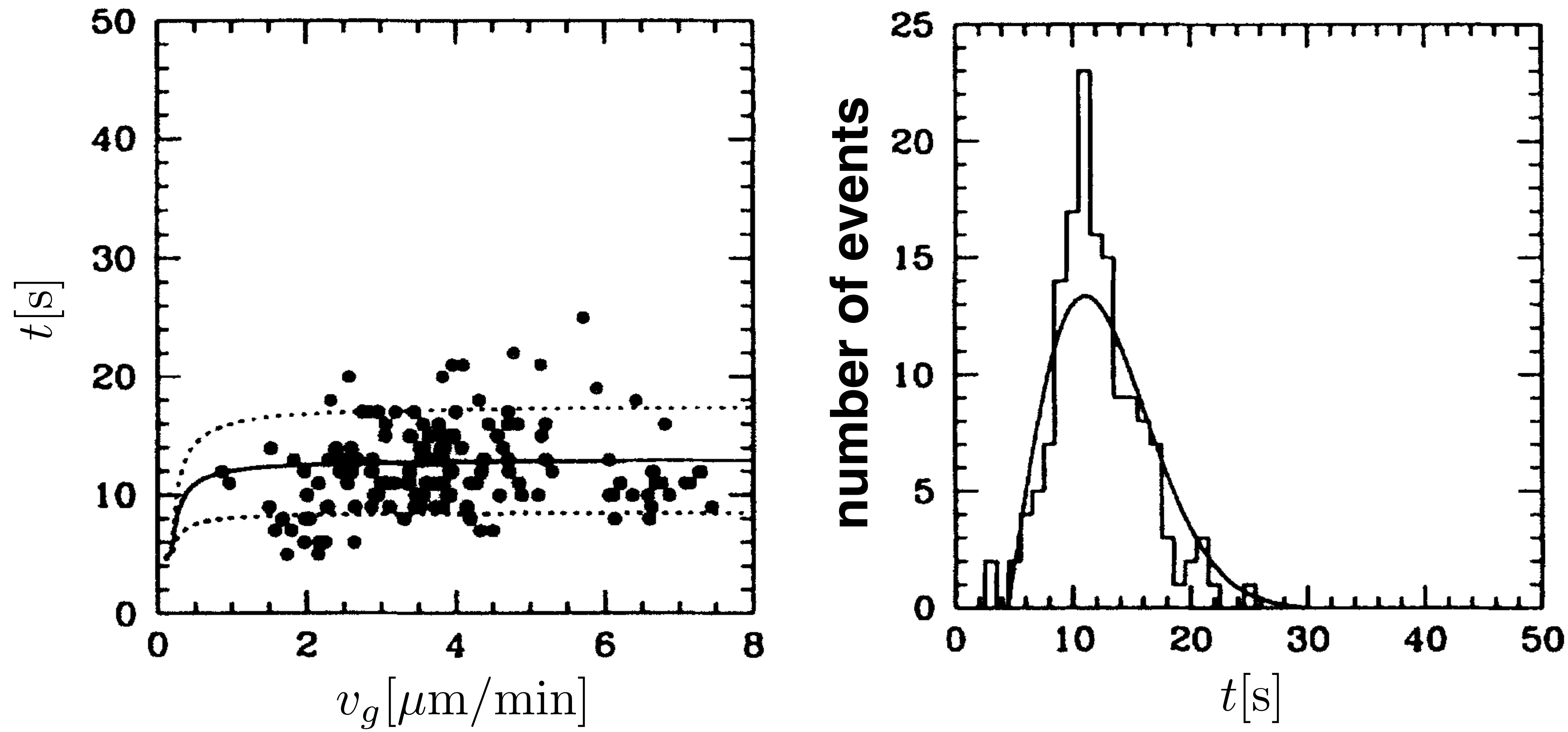
Then move microtubules to another medium without tubulin

waiting time to the catastrophe event

$$t_{\mathrm{cat}} = \frac{x}{v_{\mathrm{hyd}}} = \frac{(k_{\mathrm{on}}[T] - k_{\mathrm{hyd}})t_0}{k_{\mathrm{hyd}}}$$

Waiting time to the next catastrophe event

H. Flyvbjerg, T.E. Holy and S. Leibler,
PRL 73, 2372-2375 (1994)



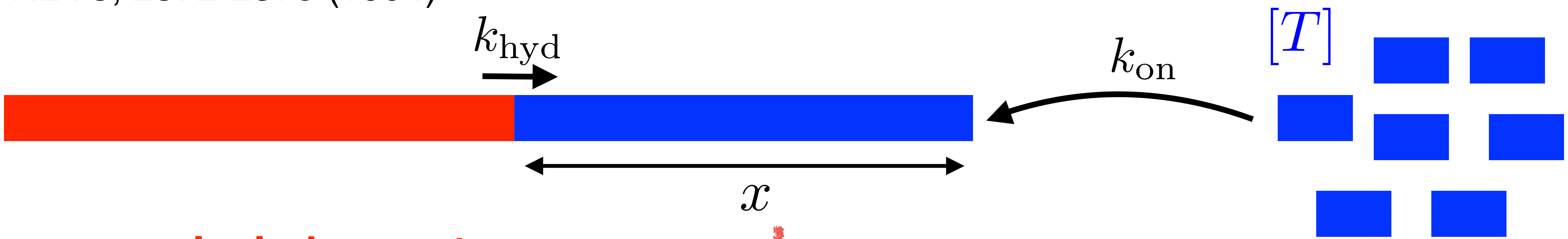
$$t_{\mathrm{cat}} = \frac{x}{v_{\mathrm{hyd}}} = \frac{(k_{\mathrm{on}}[T] - k_{\mathrm{hyd}})t_0}{k_{\mathrm{hyd}}}$$

Waiting time to the catastrophe event depends on GTP-tubulin concentration (~growth speed)!

This is in contrast with experimental observations that the waiting time to next catastrophe event is very insensitive to the tubulin concentration of the first medium!

Molecular model for growth of protective cap

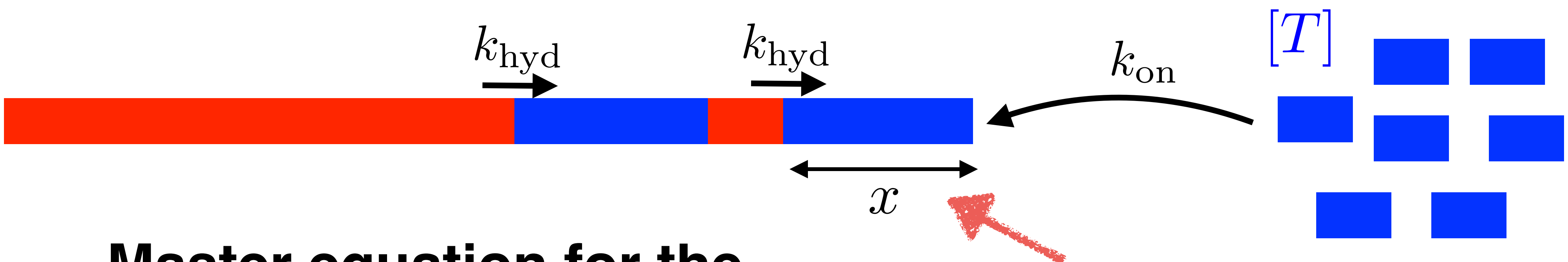
H. Flyvbjerg, T.E. Holy and S. Leibler,
PRL 73, 2372-2375 (1994)



Include spontaneous hydrolysis, that appears equally likely everywhere along the cap

$k_{\mathrm{hyd}}^{(s)}$

$a \approx 8\mathrm{nm}/13 \approx 0.6\mathrm{nm}$



only this parts serves as a protective cap that prevents catastrophe

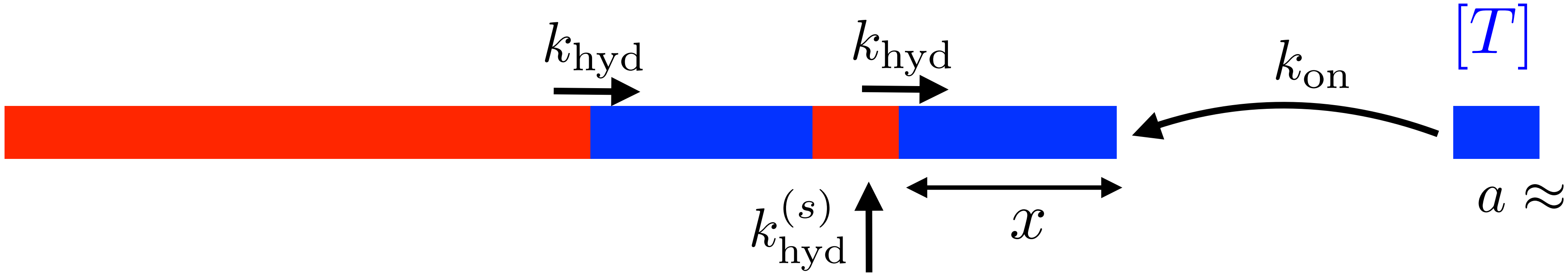
Master equation for the length *x* of protective cap

$$\frac{\partial p(x,t)}{\partial t} = k_{\mathrm{on}}[T]p(x-a,t) - k_{\mathrm{on}}[T]p(x,t) + k_{\mathrm{hyd}}p(x+a,t) - k_{\mathrm{hyd}}p(x,t)$$
$$-k_{\mathrm{hyd}}^{(s)} \sum_{y<x} p(x,t) \qquad +k_{\mathrm{hyd}}^{(s)} \sum_{y>x} p(y,t)$$

initial cap length *x*
spontaneous hydrolysis at *y* < *x*

initial cap length *y* > *x*
spontaneous hydrolysis at *x*

Molecular model for growth of protective cap



concentration of free GTP-tubulin

$a \approx 8\mathrm{nm}/13 \approx 0.6\mathrm{nm}$

Continuum limit of master equation for the length *x* of protective cap

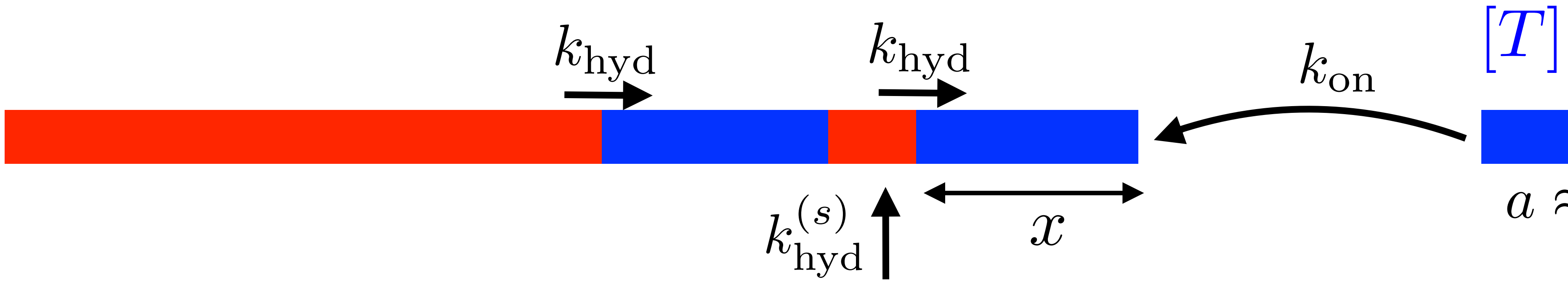
$$\frac{\partial p(x,t)}{\partial t} = -v\frac{\partial p(x,t)}{\partial x} + D\frac{\partial^2 p(x,t)}{\partial x^2} - rxp(x,t) + r\int_x^\infty dy\, p(y,t)$$

$$v = ak_{\mathrm{on}}[T] - ak_{\mathrm{hyd}} \qquad D = \frac{a^2}{2}\left(k_{\mathrm{on}}[T] + k_{\mathrm{hyd}}\right) \qquad r = \frac{k_{\mathrm{hyd}}^{(s)}}{a}$$

Rewrite equation above in terms of cumulative distribution $P(x,t) = \int_x^\infty dy\, p(y,t)$

$$\frac{\partial P(x,t)}{\partial t} = -v\frac{\partial P(x,t)}{\partial x} + D\frac{\partial^2 P(x,t)}{\partial x^2} - rxP(x,t)$$

Distribution of protective cap lengths



concentration of free GTP-tubulin

$a \approx 8\text{nm}/13 \approx 0.6\text{nm}$

$$\frac{\partial P(x,t)}{\partial t} = -v\frac{\partial P(x,t)}{\partial x} + D\frac{\partial^2 P(x,t)}{\partial x^2} - rxP(x,t)$$

cumulative distribution

$$P(x,t) = \int_x^\infty dy\, p(y,t)$$

What is the stationary distribution of protective cap lengths?

assume large GTP-tubulin concentration $v > 0$ **small diffusion** $D \approx 0$

$$\frac{\partial P^*(x,t)}{\partial t} = 0 \longrightarrow P^*(x) = e^{-rx^2/2v} \longrightarrow$$

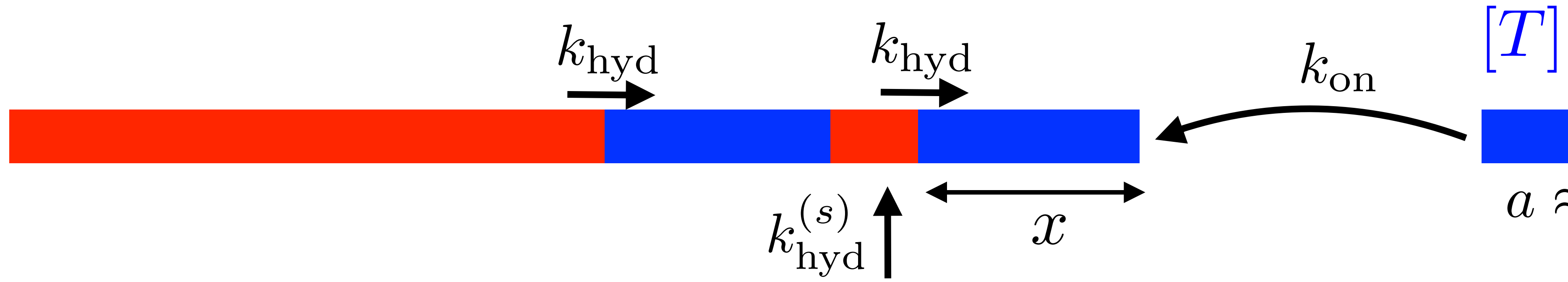
$$p^*(x) = -\frac{dP^*(x)}{dx} = \frac{rx}{v}e^{-rx^2/2v}$$

average cap length $$\overline{x} = \int_0^\infty dx\, xp^*(x) = \sqrt{\frac{\pi v}{2r}}$$

Because of spontaneous hydrolysis the typical length of protective caps remains finite!

(Note: microtubules still grow with time, because of hydrolysis of the tail)

Frequency of catastrophe events



concentration of free GTP-tubulin

$a \approx 8\text{nm}/13 \approx 0.6\text{nm}$

$$\frac{\partial P(x,t)}{\partial t} = -v\frac{\partial P(x,t)}{\partial x} + D\frac{\partial^2 P(x,t)}{\partial x^2} - rxP(x,t)$$

Catastrophe event happens when cap length shrinks to zero (*x*=0). In the absence of diffusion (*D*=0) catastrophes would never occur in GTP-tubulin rich medium. For small diffusion *D*, the probability distribution *P(x,t)* quickly approaches steady state distribution *P*(x)* multiplied by a prefactor *A(t)*, the probability that no catastrophe events occurred by time *t*.

$$P(x,t) \approx A(t)e^{-rx^2/2v} \rightarrow \frac{\partial P(x=0,t)}{\partial x} = \frac{dA(t)}{dt} = -\frac{Dr}{v}A(t) \rightarrow A(t) = e^{-Drt/v}$$

Probability distribution of waiting time to catastrophe

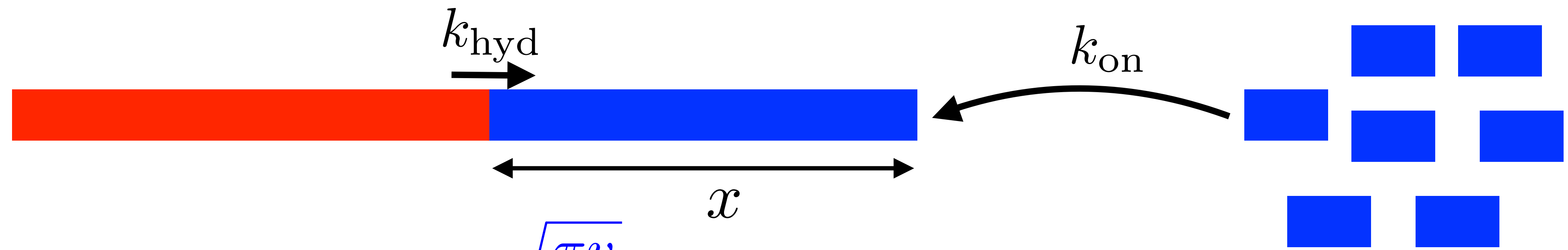
$$p_{\rm cat}(t) = -\frac{dA(t)}{dt} = \frac{Dr}{v}e^{-Drt/v}$$

$$\bar{t}_{\rm cat} = \frac{v}{Dr}$$

Waiting time to the next catastrophe event

Gedanken experiment

Grow microtubules in a medium with large GTP-tubulin concentration [*T*] for time t_0



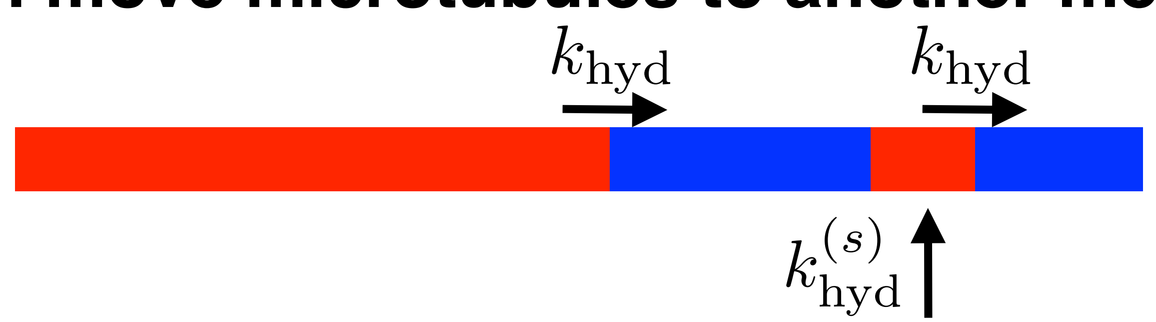
length of protective cap $\bar{x} = \sqrt{\dfrac{\pi v}{2r}} \sim 0.1\mu\mathrm{m}$

typical growth time before catastrophe $\bar{t}_{\mathrm{cat}} = \dfrac{v}{Dr} \sim 10\mathrm{min}$

$[T] \approx 10\mu\mathrm{M}$ $\quad r \approx 400\mu\mathrm{m}^{-1}\mathrm{min}^{-1}$

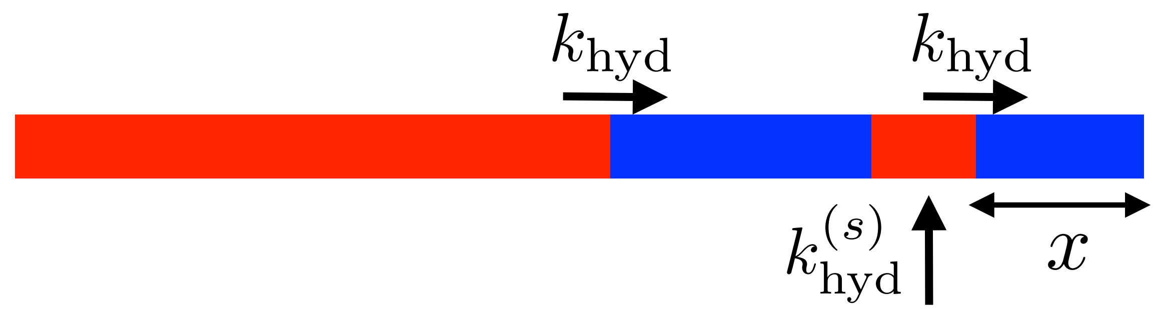
$v \approx 2\mu\mathrm{m/min}$ $\quad D \approx 10^{-3}\mu\mathrm{m}^2/\mathrm{min}$

Then move microtubules to another medium without tubulin

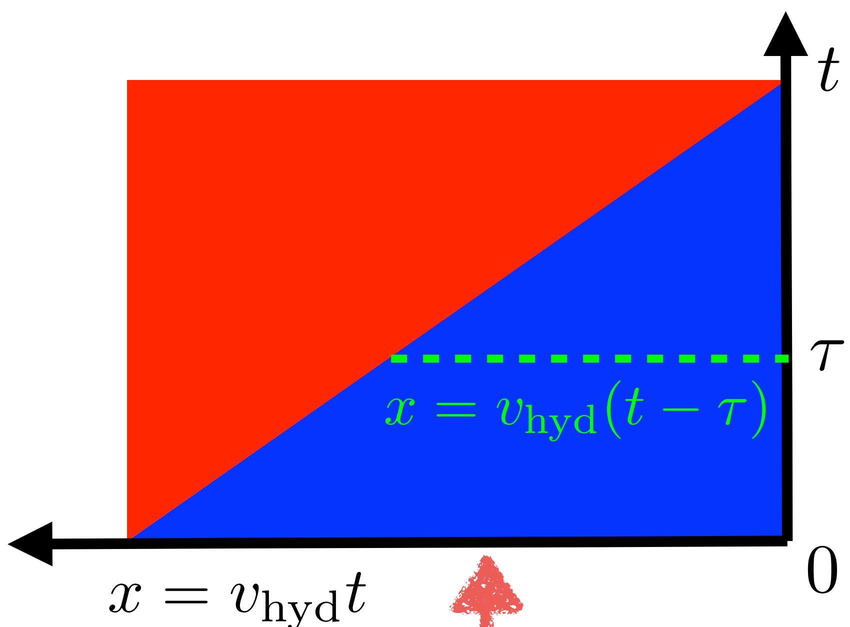


What is the probability for waiting times to the next catastrophe event in the presence of spontaneous hydrolysis?

Waiting time to the next catastrophe event



Next catastrophe event occurs at time *t* if spontaneous hydrolysis occurs at some time $0 < \tau < t$ at place $x = v_{\mathrm{hyd}}(t-\tau)$.



Any spontaneous hydrolysis inside the blue region would lead to catastrophe event at earlier time!

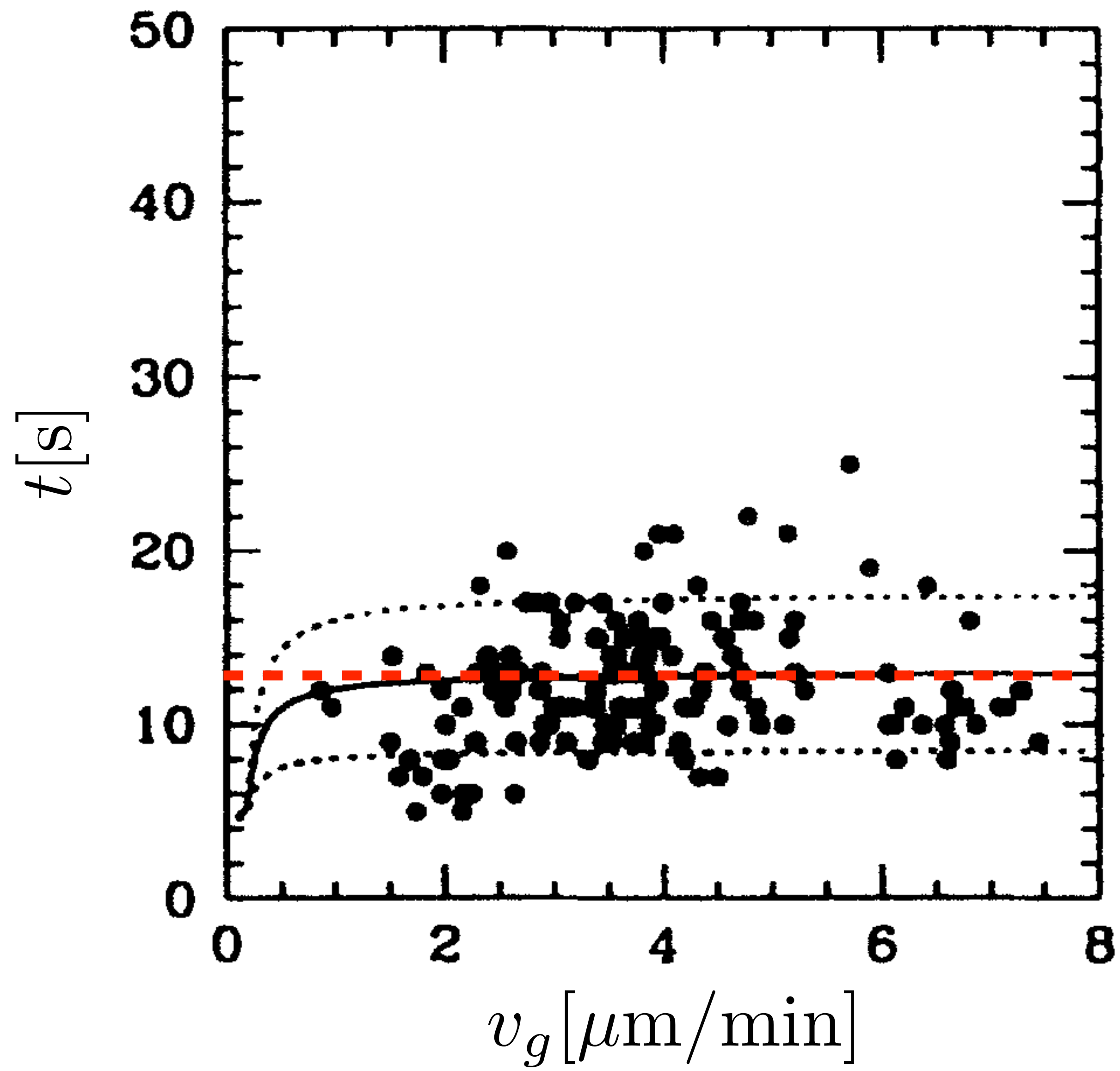
Probability distribution of waiting time to next catastrophe event

$$p_{\mathrm{cat}}(t) = r v_{\mathrm{hyd}} t \times e^{-r v_{\mathrm{hyd}} t^2/2}$$

probability that spontaneous hydrolysis happens somewhere at the interface between blue and red region

probability that no spontaneous hydrolysis happens inside blue region

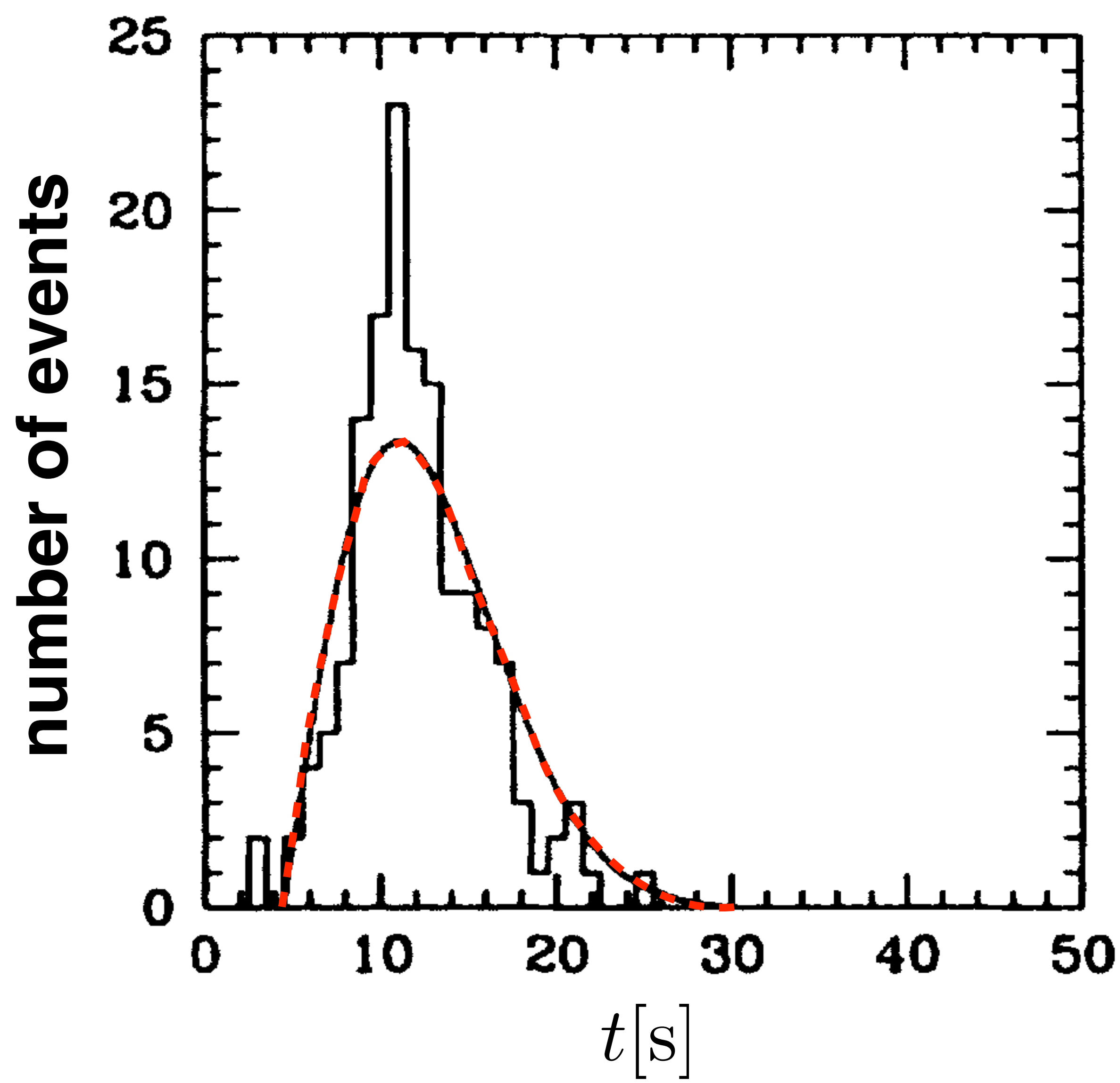
Waiting time to the next catastrophe event



$$\bar{t} = \sqrt{\frac{\pi}{2 r v_{\mathrm{hyd}}}} \sim 8s$$

Waiting time to the catastrophe event is insensitive to GTP-tubulin concentration (~growth speed)!

$r \approx 400 \mu\mathrm{m}^{-1}\mathrm{min}^{-1}$ $\quad v_{\mathrm{hyd}} \approx 0.2 \mu\mathrm{m}/\mathrm{min}$



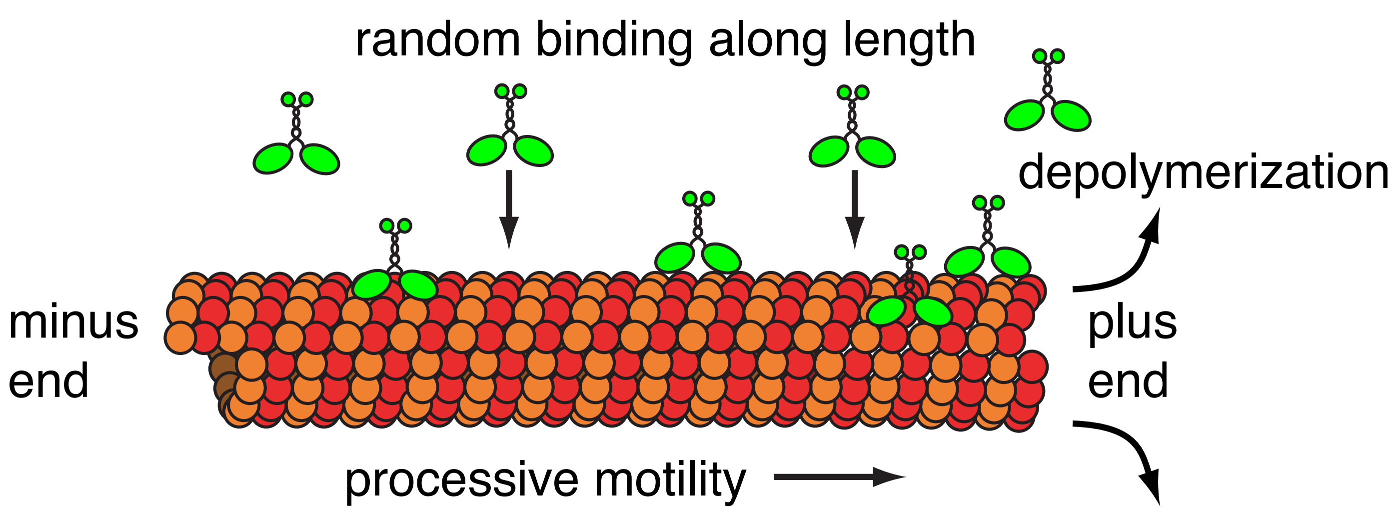
$$p_{\mathrm{cat}}(t) = r v_{\mathrm{hyd}} t \times e^{-r v_{\mathrm{hyd}} t^2/2}$$

Note: observed ~6s delay from the initiation of dilution

H. Flyvbjerg, T.E. Holy and S. Leibler, PRL 73, 2372-2375 (1994)

How cells control the total length of microtubules

Special kinesin-8 motors bind to microtubules and then walk towards the plus end, where they help detach (depolymerize) tubulin dimers

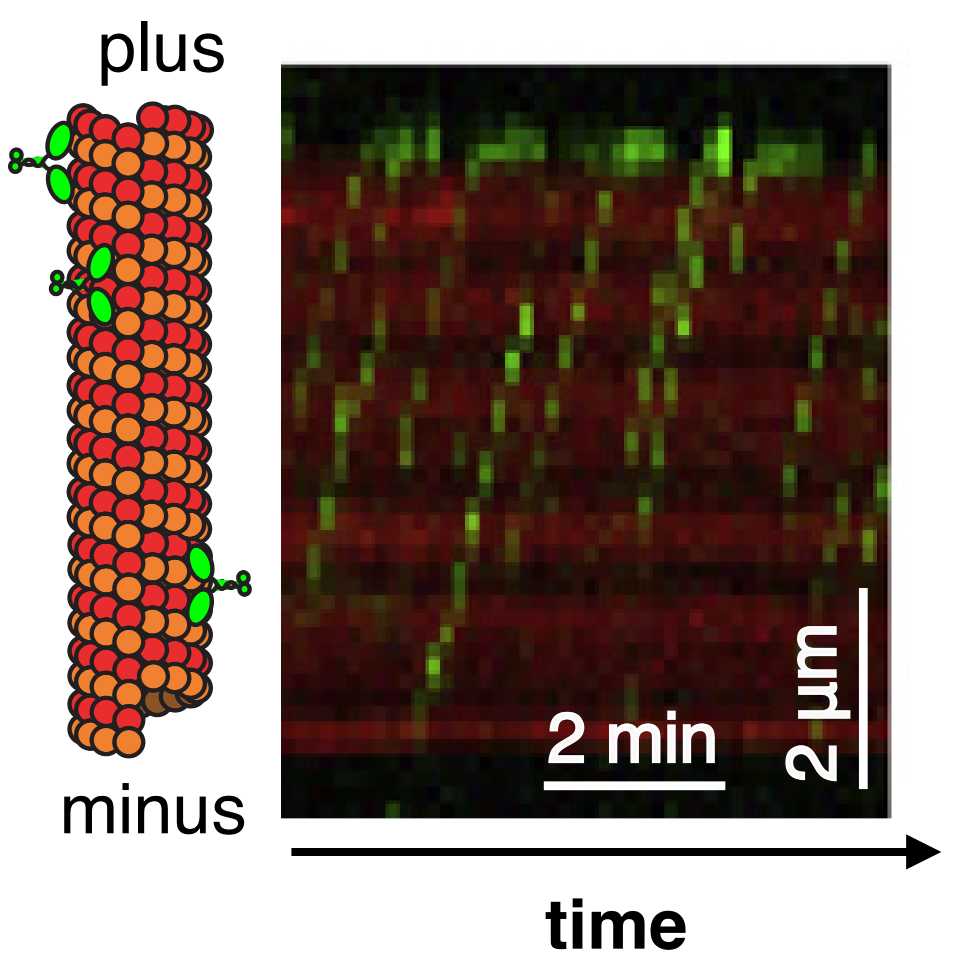


Motors walk at speed

$$v_{\rm mot} \approx 3\mu{\rm m/min}$$

kymograph

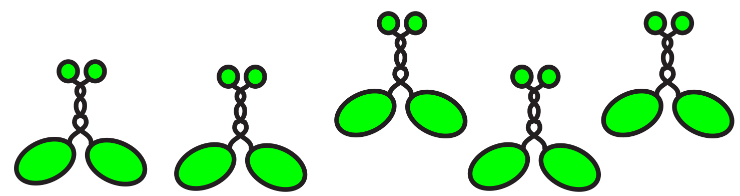
$$v_{\rm mot} \approx 3\mu{\rm m/min}$$

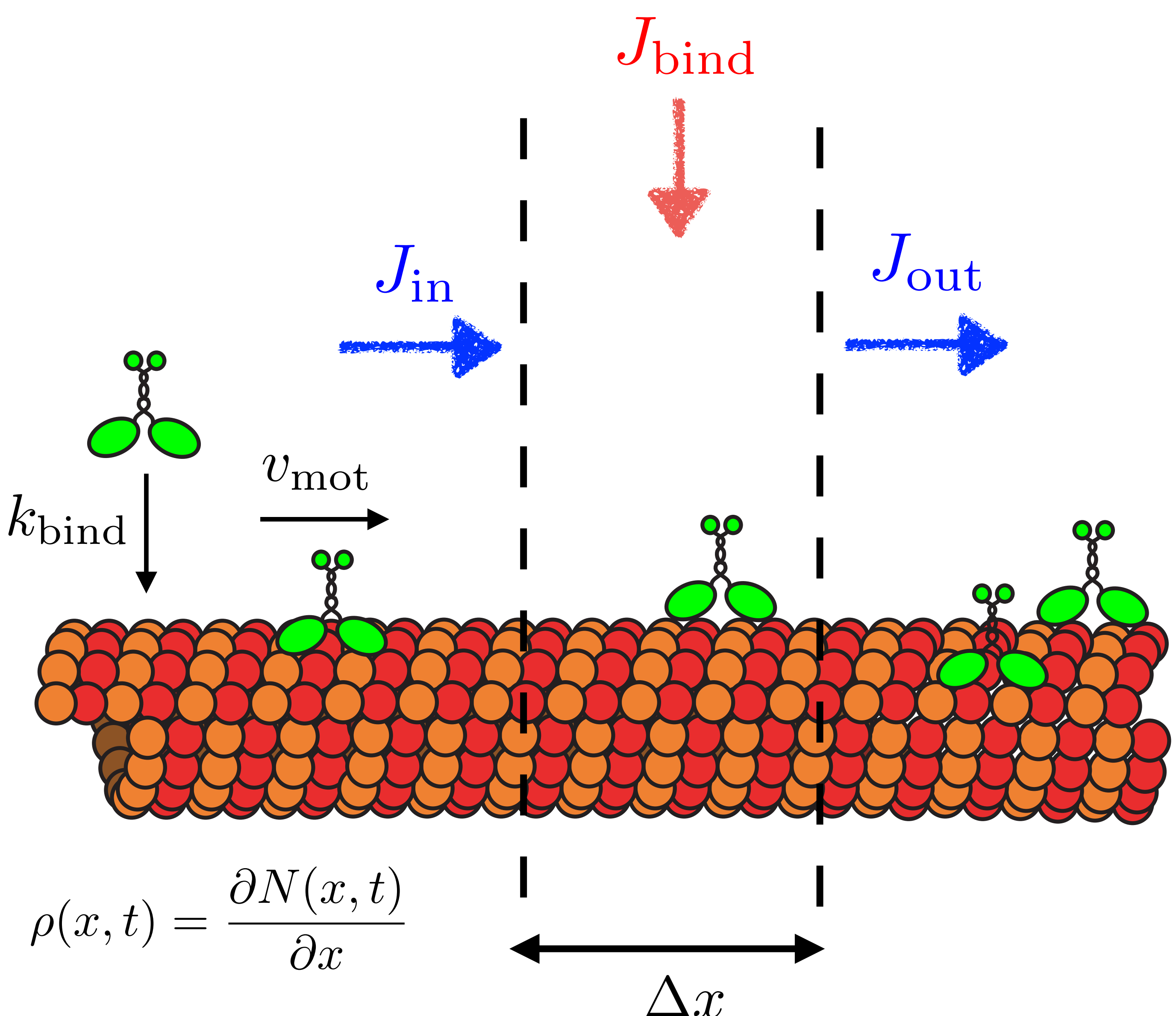


V. Varga *et al.*, Cell 138, 1174-1183 (2009)

Density of motors bound to microtubules

[*M*] concentration of unbound motors





$$\rho(x,t) = \frac{\partial N(x,t)}{\partial x}$$

density of bound motors

Conservation law for the number of bound motors

$$\frac{\Delta N}{\Delta t} = J_{\mathrm{bind}} - J_{\mathrm{out}} + J_{\mathrm{in}}$$

$$\frac{\Delta N(x,t)}{\Delta t} = k_{\mathrm{bind}}[M]\Delta x - \big(\rho(x+\Delta x,t) - \rho(x,t)\big)v_{\mathrm{mot}}$$

$$\frac{\partial \rho(x,t)}{\partial t} = k_{\mathrm{bind}}[M] - v_{\mathrm{mot}}\frac{\partial \rho(x,t)}{\partial x}$$

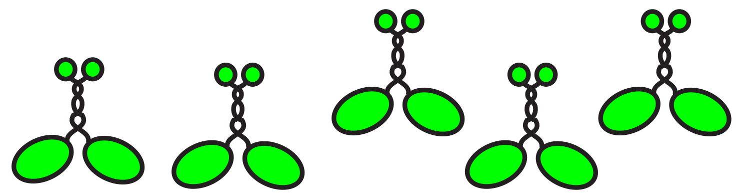
Generalized Fick's law

$$\frac{\partial \rho(x,t)}{\partial t} = r(x,t) - \frac{\partial j(x,t)}{\partial x}$$

creation/removal of particles

Density of motors bound to microtubules

[*M*] concentration of unbound motors

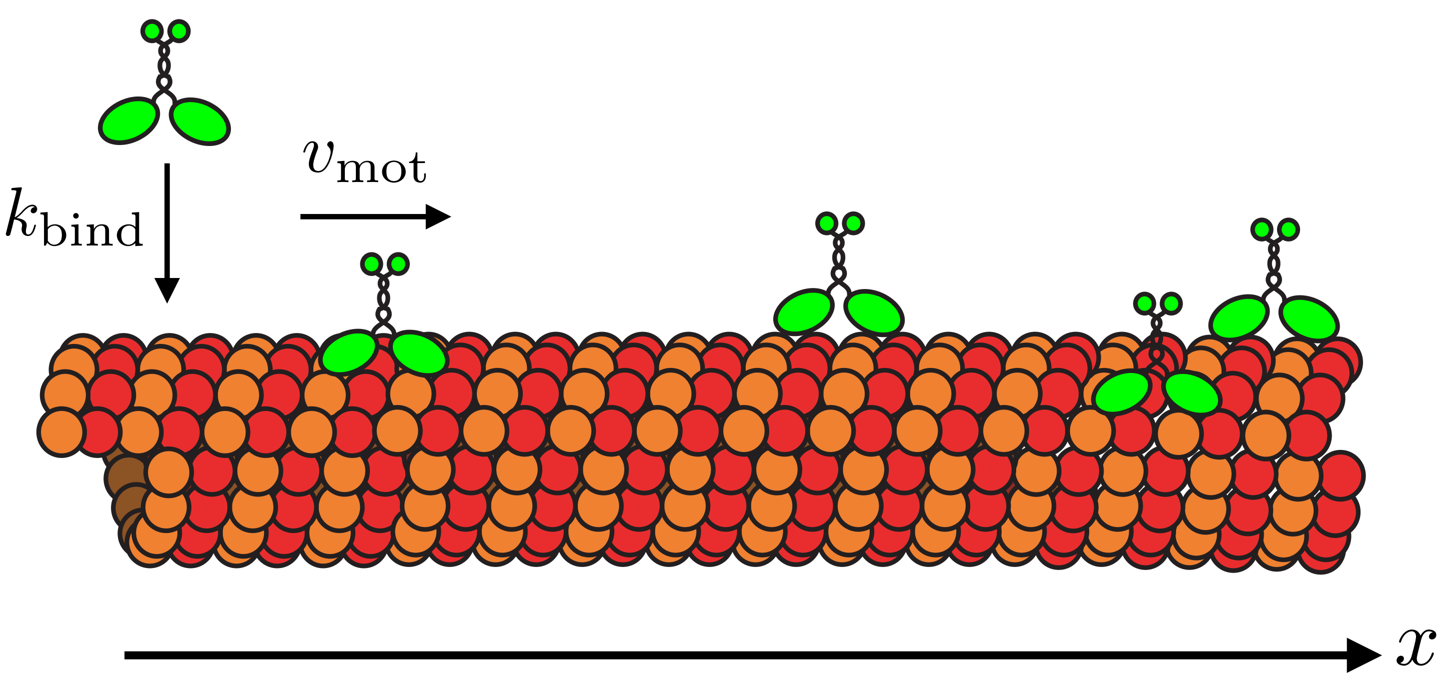


Time evolution for density of bound motors

$$\frac{\partial \rho(x,t)}{\partial t} = k_{\text{bind}}[M] - v_{\text{mot}} \frac{\partial \rho(x,t)}{\partial x}$$

For initially empty microtubule

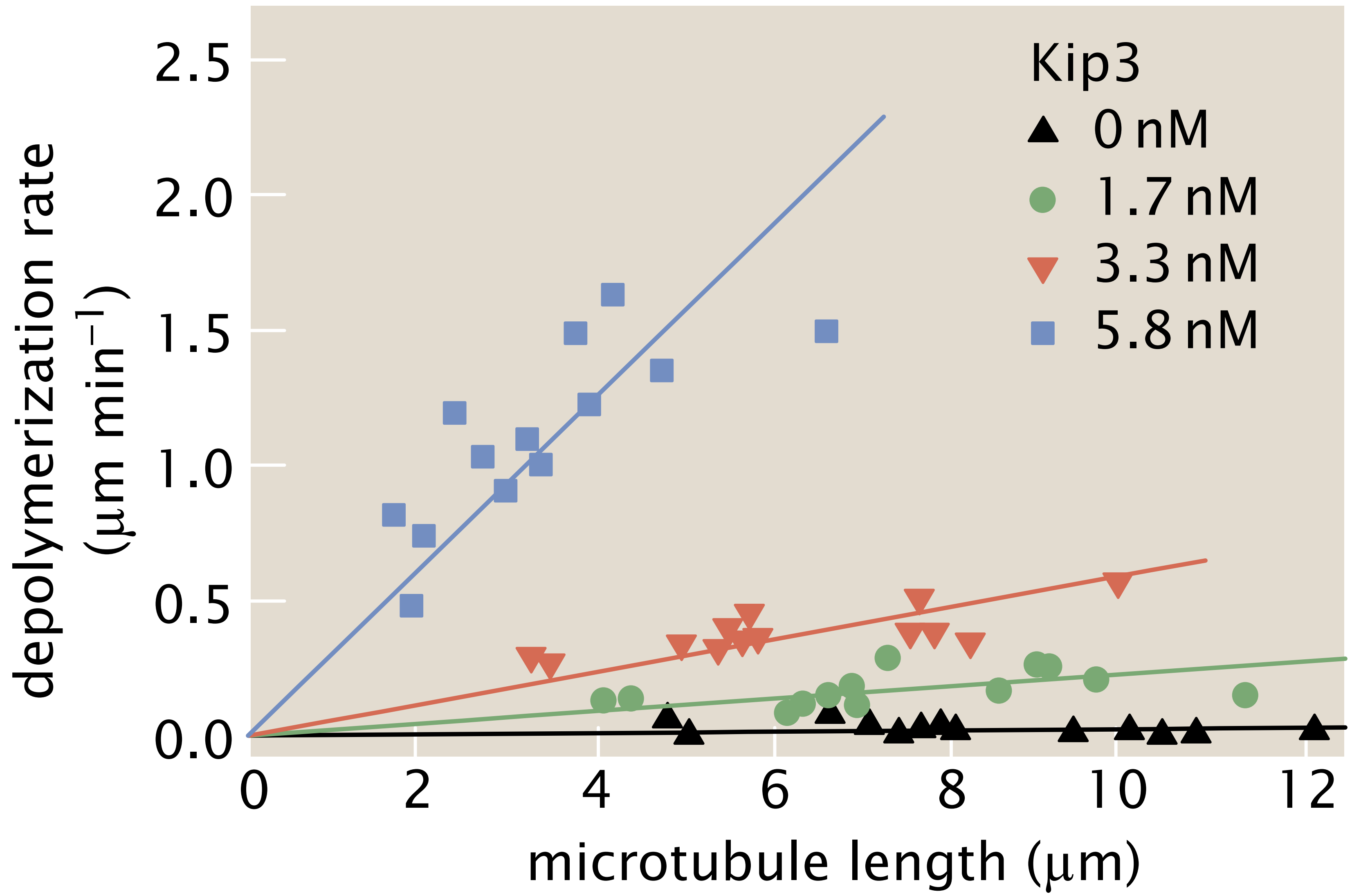
$$\rho(x,t) = \begin{cases} \frac{k_{\text{bind}}[M]}{v_{\text{mot}}}\, x, & 0 < x < v_{\text{mot}} t \\ k_{\text{bind}}[M] t, & x > v_{\text{mot}} t \end{cases}$$



Stationary density of bound motors

$$\rho^*(x) = \frac{k_{\text{bind}}[M]}{v_{\text{mot}}}\, x$$

Length dependent depolymerization rate

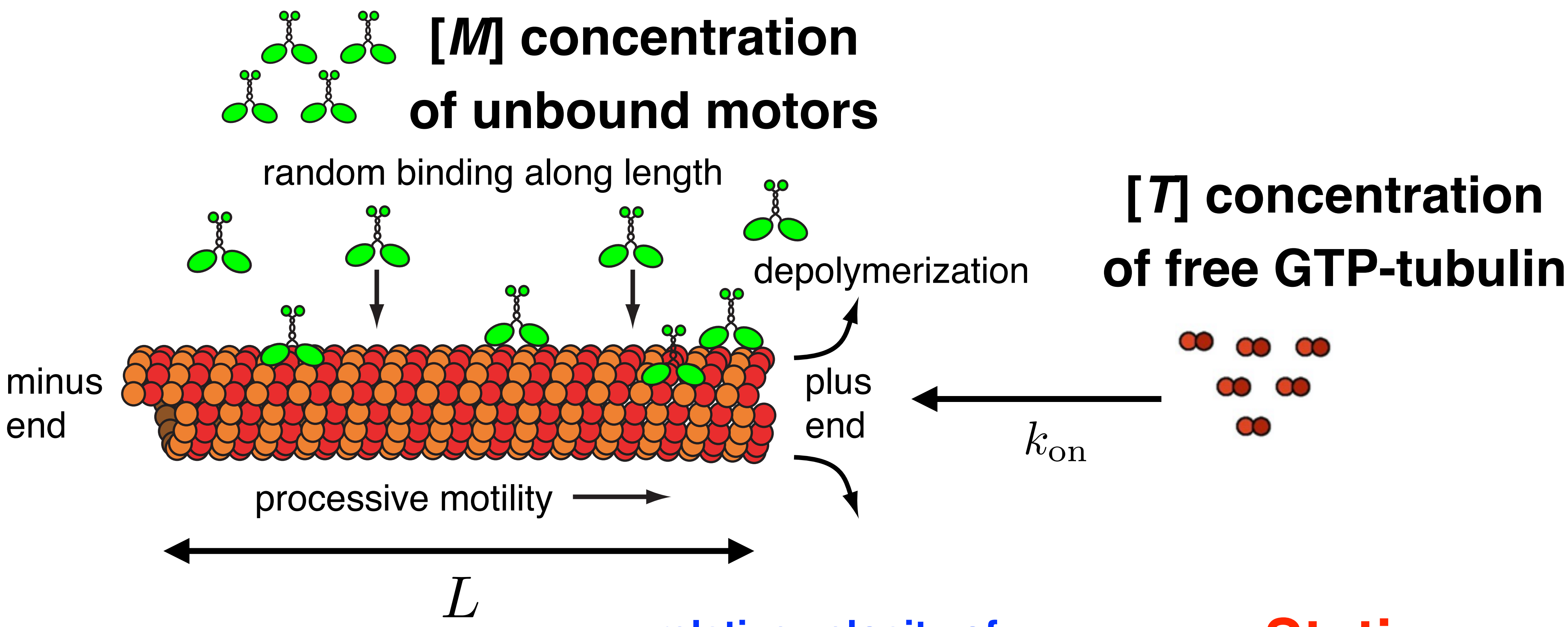


Depolymerization rate is proportional to density of Kip3 motors

$$\rho^*(L) = \frac{k_{\mathrm{bind}}[M]}{v_{\mathrm{mot}}} L$$

V. Varga *et al.*, Nat. Cell Biol. 8, 957-962 (2006)

Controlled length of microtubules



relative velocity of motors arriving to the tip

$$\frac{dL}{dt} = ak_{\mathrm{on}}[T] - a\rho^*(L)\left[v_{\mathrm{mot}} - \frac{dL}{dt}\right]$$

$$\frac{dL}{dt} = \frac{(ak_{\mathrm{on}}[T] - a\rho^*(L)v_{\mathrm{mot}})}{1 - a\rho^*(L)}$$

$$\rho^*(L) = \frac{k_{\mathrm{bind}}[M]}{v_{\mathrm{mot}}}\, L$$

Stationary length of microtubules

$$L^* = \frac{k_{\mathrm{on}}[T]}{k_{\mathrm{bind}}[M]}$$

$[T] \approx 10\mu\mathrm{M}$
$k_{\mathrm{on}} \approx 9\mu\mathrm{M}^{-1}\mathrm{s}^{-1}$
$[M] \approx 3\mathrm{nM}$
$k_{\mathrm{bind}} \approx 24\mathrm{nM}^{-1}\mathrm{min}^{-1}\mu\mathrm{m}^{-1}$

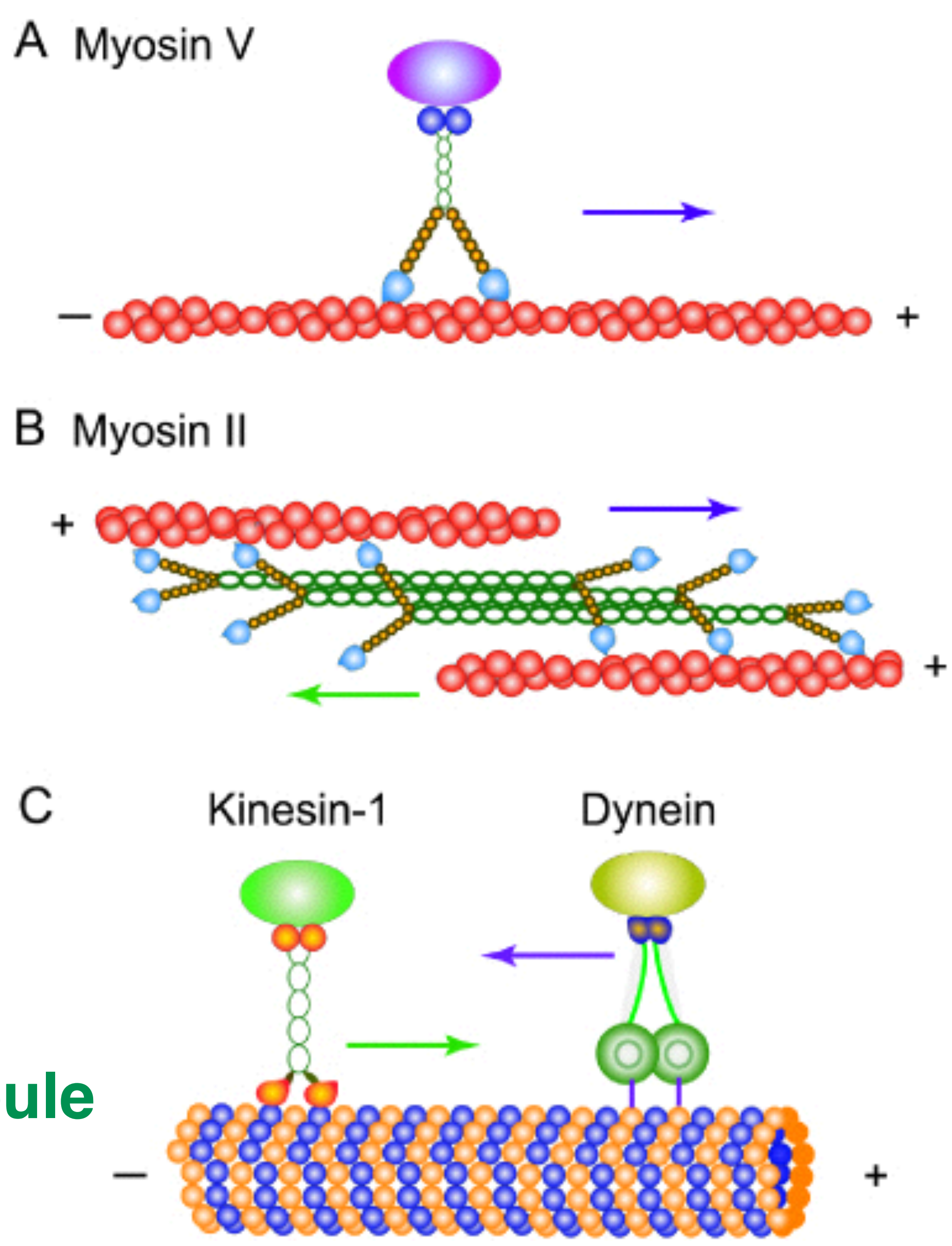
$L^* \sim 75\mu\mathrm{m}$

Molecular motors

Actin

Actin

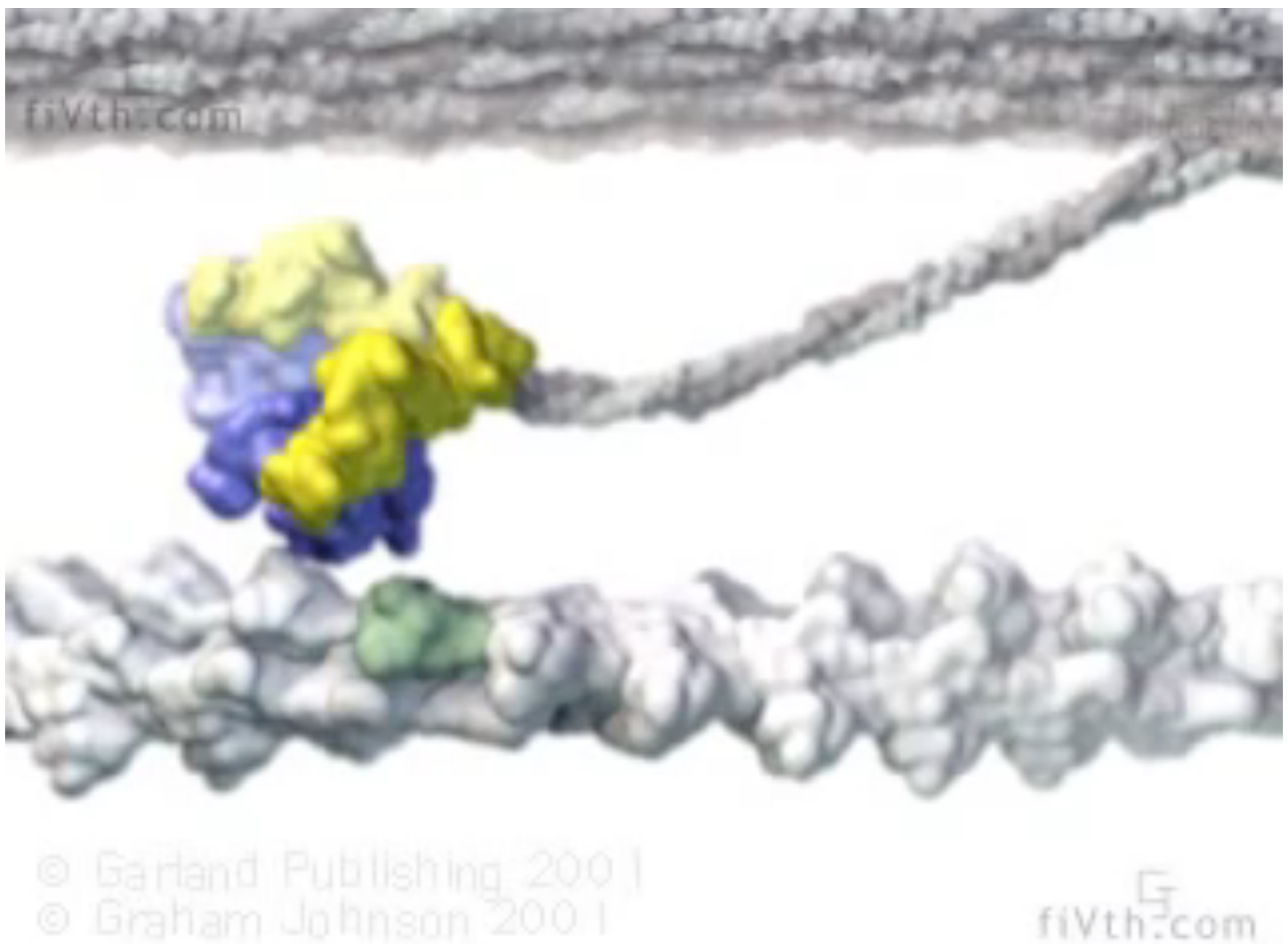
Microtubule



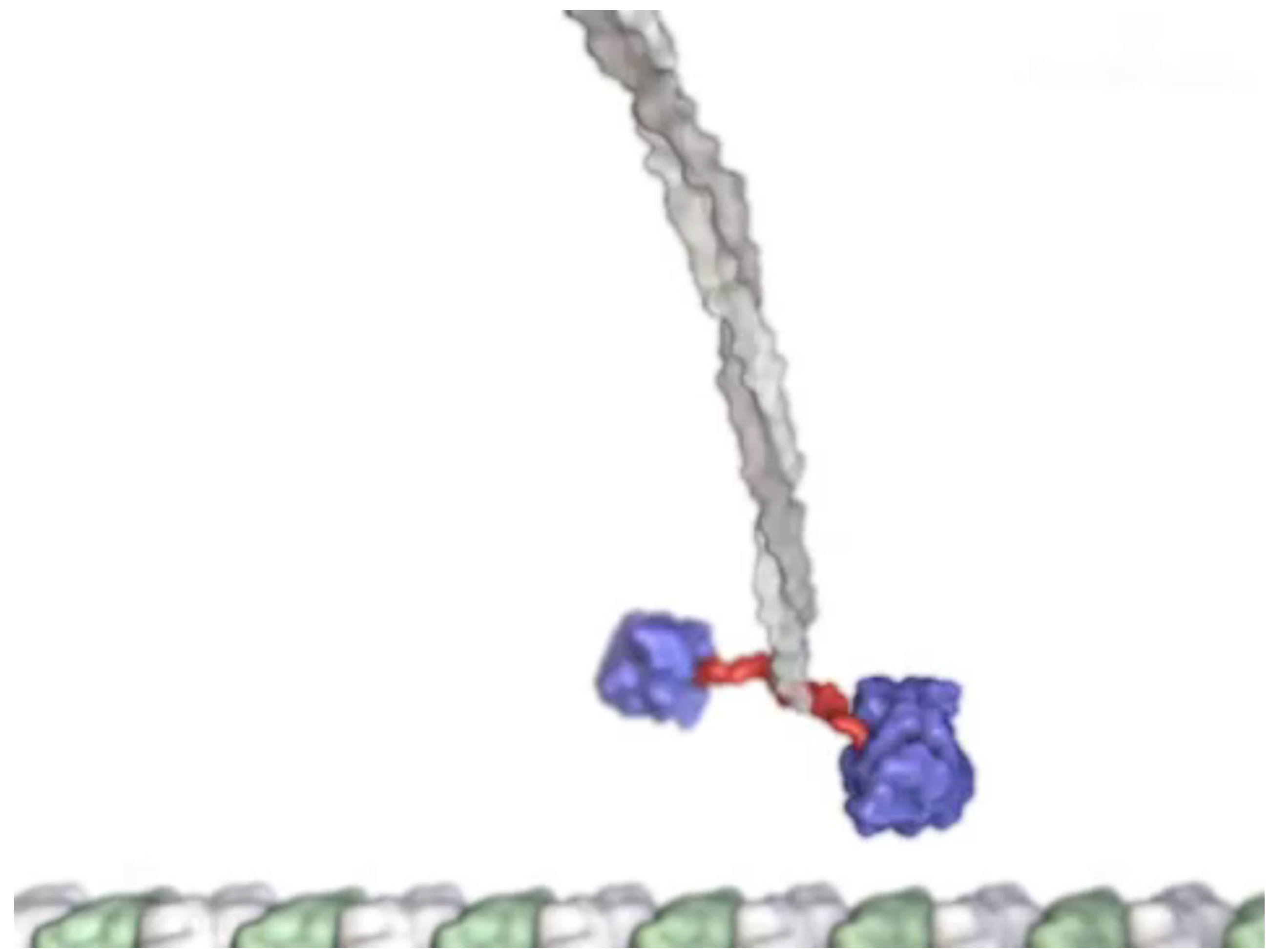
A.B. Kolomeisky, J. Phys.: Condens. Matter 25, 463101 (2013)

Movement of molecular motors is powered by ATP molecules

Myosin motor walking on actin in muscles

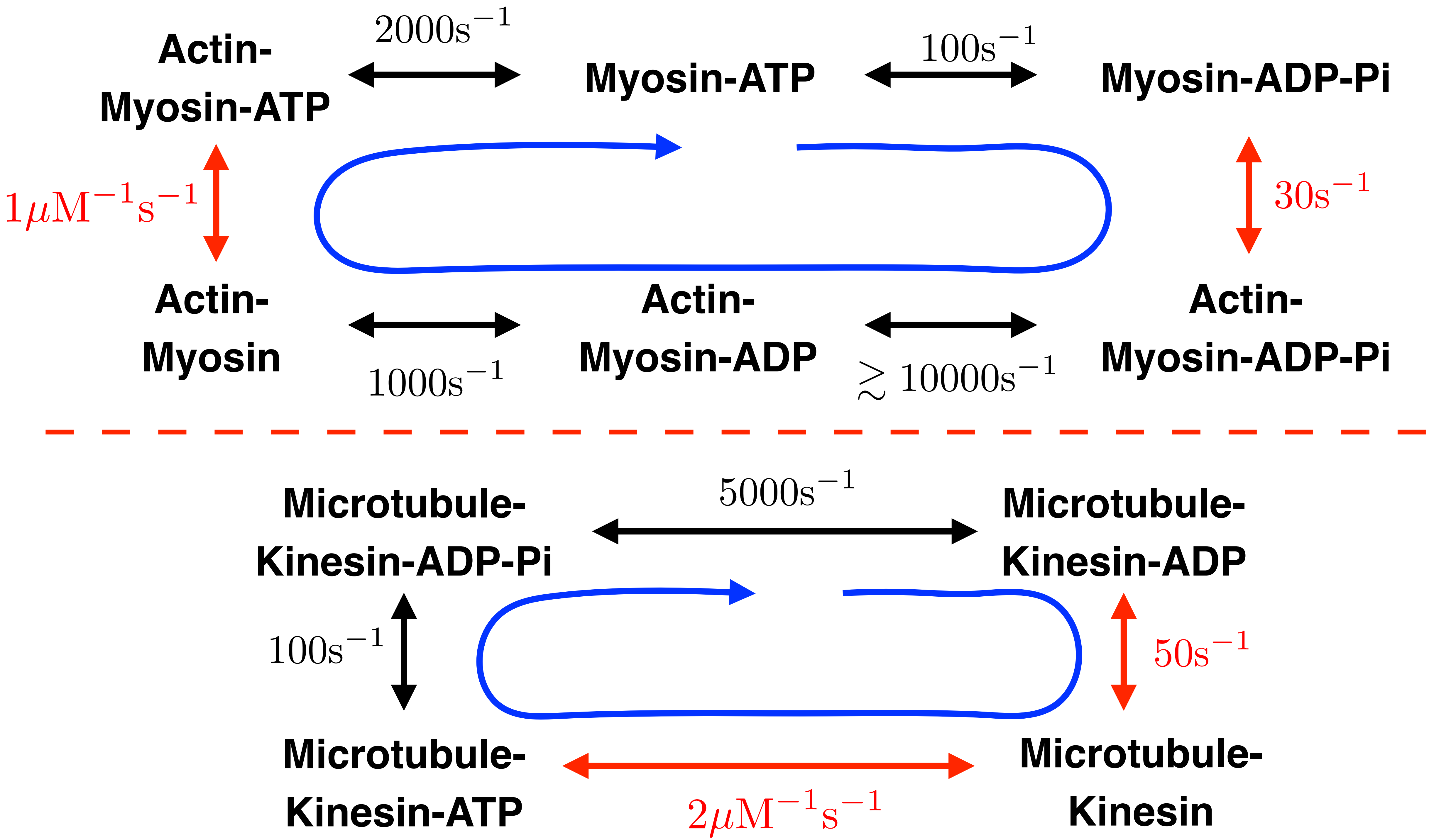


Kinesin motor walking on microtubule



Graham Johnson

Typical rates (timescales)



timescale=1/rate

stepping motion for kinesin: ~100 *μ*s

F. Fulga *et al.*, Integr. Biol. 1, 150-169 (2009)