Hitchhiking through the cytoplasm

I. M. KULIĆ and P. C. NELSON

Department of Physics and Astronomy, University of Pennsylvania Philadelphia - PA 19104, USA

Abstract – We propose an alternative mechanism for intracellular cargo transport which results from motor-induced longitudinal fluctuations of cytoskeletal microtubules (MTs). The longitudinal fluctuations combined with transient cargo binding to the MTs lead to long-range transport even for cargos and vesicles having no molecular motors on them. The proposed transport mechanism, which we call "hitchhiking", provides a possible explanation for the broadly observed yet still mysterious phenomenon of bidirectional transport along MTs. We show that cells exploiting the hitchhiking mechanism can effectively up- and down-regulate the transport of different vesicles by tuning their binding kinetics to characteristic MT oscillation frequencies.

Molecular-motor–mediated transport along microtubules (MTs) is a well-studied phenomenon *in vitro*. There is an increasing number of studies of classic microtubule motors like kinesin [1–3] and dynein [4,5] that shed light on their mechanochemistry in the idealized situation of *in vitro* single-molecule assays. Despite significant *in vitro* advances, understanding how intracellular transport works *in vivo* still remains one of the big challenges in molecular biology. Questions like how cellular cargo vesicles find their way through the cytoplasm and get targeted to their temporary or final destinations are at the heart of the problem. While the classical unidirectional transport is well understood within the "cargo hauled by a motor" model [1], the exact origin of the phenomenon called "bidirectional transport" (BDT) remains to be solved [6,7]: A large fraction of cargos in the cell move in a bidirectional and in many cases remarkably symmetric manner with similar velocity, run length, and stalling force distributions in both directions [6,7], often exhibiting only weak bias towards one microtubule end.

A first straightforward explanation for this back and forth rocking motion of cellular cargos is that opposite polarity motors kinesin and dynein both reside on the cargo and compete for the direction of motion in some way. This simple "tug of war" model came under debate after the discovery that mutations affecting either kinesin or dynein lead to a symmetric reduction of motion in both directions in several important model systems [6,7]. To explain this observation the "motor coordination hypothesis" was proposed [6] stating that a yet unknown molecular element could couple dynein and kinesin into a single functional unity. Such a coordinated molecular-motor complex would by some as-yet undetermined internal mechanism respond symmetrically to any perturbation (mutation) of its two constituent subunits kinesin and dynein. Molecules like dynactin and the klar protein known to affect BDT were suspected to be part of the complex [6] but the molecular mechanism of the coordination model remains unclear. While in some systems it was demonstrated that both motors reside on the cargos [8] a definite proof for a molecular regulator symmetrizing the behavior of two structurally markedly different motor species like dynein and kinesin is still missing to date.

Here we propose an alternative molecular model for BDT that accounts for the approximately symmetric short-time behavior in both directions. It also contains an intrinsic mechanism able to generate biased cargo distributions on longer timescales. The disruption of one motor type leads naturally to a transport reduction in both directions, as observed experimentally.

Our model has two basic ingredients: 1. *Motors of either or both types (not necessarily localized to the cargo) generate longitudinal MT fluctuations* by already known mechanisms, *e.g.* the one depicted in fig. 1a. This motor-induced confined MT sliding we call "jabberwalking" of the underlying motors. While inter-MT shearing forces are best known for motile organelles containing the axoneme structure [9], an increasing number of studies show vigorous sliding of cytoplasmic MTs when they
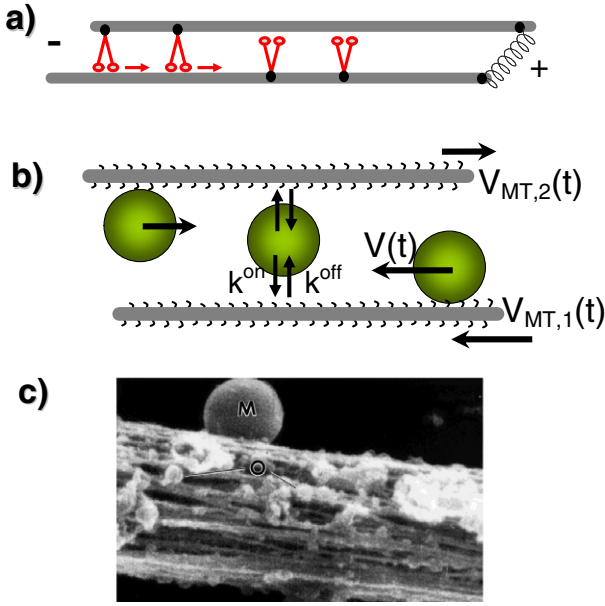
Fig. 1: a) Longitudinal MT noise caused by "jabberwalking" of double attached motors on elastically tethered MTs. b) The hitchhiking mechanism: Vesicle binding and unbinding kinetics, combined with local MT oscillations, induces diffusive, long-range transport. MT surface polarity can also give rise to a directed drift. c) *Reticulomyxa* transports microspheres (M) and organelles (O) via MT sliding (adapted from [12]).

become detached from cellular structures [10]. Two-point microrheology studies of cytoskeletal stress-strain fluctuations that demonstrate the presence of active force doublets [11] go along the same line of evidence. 2) *Vesicles transiently couple to the random, stochastically oscillating MTs* and attain their speed temporarily. As a consequence, the vesicles experience strongly enhanced diffusion. This process we call "hitchhiking" of the vesicles. The occurrence of such a mechanism is most clearly observed for reticulopodial cytoskeleton extensions [12] and chlamydomonas flagella [13]. In those systems, artificial and endogenous cargos (fig. 1c) are found to move as a consequence of motor-driven MT sliding in a bidirectional manner. The non-specific MT-cargo coupling there is likely to be established via microtubule associated proteins (MAPs) and transmembrane glycoproteins. More recently, distant vesicles moving in highly correlated fashion or in synchrony with MT tips presented strong evidence for hitchhiking in S2 drosophila cell lines [14].

We note that the hitchhiking scenario described here bears some analogy with Taylor dispersion [15] (enhanced diffusion in constant low Reynolds number tube flow generated by velocity gradients in the direction perpendicular to the diffusion axis) and with transport in disordered velocity fields [16]. However, as we will see later, hitchhiking has its own character in particular because of the stochastic MT driving mechanism and the breaking of time reversibility of the Stokes flow coming from the

cargo-MT interaction kinetics. The latter in particular can induce a net current of cargo along MTs, an effect absent in Taylor dispersion.

We first focus on the hitchhiking process and compute how a vesicle transiently coupling to a single longitudinally oscillating MT moves on long timescales (fig. 1b). Each time the cargo binds to the MT, it assumes the latter's direction and velocity $v = v_{MT}$; when it is unbound, we assume complete immobility *i.e.*, $v = 0$. We neglect here the thermal diffusion of the unbound vesicle due to its smallness compared to the active transport we consider here (cf. below). The velocity $v_{MT}(t)$ itself is a random process with the particular property that $x_{MT}(t) = \int_0^t v_{MT}(\tau) d\tau$ is a bounded variable, *i.e.*, we assume the MT to move in a spatially confined fashion. Consequently, a cargo simply permanently sticking to the MT does not get far, and so moves in a confined manner as well. However, by virtue of the switching (attachment/detachment) process, the cargo coordinate $x(t)$ can become unbounded.

The vesicle binding process is described by a 2-state random variable $B(t) \in \{0, 1\}$ with the characteristics of telegraphic noise [17]: $B(t)$ equals 1 if the cargo sticks to the MT or 0 for a detached resting cargo. We denote the stochastic switching rates between those states by k^{off} and k^{on}. We next write the velocity of the cargo vesicle as a composite random process $v(t) = B(t)v_{MT}(t)$. The cargo velocity correlation function is given by $\langle v(t_1)v(t_2) \rangle = \langle B(t_1)B(t_2)v_{MT}(t_1)v_{MT}(t_2) \rangle$, where $\langle \ldots \rangle$ denotes the ensemble average over all realizations of B and v. For simplicity, at first we focus on the limiting case of velocity-independent rates k^{off} and k^{on}. In this case, the vesicle binding becomes statistically independent of the MT motion $\langle Bv_{MT} \rangle = \langle B \rangle \langle v_{MT} \rangle$, $B(t)$ becomes a standard (asymmetric) Markovian telegraphic noise process [17], and we easily compute $\langle v(t_1)v(t_2) \rangle = C_{MT}(t_1, t_2)C_B \times (t_1 - t_2)$ with $C_{MT}(t_1, t_2) = \langle v_{MT}(t_1)v_{MT}(t_2) \rangle$ and $C_B \times (t_1 - t_2) = K^2 + K(1 - K)e^{-2|t_1 - t_2|/t_{sw}}$. The two constants K and t_{sw} characterize the binding behavior. $K = \langle B \rangle = k^{on}/(k^{on} + k^{off})$ denotes the equilibrium binding constant and $t_{sw} = 2/(k^{on} + k^{off})$ is the "mean switching time". Note that whereas the switching process $B(t)$ is assumed to be stationary in the statistical sense, the velocity $v_{MT}(t)$ can in general be a non-stationary stochastic or even a purely deterministic process, and its autocorrelation function $C_{MT}(t_1, t_2)$ is not necessarily time homogeneous. The mean-square displacement (msd) of such a vesicle is obtained from

$$\left\langle (x(t) - x(0))^2 \right\rangle = \int_0^t \int_0^t C_{MT}(t_1, t_2) C_B(t_1 - t_2) dt_1 dt_2.$$
(1)

This general expression relates the msd of the cargo to the fluctuations of a single microtubule and the binding/unbinding kinetics of the cargo to the MT. For the special case of a statistically stationary process $v_1(t)$, the correlator $C_{MT}(t_1, t_2) = C_{MT}(t_1 - t_2)$ becomes

homogeneous in time and we can further simplify eq. (1):

$$\left\langle (x(t) - x(0))^2 \right\rangle = 2 \int_0^t (t - \tau)\, C_B(\tau)\, C_{\mathrm{MT}}(\tau)\, \mathrm{d}\tau. \quad (2)$$

Now we illustrate our general formulas by focusing on two particular possible MT shaking processes: 1) Deterministic oscillation of the MT with $v_{\mathrm{MT}}(t) = V \sin \omega t$, and 2) MT motion as an overdamped harmonic oscillator driven by Markovian telegraphic noise (representing the action of motors, fig. 1a).

Periodically oscillating MT. – In the first case, we have $C_{\mathrm{MT}}(t_1, t_2) = V^2 \sin \omega t_1 \sin \omega t_2$, and it is easy to evaluate the rhs of eq. (1). The result can be simplified in two limiting cases. On short timescales $t \ll t_{\mathrm{sw}}$, the vesicle does not have enough time to bind/unbind from the MT, and we simply have $C_B(t) \approx K$. From eq. (1) we obtain $\langle (x(t) - x(0))^2 \rangle = K \omega^{-2} V^2 (1 - \cos t\omega)^2$, as expected for a particle strictly following the MT with probability K. In particular, the vesicle stays spatially confined. In the opposite limit $t \gg t_{\mathrm{sw}}$ the vesicle motion becomes diffusive: $\langle (x(t) - x(0))^2 \rangle = \mathrm{const} + 2Dt$, with the diffusion constant given by

$$D = K(1 - K)V^2 t_{\mathrm{sw}}(4 + \omega^2 t_{\mathrm{sw}}^2)^{-1}. \quad (3)$$

This expression says that a vesicle stochastically coupling to a periodically oscillating MT diffuses with an efficiency that depends on the fine tuning of the MT oscillation frequency ω and the stochastic vesicle switching time t_{sw}. For $t \gg t_{\mathrm{sw}}$ the transport efficiency is maximized for $K^{\mathrm{opt}} = 1/2$ and $t_{\mathrm{sw}}^{\mathrm{opt}} = 2\omega^{-1}$, and falls off to zero away from these values. The intuitive meaning of the first result ($K^{\mathrm{opt}} = 1/2$) is clear: If the vesicle sticks too strongly to the MT ($K = 1$) or not at all ($K = 0$), there is no long-range transport as it either moves with the MT (in a confined manner) or not at all. The optimum occurs for an intermediate value. The second finding $t_{\mathrm{sw}}^{\mathrm{opt}} = 2\omega^{-1}$, which resembles stochastic resonance phenomena [18], also has a simple interpretation: If the vesicle takes too long a ride on the MT ($t_{\mathrm{sw}} \gg \omega^{-1}$), its average displacement cancels because of the pure "back and forth" motion of the MT. If the ride is too short ($t_{\mathrm{sw}} \ll \omega^{-1}$), a similar argument applies. In this rather trivial example, we already see an interesting theme: Optimal transport of vesicles requires a fine tuning of vesicle binding and MT oscillations.

MT driven by stochastic motor noise. – In a second more realistic approach, we model longitudinal MT oscillations by an overdamped Langevin equation with a harmonic restoring force coming from MT attachment/confinement (figs. 1a,b): $\xi \dot{x}_{\mathrm{MT}} = -C x_{\mathrm{MT}} + F_{\mathrm{mot}}(\dot{x}_{\mathrm{MT}}, t)$. Here ξ is the MT longitudinal friction constant and C the MT restoring spring constant. The actively generated motor force $F_{\mathrm{mot}}(\dot{x}_{\mathrm{MT}}, t)$ depends on the detailed motor mechanochemistry, which we effectively model by a linear force-velocity relation

$v_{\mathrm{mot}}(F_{\mathrm{mot}})/v_0 = 1 - F_{\mathrm{mot}}/F_0$ with two parameters: v_0, the maximal (zero-load) velocity, and F_0, the motor stalling force. Combining this with the Langevin equation and $\dot{x}_{\mathrm{MT}} = v_{\mathrm{mot}}$ yields the effective equation of MT motion:

$$\xi_{\mathrm{eff}}(t)\dot{x}_{\mathrm{MT}} = -C x_{\mathrm{MT}} + F_0(t), \quad (4)$$

$\xi_{\mathrm{eff}}(t) = \xi + F_0(t)/v_0(t)$ is the effective friction constant.

Equation (4) states that the motors contribute to an effective external force $F_0(t)$, but also give rise to increased effective friction $\xi_{\mathrm{eff}} > \xi$. The driving motor force $F_0(t)$ and velocity $v_0(t)$ are stochastic variables, that can switch between two values. The dynamics of this switching generally depends on the MT-motor attachment geometry. In the simplest arrangement (fig. 1a), motors bind rigidly and run actively on both MTs in a symmetric manner. In this case $F_0(t)$ and $v_0(t)$ both switch between two values, which for simplicity we assume to be equal in magnitude but of opposite sign, i.e., $F_0(t) = \pm F_0$ and $v_0(t) = \pm v_0$ (same number of motors of same strength on both sides), which results in a time-independent friction constant $\xi_{\mathrm{eff}} = \xi + F_0/v_0$. We assume that the motors stochastically switch direction with an exponentially distributed switching time, i.e., $F_0(t)$ is described by symmetric Markovian telegraph noise with $p(F_0(t) = \pm F_0) = 1/2$ and $\langle F_0(t_1)F_0(t_2) \rangle = F_0^2 \exp(-2|t_1 - t_2|/T_p)$. Here T_p is the processivity time of the motors (average time between direction changes).

In the long-time limit, where x_{MT} in eq. (4) becomes a stationary process, we can exploit eq. (2) to evaluate the msd of the vesicle, provided that we can compute $C_{\mathrm{MT}}(\tau) = \langle \dot{x}_{\mathrm{MT}}(t + \tau)\dot{x}_{\mathrm{MT}}(t) \rangle$. To accomplish this, we use the solution of eq. (4) in the limit $\xi_{\mathrm{eff}}^{-1}Ct \gg 1$: $x(t) = \int_0^t \xi_{\mathrm{eff}}^{-1} F_0(\tau) e^{-\xi_{\mathrm{eff}}^{-1}C(t - \tau)}\mathrm{d}\tau$. After some calculation, this leads to [19]:

$$C_{\mathrm{MT}}(t) = \frac{F_0^2}{\xi_{\mathrm{eff}}^2} \frac{2 T_r T_p}{4 T_r^2 - T_p^2} \left(\frac{2 T_r}{T_p} e^{-\frac{2|t|}{T_p}} - e^{-\frac{|t|}{T_r}} \right). \quad (5)$$

The two characteristic timescales are now the MT relaxation time $T_r = \xi_{\mathrm{eff}}/C$ and the motor processivity time T_p. Inserting eq. (5) into eq. (2) gives a lengthy expression, which, in the limit $t \gg \max(T_p, T_r, t_{\mathrm{sw}})$, leads to

$$D = \frac{F_0^2}{C^2} \frac{2K(1 - K) t_{\mathrm{sw}} T_p}{(2 T_r + t_{\mathrm{sw}})(t_{\mathrm{sw}} + T_p)(T_p + 2 T_r)}. \quad (6)$$

As in the previous example, the vesicle mobility is always maximized for $K^{\mathrm{opt}} = 1/2$, i.e., for an intermediate binding strength. The optimal vesicle switching time $t_{\mathrm{sw}}^{\mathrm{opt}} = \sqrt{2 T_r T_p}$ is also easily obtained from eq. (6). Interestingly, for fixed T_r, eq. (6) predicts the optimal ratio $T_p/T_r = 2$. For those values we obtain the maximal diffusion constant $D_{\mathrm{max}} = F_0^2(\xi + F_0/v_0)^{-1} C^{-1}/32$, i.e., stronger and faster motors (F_0 and v_0 large), weaker MT confinement (C small) and smaller MT friction give rise to a more efficient transport.

Multiple MTs. – For n different oscillating MTs, the particle velocity is $v(t) = \sum_{k=1}^{n} \delta_{k,B(t)} v_{\mathrm{MT}_k}(t)$ with v_{MT_k} the velocity of the k-th MT, $\delta_{k,l}$ the Kroneker-delta and $B(t) \in \{0, 1, 2, \ldots, n\}$ the "binding variable" which indicates to which MT the vesicle is bound at time t (0 represents the unbound state). We assume the motor forces between the MTs to be large enough to disrupt multiple binding of the cargo to several MTs (generically moving at different speeds) at a time. For the two-MT situation in fig. 1a, $n = 2$ and $v_{\mathrm{MT}_1} = -v_{\mathrm{MT}_2}$ (anticorrelated MT velocities). Using eq. (5) as before yields the resonance condition: $k^{\mathrm{on}} \gg k^{\mathrm{off}}$, $k^{\mathrm{off}} = \sqrt{2}(T_r T_p)^{-1/2}$ and $T_p = 2T_r$, which differs slightly from the single-MT case because it is now more favorable to jump between the two MTs than to spend time in the unbound state. The corresponding diffusion constant $D_{\max} = F_0^2(\xi + F_0/v_0)^{-1} C^{-1}/8$ is 4 times larger than in the single-MT case. This indicates that the efficiency of transport, as well as the natural strategy for optimizing it, can depend on the effective number of participating MTs.

Hitchhiking vs. thermal diffusion. – In the limit of motor forces larger than viscous forces ($F_0 \gg \xi v_0$), we obtain the rough estimate $D_{\max} \approx \frac{1}{8}(F_0/C)v_0$. Typical MT sliding velocities $v_0 \approx 1$–$5\,\mu\mathrm{m/s}$ and oscillation amplitude $F_0/C \approx 0.1$–$1\,\mu\mathrm{m}$ yield $D_{\max} \approx 0.01$–$0.5\,\mu\mathrm{m}^2\,\mathrm{s}^{-1}$. In comparison, a typical organelle with diameter $500\,\mathrm{nm}$ experiences a large cellular viscosity $\eta \approx 0.3\,\mathrm{Pa \cdot s}$ [1] and has a thermal diffusion constant $D_{\mathrm{th}} \approx 3 \cdot 10^{-3}\,\mu\mathrm{m}^2\,\mathrm{s}^{-1}$. On the other hand, nanometer-sized molecules experience a much smaller effective viscosity (close to that of water $\eta \approx 10^{-3}\,\mathrm{Pa \cdot s}$) and so have $D_{\mathrm{th}} \approx 10$–$100\,\mu\mathrm{m}^2\,\mathrm{s}^{-1}$. Therefore large objects like vesicles and organelles can strongly benefit from hitchhiking while smaller molecules are more efficiently transported by thermal diffusion.

Hydrodynamic stress and biased hitchhiking. – It is straightforward to show that the hitchhiking process as described above for large t becomes an unbiased diffusive process even for an asymmetric MT shaking [14]. We consider next an interesting generalization of the hitchhiking model which gives rise to biased directed transport.

In the following, we drop the previous assumption $\langle B v_{\mathrm{MT}} \rangle = \langle B \rangle \langle v_{\mathrm{MT}} \rangle$, i.e. the statistical independence of the MT velocity and the vesicle-MT binding process. For example, a statistical coupling of binding and MT sliding can appear when the off rate k^{off} becomes velocity dependent due to a hydrodynamic drag force acting on the moving vesicle. The presence of this force can break the symmetry of transport in two different ways. The general form of the off rate will be $k^{\mathrm{off}}(v_{\mathrm{MT}})/k^{\mathrm{off}}(0) \approx 1 + c_2 v_{\mathrm{MT}}^2 + c_3 v_{\mathrm{MT}}^3 + \ldots$ with small $c_2 v_{\mathrm{MT}}^2$ and $c_3 v_{\mathrm{MT}}^3$. The linear term must vanish from the plausible requirement that k^{off} is minimal for $v_{\mathrm{MT}} = 0$. We call the coefficient c_2 the *dynamical bias coefficient*, because it gives rise to a particle drift only if the MT has different forward and backward velocity. We call c_3 the *polarity bias*, as it can give rise to drift even for a time reversal symmetric $v_{\mathrm{MT}}(t)$. Physically, this term stems from the polarity of the MT surface and the resulting polarity of the interaction with the vesicle.

We next derive the mean drift velocity $\langle v(t) \rangle$ of the vesicle in the simplifying limiting case of rapid binding equilibration, i.e. $k^{\mathrm{on}} + k^{\mathrm{off}} \gg \omega_{\mathrm{MT,char}}$, where $\omega_{\mathrm{MT,char}}$ denotes the characteristic oscillation frequency of the MT. In this limit, the vesicle binding state equilibrates at each instant of time and the conditional binding probability becomes a function of the instantaneous MT velocity: $p(B = 1|v_{\mathrm{MT}}) = k^{\mathrm{on}}/(k^{\mathrm{off}}(v_{\mathrm{MT}}) + k^{\mathrm{on}}) \approx K_0 - K_0(1 - K_0) \times (c_2 v_{\mathrm{MT}}^2 + c_3 v_{\mathrm{MT}}^3)$ with $K_0 = k^{\mathrm{on}}/(k^{\mathrm{off}}(0) + k^{\mathrm{on}})$. Consequently, $\langle v(t) \rangle = \langle v_{\mathrm{MT}}(t) p(B = 1|v_{\mathrm{MT}}(t)) \rangle = \langle v_{\mathrm{MT}}(t) \rangle K_0 - K_0(1 - K_0)(c_2 \langle v_{\mathrm{MT}}^3(t) \rangle + c_3 \langle v_{\mathrm{MT}}^4(t) \rangle)$ and its long-time mean (vesicle drift) reads

$$\overline{v} = -K_0(1 - K_0)(c_2 \overline{\langle v_{\mathrm{MT}}^3 \rangle} + c_3 \overline{\langle v_{\mathrm{MT}}^4 \rangle}), \qquad (7)$$

where the long-time average $\overline{f} \equiv \lim_{T \to \infty} T^{-1} \int_0^T f(t) \mathrm{d}t$. $\overline{\langle v_{\mathrm{MT}} \rangle}$ vanishes because of MT confinement. As an example, we consider an asymmetric square wave MT oscillation with $v_{\mathrm{MT}}(t)$ periodically switching between $v_{\mathrm{MT}} = V_1$ for a time T_1 and $v_{\mathrm{MT}} = V_2$ for a time T_2 with $V_1 V_2 < 0$ and $T_1 V_1 + T_2 V_2 = 0$ (zero mean). From eq. (7) we obtain $\overline{v} = -K_0(1 - K_0)(T_1 + T_2)^{-1}(c_2(T_1 V_1^3 + T_2 V_2^3) + c_3(T_1 V_1^4 + T_2 V_2^4))$ which in general has both the dynamical ($c_2 \ldots$) and the polar ($c_3 \ldots$) drift contributions. For a completely symmetric shaking ($V_2 = -V_1$, $T_1 = T_2$), the dynamical part vanishes as expected, whereas the polar part always stays present with a sign opposite to c_3.

Viscoelastic MT attachment. – Assuming an effective ideal harmonic spring between the MTs, as considered above, is merely an analytically tractable simplification for the restoring force stabilizing the longitudinal MT alignment. More realistically, MTs are held together by a gel-like network composed of semiflexible cytoskeletal polymers (predominantly intermediate filaments) that give rise to a complex time-dependent viscoelastic response to a motor-induced shear force. How does such a viscoelastic spring modify the cargo motion? When displacement is induced between neighboring MTs with a velocity $v_{\mathrm{MT}}(t)$ the semiflexible filament network responds with a time-dependent restoring force $F(t) = \frac{A}{D_{\mathrm{MT}}} \int_{-\infty}^t G(t - \tau) v_{\mathrm{MT}}(\tau) \mathrm{d}\tau$. Here D_{MT} is the typical distance between MTs, $A \sim R_{\mathrm{MT}} L_{\mathrm{MT}}$ the effective MT cross-sectional area over which the shear stress is acting and $G(t)$ is the time-dependent shear modulus of the interconnecting filaments. For semiflexible filaments the shear modulus obeys a power law $G(t) \propto t^{-\alpha}$ with $\alpha = 3/4$ [20] for times $t_{\mathrm{visc}} \ll t \ll t_{\mathrm{plat}}$. Here $t_{\mathrm{visc}} \propto \rho_{\mathrm{fil}}^{-4}$ is a filament density ρ_{fil} dependent timescale below which viscous fluid friction starts to dominate over filament friction. $t_{\mathrm{plat}} \sim \eta(l_p k_B T)^{-1} l_c^4$ is the timescale

at which the filament network reaches its final elastic plateau value $G_\infty \sim \rho_{\mathrm{fil}} k_B T l_p^2 / l_e^3$, with l_c the longitudinal filament crosslinking/entanglement distance and l_p their persistence length[1]. For $t \gg t_{\mathrm{plat}}$ the purely elastic spring approximation considered previously becomes valid. If, however, the cargo sticks firmly to the MT for long times $t_{\mathrm{sw}} \gg t_{\mathrm{plat}}$ but the motors switch directions before the crosslinking filaments reach their elastic plateau $t_{\mathrm{plat}} \gg T_p$ we expect: 1) Hyperdiffusive motion with $\langle (x(t) - x(0))^2 \rangle \propto t^\beta$ and $\beta = 3/2$ for $t \ll T_p$, 2) confined stochastically oscillatory motion for $T_p \ll t \ll t_{\mathrm{sw}}$, 3) diffusive motion $\langle (x(t) - x(0))^2 \rangle \propto t$ for $t \gg t_{\mathrm{sw}}$. Interestingly, behavior 1 and 3 and the cross-over between them on typical timescales of a few seconds [14,22,23] is commonly observed for many cargos in living cells consistently with the theoretical expectation of a long plateau time $t_{\mathrm{plat}} \approx 2\text{--}20\,\mathrm{s}$ for $\eta = 10^{-2}\text{--}10^{-1}\,\mathrm{Pa \cdot s}$, and a network of intermediate filaments $l_c \approx l_p \approx 1\,\mu\mathrm{m}$. The occurrence of the $\beta = 3/2$ scaling exponent *in vivo* was previously interpreted as cargo experiencing a time-dependent viscosity with respect to a stationary (viscoelastic) cytoplasm [22] or simply as a cross-over behavior between ballistic ($\beta = 2$) and diffusive ($\beta = 1$) cargo motion [23]. However, a careful analysis of the interparticle correlation behavior (as a function of their distance) by Lau *et al.* [11] revealed that a fluctuating background medium (the cytoskeleton) itself is predominantly responsible for the observed hyperdiffusive organelle motion in mammalian cells. More recently the observation of co-moving highly correlated peroxisome pairs in Drosophila S2 cells [14] showing the characteristic 3/2 exponent could be clearly attributed to moving microtubules in agreement with the predictions here.

Experimental aspects. – How could the hitchhiking hypothesis be tested experimentally? *In vivo*, one would need different markers on the cargo and on the microtubules in order to resolve unambiguously the frame of reference problem of moving *vs.* stationary MTs. A distinct signature of hitchhiking to be looked for would be the occurrence of several cargos moving in concert and cargos moving with the microtubules. Such observations from two color fluorescence imaging of microtubules and peroxisomes in drosophila S2 cells will be reported elsewhere [14]. An interesting *in vitro* experimental setup suitable for observing hitchhiking would be to use an immobilized demembranated axoneme in combination with adhering polystyrene beads as done by Kamimura and Kamiya [24]. While the beads used in [24] were firmly sticking to the oscillating MTs one could envision that an appropriate bead coating that would reduce their interaction strength with the MTs should also induce hitchhiking transport along the axoneme axis. By playing with the ATP/ADP concentration in the reactivation solution and with the type of axoneme it was possible to generate very rapid and highly asymmetric oscillations with amplitudes of $\sim 100\,\mathrm{nm}$ and speeds up to $50\,\mu\mathrm{m/s}$ [25]. As predicted above asymmetric MT sliding speeds such as in [25] should give rise to biased hitchhiking and a net current of beads towards one of the ends of the axoneme.

In conclusion, we have outlined a possible transport mechanism alternative to the standard "cargo hauled by a motor" model. Active longitudinal motions of MTs, combined with even weak non-specific cargo-MT binding, naturally lead to this transport scenario. Remarkably, even small-scale MT oscillations induce a long-distance transport on the cellular scale. Hitchhiking on approximately symmetric MT bundle arrangements as in fig. 1a inherently bears the feature of velocity and run-length distribution symmetry often observed in BDT [6,7]. Occasionally observed very large moment velocities [7,9,12] (up to $>10\,\mu\mathrm{m/s}$) —another characteristic of BDT— may be attributable to the enhanced motor efficiency known for cooperatively synchronized motor batteries performing filament sliding [26] and elastic MT relaxations [14]. Other signature behaviors of our mechanism, such as visible sliding of MTs, tandem motion of vesicles, exponential velocity relaxations and strong MT bending deformations have been observed recently [12,14]. From the perspective of our model the role of BDT regulators like dynactin or klar might be in passively attaching MTs to motors and other cellular structures (*e.g.*, nuclear lamina or cortex) and establishing the motor-cytoskeletal connectivity required for its efficient active fluctuations.

In general, we expect a combination of hitchhiking and hauling mechanisms to be present in varying proportions for different cargos and cells. Indeed in certain systems both types of motors are known to reside on the cargo [8], and likely some form of direct (possibly coordinated) competition between kinesin and dynein gives rise to bidirectional transport in those cases. At the same time vigorous MT motion seems to dominate cargo motion in other systems [12,14]. Further experiments will be needed to disentangle the relative contributions of both mechanisms and the cellular "master plan" for employing either of them in a particular system.

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[1]The typical stored slack length of each filament can be shown to be $\Delta l \approx l_c^2 / (6 l_p)$. For intermediate filaments with the persistence length $l_p \approx 1\,\mu\mathrm{m}$ [21] and with an entanglement/crosslinking distance of $l_c \approx 1\,\mu\mathrm{m}$ we have $\Delta l \approx 150\,\mathrm{nm}$. Taking into account that several parallel layers of entangled and crosslinked intermediate filaments can bridge beween the MTs proportionally larger stored lengths are possible.

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