Continuous model for microtubule dynamics with catastrophe, rescue, and nucleation processes

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Microtubules are a major component of the cytoskeleton distinguished by highly dynamic behavior both *in vitro* and *in vivo* referred to as dynamic instability. We propose a general mathematical model that accounts for the growth, catastrophe, rescue, and nucleation processes in the polymerization of microtubules from tubulin dimers. Our model is an extension of various mathematical models developed earlier formulated in order to capture and unify the various aspects of tubulin polymerization. While attempting to use a minimal number of adjustable parameters, the proposed model covers a broad range of behaviors and has predictive features discussed in the paper. We have analyzed the range of resultant dynamical behavior of the microtubules by changing each of the parameter values at a time and observing the emergence of various dynamical regimes that agree well with the previously reported experimental data and behavior.

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I. INTRODUCTION

Microtubules are protein polymers made of α/β tubulin heterodimers that form an essential part of the cytoskeleton of all eukaryotic cells. Besides giving structural stability and rigidity to a cell, microtubules play key roles in many physiological processes such as intracellular vesicle transport and chromosome separation during mitosis. An individual microtubule is a hollow cylinder of 25 nm diameter built usually from 13 protofilaments [1]. While the stable subunits of microtubules are actually heterodimers composed of α and β monomers, we will refer to them for simplicity as monomers. These monomers exist in two different energetic states, namely, bound to a molecule of guanosine triphosphate (GTP) or guanosine diphosphate (GDP), respectively. Only the GTP bound monomers are assembly competent meaning they are able to polymerize into microtubules. After the GTP monomers have been added to the growing microtubule, GTP bound to β tubulin is rapidly hydrolyzed (dephosphorylated) to form a bound GDP subunit. It has been hypothesized since the early 1980s that the so-called GTP cap on the tip of the growing microtubule gives rise to the stability of the microtubule [2]. Once the GTP cap is lost, the microtubule will switch to a "collapsing" state referred to as a catastrophe which is characterized by rapid depolymerization of the microtubule into its free subunits. However, there is also a possibility that at some point in time a catastrophically shrinking microtubule acquires a new GTP cap and thereby returns to the growing population, a situation that is referred to in the literature as a rescue event. We refer the reader to the papers [1,3–9] and the references therein for more information about this phenomenon known in the literature as *dynamical instability*. The process of microtubule polymerization dynamics both *in vitro* and *in vivo* has been exhaustively reviewed by Desai and Mitchison [10]. It should be stressed that under the conditions of high concentration of tubulin a completely different process has been observed, namely, a transition to a regime with damped oscillations of the tubulin mass polymerized into microtubules. This occurs for free tubulin concentrations exceeding a critical value, which depends on the experimental conditions and which corresponds to saturable polymerization kinetics [11,12]. Dynamic instability of microtubules has been observed *in vivo* and *in vitro* [2] and highlights both the nonequilibrium nature of the physical problem involved and the stochastic nature of the individual microtubule growth process. Interestingly, ensembles of microtubules show collective oscillations under suitable conditions first reported by Carlier *et al.* [11] and further studied by Mandelkow *et al.* [12–14] who showed that above a critical value of the free tubulin concentration, the amount of polymerized tubulin undergoes smooth oscillations which are damped out as the biochemical energy source in the form of GTP molecules is gradually depleted. Sept *et al.* [9] modeled the assembly dynamics from a chemical reaction-kinetics standpoint and found good agreement with the experimental data. While this model captures the main features of the process, it is still very empirical and incomplete. Nonetheless, to introduce the main features of the underlying biochemical processes we have described them in Appendix B. These processes involve polymerization/depolymerization, nucleation, and catastrophe events. When at least one autocatalytic reaction is added to the system, the dynamics changes significantly. Sept *et al.* [9], for example, considered an induced catastrophe event described by and incorporated into their model to reproduce oscillations observed *in vitro*. We will show in our paper that oscillations can be reproduced by including growth and nucleation in the model as the only nonlinear reactions.

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Deterministic mathematical models of polymer growth largely fall into two classes depending on whether the length of the polymer is discrete or continuous. The latter approximation is based on the assumption that the typical length of a polymer is much larger than the gain in length by adding a single monomer unit. Partial differential equation models of this type have been used, for example, in [1,7,9,14–17] for the case of microtubule dynamics and in [18] to study the dynamics of prion proliferation.

II. MATHEMATICAL MODEL

In this paper, we propose a mathematical model for the concomitant processes of microtubule growth, nucleation, catastrophic shrinking, and rescue. Roughly speaking, microtubules of length x grow at a velocity $\alpha p(t)$, where α is a constant and $p(t)$ is the concentration of free GTP tubulin. The model will therefore have the form of a nonlinear transport equation. This is essentially also the approach taken in some of the earlier works in this area [1,6,7,19]. Our intention is to develop a generalized model that accounts for the wealth of observed behavior including nucleation, growth to saturation, and synchronized oscillations, in addition to catastrophes and rescues. Moreover, we have explicitly included the presence of a GTP cap and a measure of the microtubule's age. While striving for completeness in the mathematical description we have also attempted to introduce a minimal number of model parameters. Our model contains seven empirical parameters, most of which can be determined from experimental data.

Let $Y=\{(x,y)\in\mathbb{R}^2: x>y>0\}$ be the state space for microtubules with a GTP cap. For $(x,y)\in Y$, let $u(x,y,t)$ denote the population density of microtubules of total length x that have a GTP cap and whose GDP domain has length y. This implies that

$$\|u(\cdot,\cdot,t)\|_{dydx}=\int_0^\infty\int_0^x u(x,y,t)dydx$$

is the total number of microtubules in a reference volume. We assume that the GDP domain forms a connected set and that the remainder of the microtubule is the GTP domain of length $x-y$; we will return to this assumption in Sec. IV below. If the microtubule has no GTP cap then it will undergo catastrophic depolymerization. In order to keep track of this process, let $v(x,t)$ denote the density of microtubules of length x without a GTP domain, again in the sense of a concentration. In addition, we introduce the concentrations of free GTP monomers p and free GDP monomers q. Catastrophic depolymerization of microtubules results in the release of GDP monomers which are then biochemically converted into GTP monomers in a reaction sometimes referred to as "pumping" since it involves a biochemical energy input from the solution. The new GTP monomers then become available for further microtubule growth, rescue, and nucleation. Figure 1 provides a schematic depiction of the tubulin cycle. The equation for u is given by

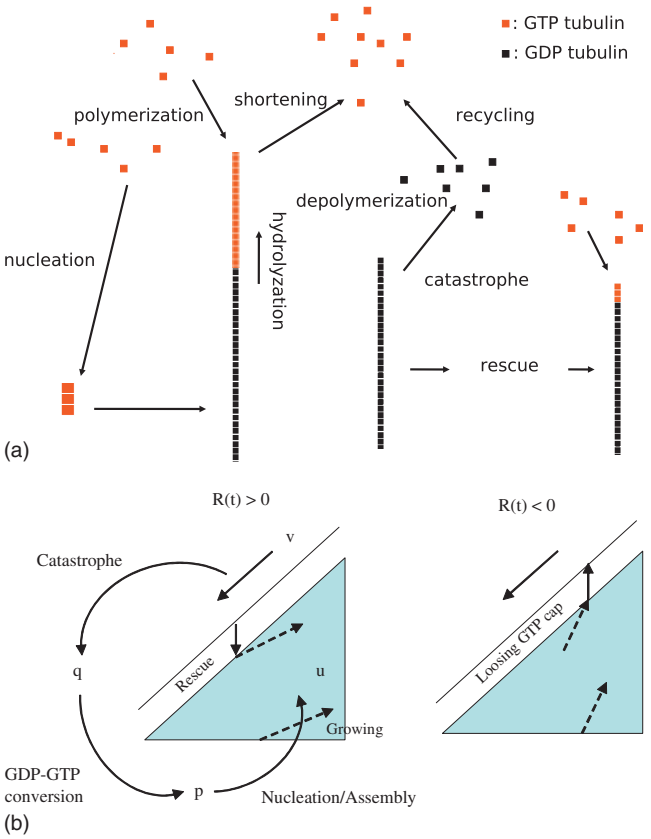

FIG. 1. (Color) Schematic representation of the tubulin cycle (top) and our model presented in this paper (bottom). In the case $R(t)=\alpha p(t)-\beta-\gamma>0$, the system is in a phase of growth (dashed arrows, left panel). If $R(t)<0$, the system is in a phase of shrinking (dashed arrows, right panel). The recycling of free monomers is identical in both cases and only depicted in the left panel.

$$\frac{\partial}{\partial t}u(x,y,t)+[\alpha p(t)-\beta]\frac{\partial}{\partial x}u(x,y,t)+\gamma\frac{\partial}{\partial y}u(x,y,t)=0. \tag{1}$$

The new monomers are added at rate $\alpha p(t)$ and result in an increase in the overall lengths of the microtubules. The constant $\gamma>0$ is the progression rate of the GDP zone (i.e., the speed of hydrolysis within the microtubule). The rate β can be positive if occasionally a GTP bound monomer is lost from the microtubule. Notice that both factors $\alpha p-\beta$ and γ have the dimension LT^{-1}. The characteristic curves for Eq. (1) are given by

$$\frac{dx}{dt}=\alpha p(t)-\beta,\quad \frac{dy}{dt}=\gamma.$$

In view of the second of these equations, the variable y can also be interpreted as the "age" of the microtubule, since hydrolysis is assumed to start immediately upon nucleation. The boundary condition on $\Gamma_1=\{(x,y)\in\mathbb{R}^2_{\geq 0}: y=0\}$ incorporates the nucleation of microtubules without a GDP domain. Let $\psi(x)$ be the length distribution of such freshly nucleated microtubules without a GDP domain. This can be a uniform distribution on some interval $[x_-,x_+]$ or a (narrow) Gaussian

distribution centered at some point x^0. Let $L^*=\int_0^\infty x\psi(x)dx$ be the average length of freshly nucleated microtubules. The nucleation reaction is generally assumed to be a nonlinear reaction although the exact number of monomers n that need to come together is a matter of discussion [4,20,21]. With the rate of nucleation $\mu>0$, the boundary condition on Γ_1 is

$$\gamma u(x,0,t)=\frac{\mu}{L^*}p^n(t)\psi(x). \tag{2}$$

Thus, Γ_1 is part of the inflow boundary of the domain Y at all times. The boundary $\Gamma_2=\{(x,y)\in\mathbb{R}^2_{\geq 0}: x=y\}$ may be part of the inflow boundary, respectively, of the outflow boundary, depending on whether the growth of the entire microtubule is faster than the progression of the GDP domain. Precisely, let $R(t)=\alpha p(t)-\beta-\gamma$. If $R(t)>0$, then we say that the system is in a growth phase. This allows microtubules without a GTP cap to be rescued. The boundary condition on Γ_2 is then given by

$$R(t)u(x,x,t)=\lambda v(x,t), \quad \text{if } R(t)>0, \tag{3}$$

where $\lambda\geq 0$ is the propensity of shrinking microtubules to be rescued (it has the dimension T^{-1}). If, on the other hand, $R(t)<0$, then we say that the system is in a state of shrinking and Γ_2 is part of the outflow boundary of the domain Y. Microtubules reaching the boundary Γ_2 are transferred to the population of microtubules without a GTP cap $v(x,t)$. It will be helpful later to write Eq. (1) in divergence form. Let

$$\boldsymbol{b}(t)=\begin{pmatrix}\alpha p(t)-\beta\\ \gamma\end{pmatrix}, \tag{1'}$$

then Eq. (1) can be written as

$$u_t+\nabla\cdot[\boldsymbol{b}(t)u]=0.$$

Microtubules without a GTP cap are shrinking at a rate $\delta>0$ (which represents the loss of length per unit time). This population has a source or a loss term, again depending on the sign of the function R. The equation for v is

$$\frac{\partial}{\partial t}v(x,t)-\delta\frac{\partial}{\partial x}v(x,t)=\begin{cases}-R(t)u(x,x,t), & \text{if } R(t)<0\\ -\lambda v(x,t), & \text{if } R(t)>0\end{cases}. \tag{4}$$

If $R(t)<0$ the system is in a state of shrinking and microtubules that lose their GTP cap obviously enter the population of microtubules without a GTP cap. If the system is in a state of growth then microtubules without a GTP cap are rescued and re-enter the class u through boundary condition (3) on Γ_2.

GDP-bound monomers are gained by catastrophes (there are no intermediate depolymerization products) and are converted to GTP-bound monomers by biochemical pumping. The gain is proportional to the number of microtubules without a GDP domain. Hence,

$$\frac{d}{dt}q=\delta\int_0^\infty v(x,t)dx-\kappa q. \tag{5}$$

The constant $\kappa>0$ denotes the rate of the (first order) pumping reaction. We assume that there is always enough GTP available in the solution to ensure a constant rate of pumping, but in the future we may also include free GTP as a variable in the model. Recall that q and p are concentrations of "lengths" of microtubules stored in free GDP or GTP-bound tubulin monomers, respectively. In order to obtain the concentrations of molecules, one can calculate $\tilde{q}=\ell^{-1}q$, where ℓ is length gained by adding a single monomer. Flyvbjerg *et al.* [6,7] used a simple conversion where $\ell=8$ nm$/13=0.6$ nm, since 8 nm is the length of a single tubulin heterodimer and 13 is the number of protofilaments in a microtubule.

The population of free GTP-bound tubulin monomers is replenished by the conversion of GDP monomers while losses occur due to growth and nucleation of microtubules. Therefore,

$$\frac{d}{dt}p=-(\alpha p-\beta)\int_0^\infty\int_0^x u(x,y,t)dydx+\kappa q-\mu p^n. \tag{6}$$

The last term in Eq. (6) indicates that n individual monomers combine during nucleation. The set of initial conditions is

$$u(x,y,0)=u^0(x,y), \quad v(x,0)=v^0(x), \quad p(0)=p^0, \quad q(0)=q^0.$$

We calculate the total length $\|u\|_{xdydx}$ of GDP- and GTP-bound tubulin found in microtubules with a GTP cap by integrating with the weight $xdydx$,

$$\|u(\cdot,\cdot,t)\|_{xdydx}=\int_0^\infty\int_0^x u(x,y,t)xdydx.$$

The total length of GDP-bound tubulin in collapsing microtubules is given by a similar expression,

$$\|v(\cdot,t)\|_{xdx}=\int_0^\infty v(x,t)xdx.$$

The model [Eqs. (1)–(6)] conserves the total length of bound and free tubulin $\|u\|_{xdydx}+\|v\|_{xdx}+q+p$, i.e.,

$$\frac{d}{dt}[\|u(t)\|_{xdydx}+\|v(t)\|_{xdx}+q(t)+p(t)]=0. \tag{7}$$

See Appendix A for details. It is also possible to calculate the total length of GDP-bound tubulin found in microtubules with a GTP cap as

$$\ell_{\text{bound-GDP}}(t)=\int_0^\infty\int_0^x u(x,y,t)ydydx, \tag{8}$$

and so the complementary quantity, the total length of GTP-bound tubulin, is

$$\ell_{\text{bound-GTP}}(t)=\int_0^\infty\int_0^x u(x,y,t)(x-y)dydx.$$

III. PARAMETRIZATION AND NUMERICAL RESULTS

The parameters of our model are the following, each given with their physical dimensionality:

TABLE I. Experimental and/or computational estimates for parameters published in the literature, some of which are used in the model. k_{res} and k_{cat} are rescue and catastrophe rates that can be used to fine tune λ, κ, and γ.

Parameter	Value	Reference
α	0.5–11.5 μm min^{-1} μM^{-1}	[22–26]
β	1.6–35 μm min^{-1}	[1,22–28]
γ	0.25 μm min^{-1}	[7]
δ	44–50 μm min^{-1}	[26]
n	1–12	[4,9,20,21]
μ	5.9×10^3 M^{-1} min^{-1}	[21]
κ	3–120 min^{-1}	[4,9]
k_{res}	2–10 min^{-1}	[26,29]
k_{cat}	0.1–1 min^{-1}	[26,29]

(i) α, the growth rate of microtubules such that $\alpha p(t)$ has dimension LT^{-1};
(ii) β, rate of loss of a GTP-bound monomer (LT^{-1});
(iii) γ, progression rate of the GDP zone (LT^{-1});
(iv) δ, depolymerization rate of microtubules without a GTP cap (LT^{-1});
(v) κ, rate of the pumping reaction that converts GDP-bound monomers into GTP-bound monomers (T^{-1});
(vi) λ, rescue propensity for microtubules undergoing a catastrophe (T^{-1});
(vii) μ, rate of nucleation ($[p]^{-(n-1)}T^{-1}$);
(viii) n, the order of the nucleation reaction;
(ix) ψ, distribution of lengths of freshly nucleated microtubules;
(x) L^*, average length of freshly nucleated microtubules (L).

The dimensions of α and μ require some discussion. From Eq. (6) we see that $[\alpha p]=[\beta]=LT^{-1}$ and therefore $[\alpha]=[p]^{-1}LT^{-1}$. Hence, we are only able to use sources that report the growth rate as dependent on the concentration of free GTP-bound tubulin. Equation (6) implies further that $[\mu]=[p]^{-(n-1)}T^{-1}$. We collect in Table I a set of representative numerical values for the parameters that have been published in the literature.

We have implemented our model numerically using MATLAB; the codes will be available from the authors upon request. To simulate the system of Eqs. (1)–(6), we discretize Y into 500×500 cells where each cell has a dimension of 200 nm$\times$200 nm. We use an upwind scheme with adaptive time step for the partial differential equations and the explicit Euler method for the ordinary differential equations [30]. Since the amount of free GTP-tubulin $p(t)$ changes over time, so does the growth velocity of the microtubules and hence an adaptive choice of the time step is necessary to guarantee the Courant-Friedrichs-Lewy condition [30]. We continuously keep track of the total amount of tubulin in all its forms to guarantee that our numerical solution satisfies conservation law (7). Using the length of a single unit as stated above, we can convert a concentration of tubulin as follows:

$$1\ \mu\mathrm{M} \equiv 3.76\times10^{14}\frac{\mu\mathrm{m}}{\mathrm{L}} =: \mathrm{C}.$$

For consistency and comparison we choose most of the parameters from only two experimental sources [21,26]. Walker *et al.* [26] provided experimental estimates of the polymerization rate α, the loss rate of GDP monomers β, and the depolymerization rate δ. The values (summed on both plus and minus ends) are

$$\alpha = 2.5\ \mu\mathrm{m\ min^{-1}\ \mu M^{-1}} \equiv 1.33\times10^{-15}\ \mathrm{L\ min^{-1}}\left(=\frac{\alpha}{\mathrm{C}}\right),$$

$$\beta = 2.4\ \mu\mathrm{m\ min^{-1}}, \quad \delta = 50\ \mu\mathrm{m\ min^{-1}}.$$

These authors also estimate the rescue and catastrophe rates as 10 min^{-1} and 0.36 min^{-1} at 10 μM tubulin concentration, respectively. The latter are used to fine tune the other parameters of our model, i.e., γ, κ and λ. We note that the integral on the boundary Γ_2 in Eq. (A1) can serve as a definition of the catastrophe, respectively, rescue frequency, depending on the sign of R. Therefore, we define the time-average rescue and catastrophe rates from our model as follows:

$$k_{cat} = -\frac{1}{T}\int_0^T\int_{\Gamma_2} u(x,x,t)dxR(t)dt, \quad \text{if } R(t)<0,$$

$$k_{res} = \frac{1}{T}\int_0^T\int_{\Gamma_2} u(x,x,t)dxR(t)dt, \quad \text{if } R(t)>0, \qquad (9)$$

where T is the total simulation time. For the nucleation reaction we assume that $n=2$ and $\mu=5.9\times10^{-3}$ μM^{-1} min^{-1} ([21], Table I).

As one initial distribution of microtubules with a GTP cap we choose

$$u^0(x,y) = c\ \exp\left(-\frac{(x-10)^2}{5^2}-\frac{(y-5)^2}{2.5^2}\right), \qquad (10)$$

where the constant c is chosen such that $\|u^0\|_{xdydx}\equiv5$ μM; this is half the concentration of the total bound tubulin. The initial concentration of free GTP-bound tubulin is $p^0\equiv5$ μM. The remaining two initial data are chosen to be 0.

In the first modeling scenario we set the parameters as $\kappa=\lambda=\mu=0$ (no conversion of GDP monomers, no rescue, and no nucleation); see Fig. 2. We plot the time evolution of microtubules in u (total amount of tubulin bound in microtubules with a GTP cap, solid red curve) and v (total amount of tubulin bound in microtubules without a GTP cap, blue curve) pools, and GTP and GDP tubulin dimers in p (green curve) and q (black curve) pools, respectively. After an initial period of growth, microtubules enter the depolymerization phase due to low concentration of the GTP tubulin (green curve). This is because the GDP-GTP conversion process of monomers is turned off ($\kappa=0$). As a result, the total length of microtubules decreases. Due to lack of nucleation and rescue ($\mu=0=\lambda$), this leads to complete depolymerization of all microtubules in the system, as expected. The dashed red curve in Fig. 2 represents the length of GDP-bound tubulin within microtubules with a GTP cap [see Eq. (8)].

In agreement with experimental observation [13], a solution showing damped oscillations can be found by introduc-

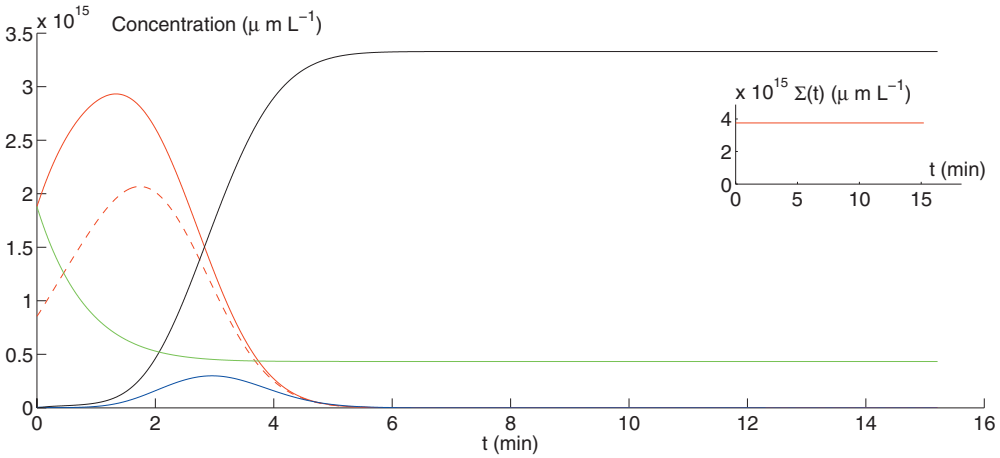

FIG. 2. (Color) Time evolution of microtubules for the parameter set $\alpha=2.5\ \mu\text{m min}^{-1}\ \mu\text{M}^{-1}$, $\beta=2.4\ \mu\text{m min}^{-1}$, $\gamma=5.4\ \mu\text{m min}^{-1}$, $\delta=50\ \mu\text{m min}^{-1}$, and $\mu=\kappa=\lambda=0$. Shown are the total amounts of tubulin in each of its forms, namely, bound in microtubules with a GTP cap $\|u\|_{xdydx}$ (solid red curve), bound in microtubules without a GTP cap $\|v\|_{xdx}$ (blue curve), and tubulin monomers bound to GTP p (green curve) and bound to GDP q (black curve). All quantities are in units of μmL^{-1} of tubulin. The total length of microtubules is decreasing at later times due to complete depolymerization of microtubules without a GTP cap and lack of nucleation. The red dashed curve represents the amount of bound GDP tubulin in microtubules with a GTP cap. The inset plot shows the conservation of the total amount of tubulin confirming Eq. (7).

ing recycling of GDP tubulin, and nucleation and rescue processes. Using the parameters $\kappa=1\ \text{min}^{-1}$, $\mu=5.9\times10^{-3}\ \mu\text{M}^{-1}\ \text{min}^{-1}$, and $\lambda=0.136\ \text{min}^{-1}$, the result is shown in Fig. 3. From now on we refer to this parameter set along with $\alpha=2.5\ \mu\text{m min}^{-1}\ \mu\text{M}^{-1}$, $\beta=2.4\ \mu\text{m min}^{-1}$, and $\delta=50\ \mu\text{m min}^{-1}$ as the "standard" and make all changes with respect to these values. In Fig. 3, kinks in the curves indicate rescue events that occur when microtubules without a GTP cap acquire a GTP cap under favorable growth conditions. Setting the rescue rate λ to zero results in very similar curves without the discontinuities (not shown). The resulting rescue and catastrophe rates are $k_{\text{res}}=6.7899\ \text{min}^{-1}$ and $k_{\text{cat}}=0.1392\ \text{min}^{-1}$ which are within experimentally observed ranges (see Table I). Another interesting observation in Fig. 3 is that the GDP zone (of the entire population) follows quite closely the total length. This means that on average, microtubules maintain their GTP cap. Figure 4 depicts the time evolution of the population density u for the set of parameters described in Fig. 3. As shown, the microtubules continue to grow and shrink in time. Another interesting initial distribution of microtubules with a GTP cap is $u^0(x,y)=0$ and $p^0\equiv10\ \mu\text{M}$. In this scenario the influence of nucleation can be studied; see Fig. 5. As shown, microtubules are nucleating, polymerizing, and then growing while the GDP-bound portion is also progressing.

We have tested numerically the influence of the individual parameters on the behavior of our model. Using the initial datum from Eq. (10), we have varied one parameter at a time. In order to unify the picture somewhat, we arrange several growth curves in two-dimensional slices of the pa-

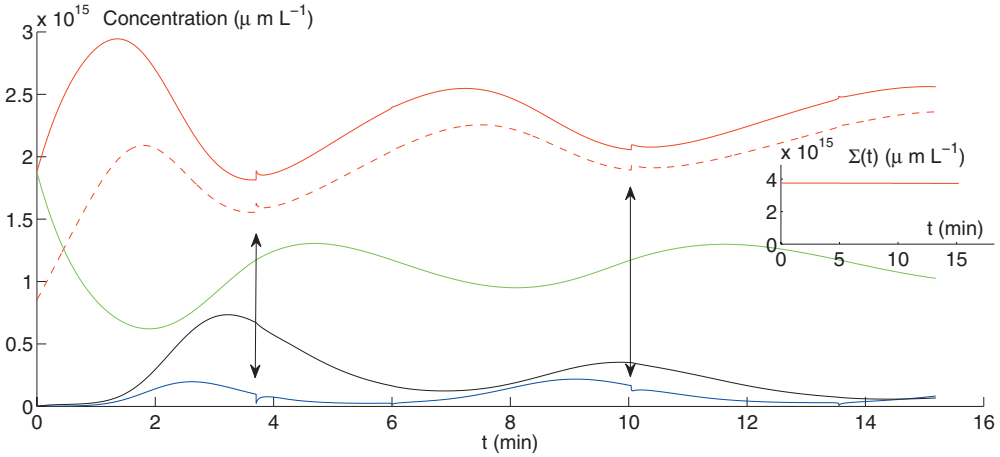

FIG. 3. (Color) Same as Fig. 2 but for the parameter set $\mu=5.9\times10^{-4}\ \mu\text{M}^{-1}\ \text{min}^{-1}$, $\kappa=1\ \text{min}^{-1}$, and $\lambda=0.136\ \text{min}^{-1}$. The arrows indicate rescue events; the conservation of the total amount of tubulin is shown in the inset plot. The resulting catastrophe and rescue rates are $k_{\text{cat}}=0.1394\ \text{min}^{-1}$ and $k_{\text{res}}=6.7899\ \text{min}^{-1}$ [see Eq. (9)]. The parameters α, β, γ, and δ used in Fig. 2 and μ, κ, and λ used here are referred to as to the "standard" parameter set from now on.

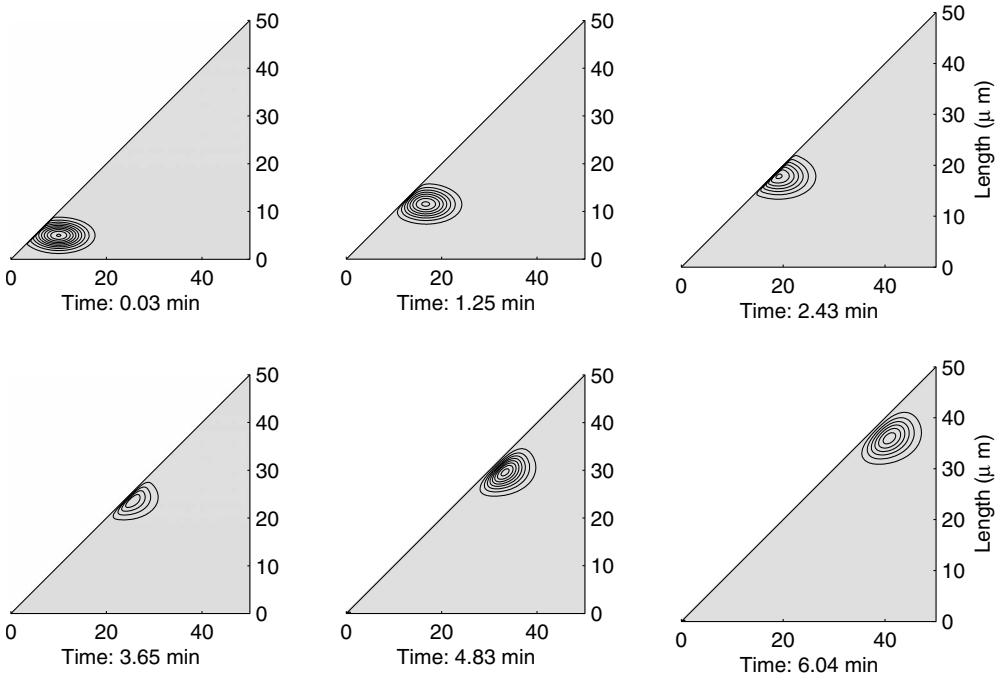

FIG. 4. Time evolution of the population density u for the set of parameters used in Fig. 3. Plotted are equidensity contours beginning from a Gaussian profile. Note the change in direction of the center as time progresses due to the activation of the rescue process.

rameter space. In the following, we state variation in the relevant parameter as a multiple of the standard value, which is denoted by an asterisk. A decreased rate of monomer addition $\alpha \leq 0.3\alpha^*$ results in no growth of microtubules during 10 min (Fig. 6, left column). An increase in the loss rate of GTP-bound monomers to $6\beta^*$ results in the formation of much shorter microtubules with a very short GTP cap (Fig. 7). A reduced rate of GTP hydrolysis allows the formation of large GTP domains and delays the onset of the oscillation phase (Fig. 8, bottom row). On the other hand, large values of γ are able to suppress microtubule formation completely (Fig. 8, top row). It is worth noting that in Fig. 3 we observe a crossover from a regime dominated by low-level microtubule assembly to saturation kinetics followed by a polymerization "overshoot" with damped oscillations. The latter is a characteristic feature found experimentally by Marx *et al.* [12] almost 20 years ago.

Changes in the depolymerization rate of microtubules without a GTP cap δ lead to interesting behavior. Strong oscillations and a prolonged existence of microtubules without a GTP cap are seen (Fig. 9) when δ is small and rescue events are more pronounced due to a longer survival of microtubules without a GTP cap. A very important role is played by the rate κ at which GDP monomers are recycled to GTP monomers. Even a very small value such as $0.1\kappa^*$ suffices to induce oscillations, although their period is much longer (Fig. 6, bottom row). On the other hand, a large value such as $10\kappa^*$ results in a quick dampening of the oscillations (Fig. 6, top row). Finally, the parameter μ influences the time

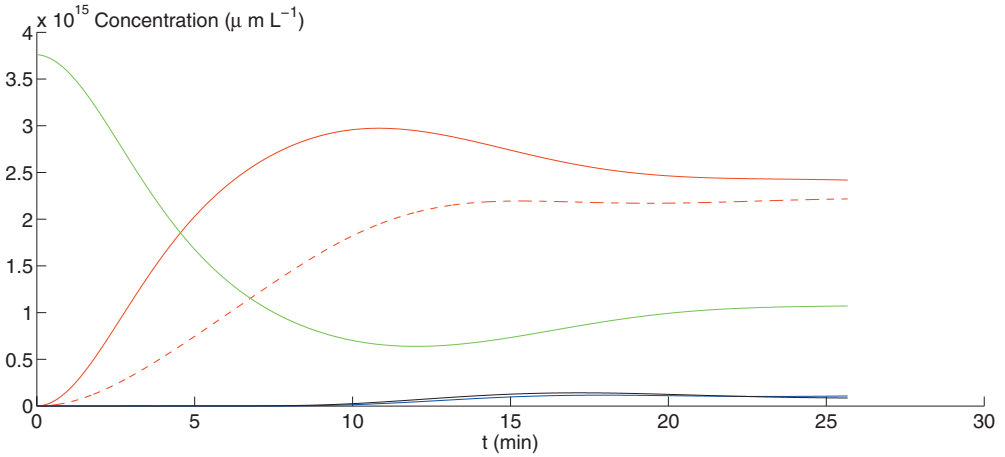

FIG. 5. (Color) Shown are again the total amounts of tubulin in each of its forms ($\|u\|_{xdydx}$, solid red curve; $\|v\|_{xdx}$, blue curve; p, green curve; and q, black curve). Parameters are the same as in Fig. 3 but with initial conditions $\|u^0\|_{xdydx}=0$ and $p^0=10$ μM.

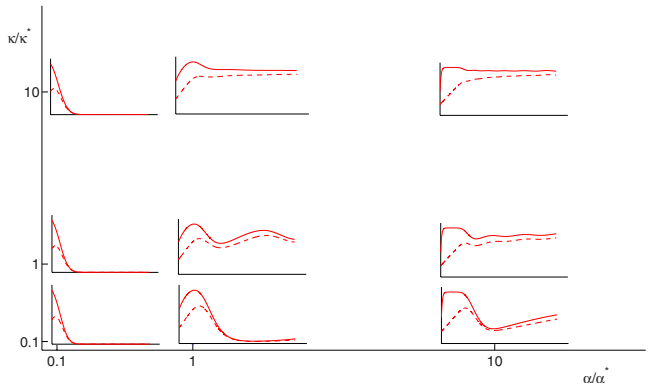

FIG. 6. (Color online) The behavior at several points in parameter space when α and κ are varied simultaneously. Each subfigure shows only the amount of tubulin in microtubules with a GTP cap (solid red curves) and the length of the trailing GDP subdomain (dashed red curves). Parameters are varied with respect to the values $\alpha^*=2.5\ \mu\text{m min}^{-1}\ \mu\text{M}^{-1}$ and $\kappa^*=1\ \text{min}^{-1}$. The time axis covers 10 min in each case.

scale for the nucleation process but does not change the general behavior drastically (not shown).

In addition to these variations in parameters we also varied one initial datum, namely, the concentration of free GTP tubulin p^0. Now using the standard parameter set again, we see that a high concentration of $p^0 \equiv 10\ \mu\text{M}$ and $u^0(x,y)$ as in Eq. (10) results in a longer persistence of oscillations (Fig. 10). Our model provides results regarding the nature of the oscillatory kinetics for microtubule assembly which is consistent with the original experimental observations reported by Carlier *et al.* [11], specifically the data shown in their Fig. 1 where the amplitude of oscillations and their persistence diminish as the concentration of free tubulin decreases from 150 to 50 μM. The actual values of the tubulin concentration used in the experiment may not be directly comparable to our values due to the specific experimental conditions under which the measurements were made.

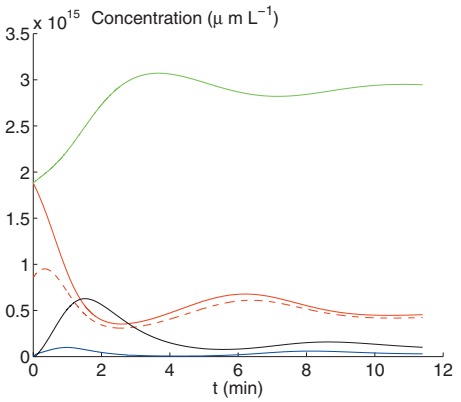

FIG. 7. (Color) An increase in the loss rate of GTP monomers β to $6\beta^*$ does not prevent formation of microtubules entirely but reduces drastically their average length. Notice also the extremely short GTP cap. All other parameters are the same as in Fig. 3.

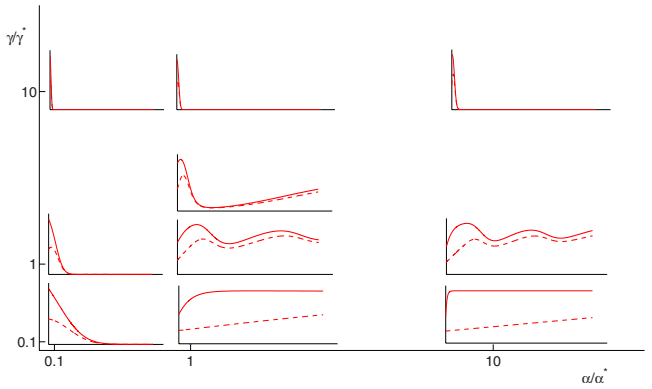

FIG. 8. (Color online) As in Fig. 6, but now α and γ are varied simultaneously.

IV. DISCUSSION

The dynamical behavior of microtubules has attracted many investigators over the past few decades to examine the microtubule behavior in regard to many biophysical aspects [1,3,4,6–9]. In this paper, we have proposed a mathematical model that includes all processes taking place during microtubule polymerization/depolymerization, namely: growth, nucleation, catastrophic shrinkage, and rescue events. Our model contains the amounts of free tubulin in both its energetic forms (GTP and GDP bound) as dependent variables. This results in a nonlinear transport equation whose mathematical analysis will pose a serious challenge. Nevertheless, we think this will give valuable insight into the role of the GTP cap in maintaining microtubule stability. The earlier paper by Houchmandzadeh and Vallade [17] contained expressions for free GTP and GDP tubulin; it did not contain a growth velocity for microtubules that was dependent on the amount of free GTP tubulin. GTP monomers continue to be lost, even beyond the point when they have become completely depleted [[17], Eq. (9)]. This results in possibly negative concentrations of GTP monomers avoided in our treatment.

A crucial assumption of our model is that the microtubules consist of two separated and connected domains, a GTP domain where the growth occurs and a "trailing" GDP domain. Only very recently, a paper by Dimitrov *et al.* [31] provided experimental evidence for the presence of a GTP cap in microtubules *in vivo*. The authors of [31] suggest further that remnants of GTP tubulin left in the GDP domain play a role during rescue events and that growth resumes after such remnants have been exposed during a catastrophic depolymerization. This is clearly an exciting new development. Nevertheless, the remnants are likely to be very short (say, one layer of GTP tubulin). It should be clarified that our model describes a large population of growing and shrinking microtubules where stochastic events in an individual microtubule have been averaged out.

We should also point out that our model does not contain a diffusion term of the type $\text{div}(D\nabla u)$ on the right-hand side of Eq. (1). Such a term has been a prominent feature of earlier models [6,7] although it is not contained in other models [19]. The role of "length diffusion" in a mathematical

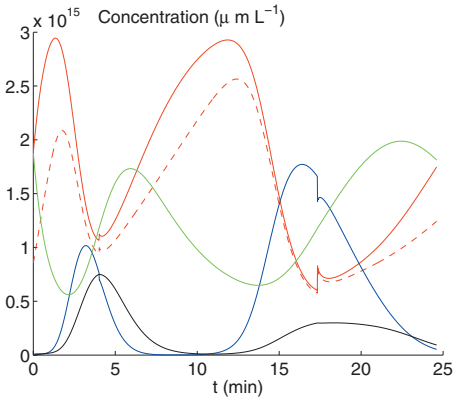

FIG. 9. (Color) A decrease in the depolymerization rate of collapsing microtubules to $0.2\delta^*$ results in irregular oscillations and longer presence of microtubules without a GTP cap. All other parameters are the same as in Fig. 3.

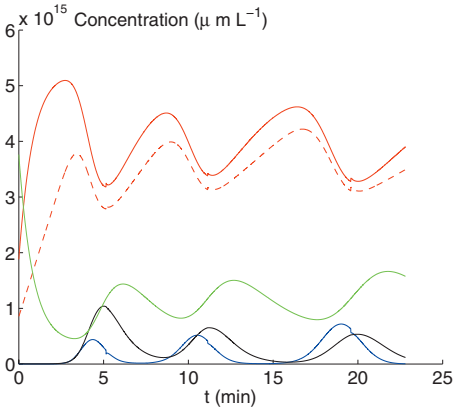

FIG. 10. (Color) Reverting to the standard parameter set (see Fig. 3) and using the initial condition $p^0 \equiv 10$ μM gives oscillations that persist for longer times.

model for linear polymer accretion has been investigated from a mathematical point of view [32,33]. Collet *et al.* [32] and Laurençot and Mischler [33] discussed convergence of the solutions of the discrete Becker-Döring system to solutions of the continuous Lifshitz-Slyozov equation under certain scaling assumptions. The "standard" version of the Lifshitz-Slyozov equation in [32,33] does not contain a diffusion term implying that a population highly concentrated at a certain length initially does not disperse later on.

We have performed numerical simulations using mainly parameters from only two experimental sources, namely, [21,26]. Already with a few choices and variations in parameters, we are able to reproduce commonly seen dynamical behaviors such as complete depolymerization in case of lacking recycling of GDP monomers (see Fig. 2) and damped oscillations in a growing population (Fig. 3). The parameters that have not yet been determined experimentally are—to the best of our knowledge—the rescue rate λ and the pumping rate κ, although the order of magnitude of κ has been estimated by theoretical arguments [4,9]. By varying these parameters in simulations we can predict their influence on the growth behavior and suggest experimental scenarios to look for. While outside of the scope of the present paper, we want to point out that microtubule polymerization and depolymerization is the target of many cancer chemotherapy drugs. The precise mechanism by which some drugs (such as vinblastine and taxol) suppress dynamic instability is a topic for future modeling and experimental research.

Some important points that we plan to address in the future are [34] as follows:

(i) The GTP zone is generally believed to be short, a few helical rings or 40 nm at most [35]. This would imply that hydrolysis, under typical conditions, proceeds at roughly the same speed as the growth of the microtubules which in turn is dependent on the concentration of unpolymerized GTP-bound tubulin. Is hydrolysis of polymerized GTP-bound tubulin a catalyzed reaction?

(ii) It is an open question whether the model developed in this paper can be adopted to describe situations corresponding to *in vivo* conditions such as the presence of microtubule associated proteins during the polymerization process and the existence of discrete microtubule organizing centers. Some recent papers emphasize their role [36,37] in realistic representations of cellular processes.

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APPENDIX A: BALANCES FOR TUBULIN AND MICROTUBULES

We integrate Eq. (1) over the domain Y and apply the divergence theorem for weighted integrals

$$\int_\Omega (\nabla \cdot \boldsymbol{b}) w(\xi) d\xi = \int_{\partial\Omega} (\boldsymbol{b} \cdot \boldsymbol{n}) w(\xi) d\sigma(\xi) - \int_\Omega \nabla w \cdot \boldsymbol{b} d\xi,$$

where $\boldsymbol{n}$ is the outer normal vector. With $w(x,y)=x$ and $\boldsymbol{n}=\binom{0}{-1}$ on Γ_1 and $\boldsymbol{n}=\binom{-1}{1}$ on Γ_2 this gives, interchanging the order of integration and differentiation,

$$\frac{d}{dt}\|u(t)\|_{xdydx} = \int_{\Gamma_1} \gamma u(x,0,t) x dx - \int_{\Gamma_2} \{-[\alpha p(t) - \beta] + \gamma\} u(x,x,t) x dx + \int_Y [\alpha p(t) - \beta] u(x,y,t) dy dx = \mu p^n(t)$$

$$+\left\{\begin{array}{ll} R(t)\displaystyle\int_0^\infty u(x,x,t)xdx, & \text{if } R(t)<0 \\ \lambda\displaystyle\int_0^\infty v(x,t)xdx, & \text{if } R(t)>0 \end{array}\right\}$$

$$+[\alpha p(t)-\beta]\int_0^\infty\int_0^x u(x,y,t)dydx.$$

Likewise, we integrate Eq. (4) with weight xdx. Again, after integration by parts, we have

$$\frac{d}{dt}\|v(t)\|_{xdx}=-\left\{\begin{array}{ll} R(t)\displaystyle\int_0^\infty u(x,x,t)xdx, & \text{if } R(t)<0 \\ \lambda\displaystyle\int_0^\infty v(x,t)xdx, & \text{if } R(t)>0 \end{array}\right\}$$

$$-\delta\int_0^\infty v(x,t)dx.$$

Adding these two results and Eqs. (5) and (6) yields Eq. (7).

If the weight x is removed then we obtain the total number of microtubules with or without a GTP cap. For microtubules with a GTP cap we obtain

$$\frac{d}{dt}\|u(t)\|_{dydx}=\int_{\Gamma_1}\gamma u(x,0,t)dx+[\alpha p(t)-\beta$$

$$-\gamma]\int_{\Gamma_2}u(x,x,t)dx=\mu p^n(t)\int_{\Gamma_1}\psi(x)dx$$

$$+R(t)\int_{\Gamma_2}u(x,x,t)dx, \tag{A1}$$

that is, such microtubules are gained through nucleation and lost or gained [depending on the sign of $R(t)$] through exchange with the population without a GTP cap. For the latter we have

$$\frac{d}{dt}\|v(t)\|_{dx}=-\delta v(0,t)-R(t)\int_{\Gamma_2}u(x,x,t)dx,$$

i.e., microtubules without a GTP cap are lost by complete depolymerization and gained or lost through exchange with the population with a GTP cap. Taken together

$$\frac{d}{dt}[\|u(t)\|_{dydx}+\|v(t)\|_{dx}]=\mu p^n(t)\int_{\Gamma_1}\psi(x)dx-\delta v(0,t),$$

that is, only nucleation and complete depolymerization change the total number of microtubules.

APPENDIX B: BIOCHEMISTRY OF TUBULIN POLYMERIZATION

The principal elements in the model of Sept *et al.* [9] can be summarized by the equations that follow. For simplicity, the microtubule is considered as a linear polymer rather than an object of 13 protofilaments. We shall denote a microtubule of n subunits by MT_n. Note that in solution, the free tubulin subunits may be bound to either GTP or GDP at their exchangeable nucleotide site and we denote them as T_{GTP} and T_{GDP}, respectively. Note that only tubulin bound to GTP is able to polymerize.

The reaction set involved in the process consists of addition, nucleation, and catastrophic collapse given by

$$\mathrm{MT}_n+T_{GTP}\leftrightharpoons\mathrm{MT}_{n+1},$$

$$nT_{GTP}\overset{k_n}{\longrightarrow}\mathrm{MT}_n,\quad\text{and}$$

$$\mathrm{MT}_n\overset{k_c}{\longrightarrow}nT_{GDP}.$$

For simplicity, it is assumed here that all collapses are complete and the number of dimers, n', required for nucleation is an adjustable parameter not available directly from experiment. These equations can also be supplemented by the reactivation of tubulin, to make it assembly competent, which will occur when the concentration of GTP is high,

$$T_{GDP}+\mathrm{GTP}\overset{k_r}{\longrightarrow}T_{GTP}+\mathrm{GDP}.$$

The free-energy change associated with this reaction is less than the free-energy change of GTP hydrolysis in solution and the difference is attributed to a structural change in the tubulin dimer. It is this conformational change which presumably makes assembly possible.

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