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ORIGINAL ARTICLE

A Quasistationary Analysis of a Stochastic Chemical Reaction: Keizer's Paradox

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Abstract For a system of biochemical reactions, it is known from the work of T.G. Kurtz [J. Appl. Prob. 8, 344 (1971)] that the chemical master equation model based on a stochastic formulation approaches the deterministic model based on the Law of Mass Action in the infinite system-size limit in finite time. The two models, however, often show distinctly different steady-state behavior. To further investigate this "paradox," a comparative study of the deterministic and stochastic models of a simple autocatalytic biochemical reaction, taken from a text by the late J. Keizer, is carried out. We compute the expected time to extinction, the true stochastic steady state, and a quasistationary probability distribution in the stochastic model. We show that the stochastic model predicts the deterministic behavior on a reasonable time scale, which can be consistently obtained from both models. The transition time to the extinction, however, grows exponentially with the system size. Mathematically, we identify that exchanging the limits of infinite system size and infinite time is problematic. The appropriate system size that can be considered sufficiently large, an important parameter in numerical computation, is also discussed.

Keywords Stochastic models · Nonlinear · Chemical kinetics · Quasistationary · Uniform convergence

1. Introduction

Mathematical models provide quantitative characterizations of chemical and biochemical reaction kinetics (Érdi and Tóth, 1989; Epstein and Pojman, 1998). Traditional chemical kinetics in aqueous solutions, in terms of the concentrations as function of time, are usually modeled by deterministic differential equations based on the Law of Mass Action. Such models give satisfactory predictions for

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well mixed, macroscopic reaction systems. One of the most celebrated examples is the Oregonator: the mathematical theory for the Belousov–Zhabotinsky reactions (Noyes and Field, 1974; Epstein and Pojman, 1998; Murray, 2002).

Mathematical modeling of biochemical reaction systems in a living cell, however, requires a different approach. Many biochemical reactions responsible for signal transduction and gene regulations inside living cells involve protein molecules with only a small number of copies (Smolen et al., 2000; Turner et al., 2004). The fluctuations in the number of molecules may be of biological significance (Paulsson et al., 2000; Samoilov et al., 2005). Kinetics of such reactions, thus, are more realistically described by stochastic models which emphasize the discrete nature of molecular reactions and the randomness of their collisions (Érdi and Tóth, 1989).

The mathematical basis for stochastic chemical kinetics is the discrete-state, continuous-time Markov jump processes, known as birth-death processes in the probability literature (Taylor and Karlin, 1998) and master equations in the physics literature (Schnakenberg, 1976). This tradition began in the 1930s with the work of M.A. Leontovich for gas phase reactions (Leontovich, 1935) and was continued, sometime independently, by A.J.F. Siegert, M. Kac, M. Delbruck, A. Renyi, M. Lax and D.A. McQuarrie, among others. Comprehensive reviews of some of the history of this formulation can be found in McQuarrie (1967), Érdi and Tóth (1989), and Keizer (1987).

Stochastic simulations of complex chemical reaction systems were realized in the early 1970s (Sipos et al., 1974a,b). Current software packages used for the simulation of biochemical reactions commonly make use of algorithms based on the influential work of Gillespie (1976, 1977). An analytical solution to stochastic, open unimolecular reaction networks can be found in Heuett and Qian (2006). It is not possible, in general, to obtain analytical solutions to an open, non-unimolecular reaction system.

The relation between the stochastic theory of chemical kinetics and its deterministic counterpart has been extensively studied by Kurtz (1971, 1972) who has shown that in the limit of large system size and number of molecules (i.e. thermodynamic limit), the stochastic model becomes the expected deterministic ordinary differential equation (ODE). Furthermore, solutions with given initial values to the stochastic model approach to the respective solutions to the ODE (Kurtz, 1971; Ethier and Kurtz, 1986). Approximating the stochastic jump processes by diffusion processes with continuous fluctuations, however, is still not well understood (Hänggi et al., 1984; Baras et al., 1996; Ashih, 2001). The delicate issue is related to exchanging the limits for large number of molecules and for long time (Luo et al., 1984).

As the mathematical foundation of chemical reaction theory therefore, the chemical master equation approach has superceded the traditional deterministic models. Within the stochastic formulation, however, one of the unsolved theoretical problems is how to asymptotically obtain steady-state fluctuations in the limit of large system size. A resolution to this problem will significantly extend the validity of the stochastic approach to biochemical reaction systems in cells.

Interestingly, the issue of exchanging limits is also present even between a stochastic jump process and the ODE model. It is intimately related to the time

scales and how they depend upon the system size. This is most tellingly illustrated, through an example, by the late Professor Keizer in his wonderful text on nonequilibrium statistical thermodynamics (Keizer, 1987). The long-time behavior predicted by the stochastic model, if not interpreted properly, appears in direct disagreement with that of the ODE model. It was concluded that "A weakness of the master equation formulation is that if used uncritically, it can lead to physically meaningless results" (Keizer, 1987, p. 164).

In this paper, we present an extended analysis of the limiting processes of large N and large t through the stochastic mathematical technique known as quasistationarity (Allen, 2003). This technique has been used to resolve similar paradoxical issues in models involving population biology (Reddy, 1975; Nasell, 2001). The issue has not been as closely studied in terms of chemical reactions however. For example, in a comparison of deterministic and stochastic models of intracellular kinetics (Srivastava et al., 2002), extinction was noted as a possibility only in the stochastic model but its significance was not throughly discussed. The model presented here is meant to give a deeper understanding of the problem.

This paper is organized as follows. In Section 2, both the deterministic and stochastic models are analyzed; their steady states obtained. Materials in this section are elementary; nevertheless they are included for completeness. In Section 3, the time to extinction (i.e. the stochastic steady state), τ_m, is compared with the time to quasistationarity (which corresponds to the deterministic steady state), T_m, where m is the initial number of molecules in the system. It is shown that with increasing volume size, V, τ_m increases exponentially while T_m remains constant. Hence with sufficiently large V, that is, in the thermodynamic limit, the quasistationary state becomes the de facto stationary state, and the probability of approaching the stochastic stationarity becomes exponentially small. This is the salient feature in the disagreement between the stochastic jump processes and its diffusion processes approximation (Hänggi et al., 1984; Baras et al., 1996; Ashih, 2001). In Section 4, we discuss the significance of open system and irreversibility in the "paradox." In fact, for a closed system none of the above complex behavior is possible. Section 5 provides conclusions and some discussions.

2. Deterministic and stochastic models: Steady-state analysis and the Keizer's paradox

Let us consider the autocatalytic reaction system

$$A + X \underset{k_{-1}}{\overset{k_1}{\rightleftharpoons}} 2X, \quad X \xrightarrow{k_2} C. \tag{1}$$

This is a modified version of Keizer's original example which assumes $k_{-1} = 0$. In this reaction, the molecule X acts as a catalyst in transforming an A molecule into an extra X molecule. This reaction is reversed if two X molecules react and form one X and one A molecule. The X molecule can also be transformed into a C molecule by a unimolecular reaction, which is assumed to be irreversible. All chemical reactions have to be reversible in reality; in our case the backward rate constant in the second reaction is sufficiently small so it is negligible.

The system in (1) is assumed to be in an open environment, that is, the number of A molecules, n_a, remains constant. This means the level of A is constantly being monitored and a feedback system which controls the n_a exists. Therefore, while it is unlikely, X can be continuously created from A just by chance. It follows that there is no limit on the number of X in the stochastic system, in principle.

2.1. Deterministic model with the Law of Mass Action

The deterministic ODE model of this reaction is

$$\frac{\mathrm{d}x}{\mathrm{d}t} = k_1 x a - k_{-1} x^2 - k_2 x, \tag{2}$$

where $a = [A]$, the concentration of chemical A, $x = [X]$, the concentration of chemical X, and k_1, k_{-1}, k_2 are the reaction rate constants. This equation is derived from the Law of Mass Action. The ODE system has two steady states:

$$x_1^* = 0 \quad \text{and} \quad x_2^* = \frac{k_1 a - k_2}{k_{-1}}. \tag{3}$$

We assume that $a > \frac{k_2}{k_1}$.[1] Thus, the first zero steady state is unstable and the second positive one is stable (Murray, 2002).

The differential equation is separable and the explicit solution is (Murray, 2002):

$$x(t) = \frac{x_0(k_1 a - k_2)\mathrm{e}^{(k_1 a - k_2)t}}{k_1 a - k_2 - k_{-1}x_0 + x_0 k_{-1}\mathrm{e}^{(k_1 a - k_2)t}}. \tag{4}$$

It shows that if the concentration of X starts at either one of the values in Eq. (3), it will never change. If the system starts from any other point, the concentration will tend to the nonzero steady state $x_2^* = (k_1 a - k_2)/k_{-1}$ as time tends toward infinity.

The explicit solution indicates that although $x(t)$ tends toward x_2^*, the concentration only reaches this value in the limit as time goes to infinity. For a more realistic understanding of the solution, we can calculate how long it would take for $x(t)$ to come within a certain percentage of the steady state concentration. Substituting the expression $x_2^*(1-\epsilon)$ for $x(t)$ in Eq. (4) and solving for t, we have

$$t(\epsilon, x_0) = \frac{\ln\left(\frac{(1-\epsilon)(k_1 a - k_2 - k_{-1}x_0)}{x_0 k_{-1}\epsilon}\right)}{k_1 a - k_2} \tag{5}$$

$$\approx -\frac{\epsilon - \ln(k_1 a - k_2 - k_{-1}x_0) + \ln(k_{-1}) + \ln\epsilon}{k_1 a - k_2}, \tag{6}$$

[1] When $a < k_2/k_1$, $x_1^* = 0$ is the only physically meaningful steady state, which is stable. It is interesting to note that k_2/k_1 has a dimension of $[\text{volume}]^{-1}$. Hence, with decreasing system size, k_2/k_1 increases. This observation suggests another way to compare the deterministic and stochastic models while ak_1/k_2 are kept constant rather than $ak_1/k_2 \to 0$ for small system.

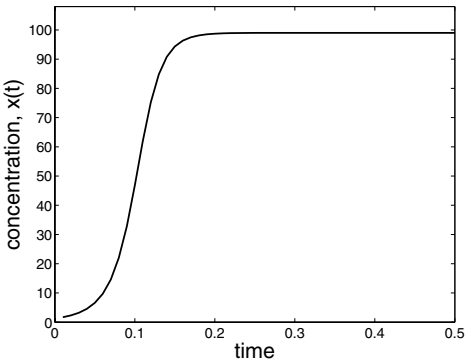

Fig. 1 Graph of concentration, $x(t)$ versus time in the deterministic model, with initial value $x(0) = 1$, according to Eq. (4). The parameter values are $k_1 = k_{-1} = k_2 = 0.05$, $a = 100$. Hence $x_2^* = 99$.

when ϵ is very small. The expression $t(\epsilon, x_0)$ gives us the time that it takes an initial condition x_0 to come within $100\epsilon\%$ of the stable steady state concentration, x_2^*. Note that for a fixed initial condition, $t(\epsilon, x_0)$ is asymptotically proportional to $-\ln \epsilon$, which is expected from any exponential relaxation process.

Figure 1 illustrates the behavior of the deterministic solution for parameter values $k_1 = k_2 = k_{-1} = 0.55$ and $a = 100$, using an initial condition of $x(0) = 1$. Equation (5) shows that the concentration $x(t)$ will reach 99.98% of the steady state concentration before $t = 0.25$. The concentration here is considered in units of $\frac{\text{(number of molecules)}}{\text{(volume)}}$ in order to match the units of the stochastic model in the following sections.

2.2. Stochastic model with exponential reactions

In the stochastic model for the reactions in (1), one considers the reactions in terms of their discrete, molecular events. That is, one considers the number of molecules of X, n, as opposed to the continuous concentration, $x = [X]$. Because of the Poissonian nature of the reaction, n is no longer a deterministic function of time. Rather, it is a random variable. Instead of asking "What is the number of molecules of X at time t?", we should now ask "What is the probability of the number of molecules of X being n at time t (called $p_n(t)$)?". This corresponds to a continuous-time, discrete-state Markov process with $n = 0, 1, 2, \ldots$. According to the Poissonian assumption, the number of molecules may only change by one at a time (Taylor and Karlin, 1998).

The mathematics for the stochastic model is a set ordinary differential equations, known as the forward Kolmogorov equation, describing the change of the probability $p_n(t)$ in time for each possible n. At each time step, there are three contributions to the change in $p_n(t)$: the two forward reactions and the backward reaction. Analogous to the deterministic model, each reaction is represented as the reaction rate multiplied by the product of the amount of each reactant.

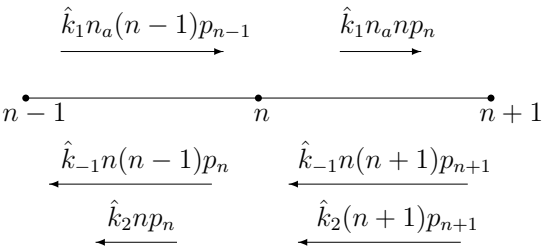

Fig. 2 Probability change from state P_n, where n_a is the number of A molecules.

The variable n in the stochastic model is related to the concentration x by

$$n = xV, \tag{7}$$

where V is the volume of the system. This parameter appears implicitly in both models of the system. For example, in the deterministic model, reaction rates k_1 and k_{-1} have units of V/t, and k_2 has units of $1/t$. The reaction rates in the stochastic model are related to these rates by

$$\hat{k}_1 = k_1/V \tag{8a}$$

$$\hat{k}_{-1} = k_{-1}/V \tag{8b}$$

$$\hat{k}_2 = k_2. \tag{8c}$$

These reaction rates are scaled so that the units agree in the Kolmogorov forward equation (see Eq. 9). Unless otherwise noted, we will use a value of $V = 1$ in calculations for simplicity.

Figure 2 illustrates how the probability of each state n is affected by these reactions. The change in each state, $\frac{\mathrm{d}p_n}{\mathrm{d}t}$ is the sum of the changes due to each of the three reactions. Thus, the stochastic model is the system of equations:

$$\frac{\mathrm{d}p_o}{\mathrm{d}t} = \hat{k}_2 p_1 \tag{9a}$$

$$\begin{aligned}\frac{\mathrm{d}p_n}{\mathrm{d}t} &= \hat{k}_1 n_a(n-1)p_{n-1} + (\hat{k}_{-1}n(n+1) + \hat{k}_2(n+1))p_{n+1} \\ &\quad - (\hat{k}_{-1}n(n-1) + \hat{k}_1 n_a n + \hat{k}_2 n)p_n \end{aligned} \tag{9b}$$

$$\vdots$$

where n_a represents the number of A molecules, which does not change in the model.

To find the steady state, we again set the equations $\mathrm{d}p_n/\mathrm{d}t = 0$. This yields an infinite set of homogeneous equations:

$$\frac{\mathrm{d}p_0}{\mathrm{d}t} = \hat{k}_2 p_1 = 0 \tag{10a}$$

$$\frac{\mathrm{d}p_1}{\mathrm{d}t} = (2\hat{k}_{-1} + 2\hat{k}_2)p_2 - (\hat{k}_1 n_a + \hat{k}_2)p_1 = 0 \tag{10b}$$

$$\frac{\mathrm{d}p_2}{\mathrm{d}t} = \hat{k}_1 n_a p_1 + (6\hat{k}_{-1} + 3\hat{k}_2)p_3 - (2\hat{k}_{-1} + 2\hat{k}_1 n_a + 2\hat{k}_2)p_2 = 0 \tag{10c}$$

$$\vdots$$

By induction, all probabilities p_n for $n > 0$ are zero. Because the sum of all the probabilities must add to one, this forces $p_0 = 1$. So we have

$$p^*(0) = 1 \quad \text{and} \quad p^*(n) = 0, \quad n > 0 \tag{11}$$

as the probability distribution for the unique steady state of the stochastic model. Note that the steady state of the deterministic model are fixed points, and the steady state of the stochastic model has a distribution.

The stochastic model shows that eventually there will be no X left in the system. This is in striking contrast to the previous deterministic model, which predicts that the concentration of X will stabilize at the nonzero x_2^*, while $x_1^* = 0$ is unstable. This is the Keizer's paradox.

We introduce the Keizer's paradox because the mathematical problem in the current example, though extremely simplistic, is at the core of the more general problem of approximating the discrete, master equation approach to biochemical kinetic systems with continuous Langevin dynamics with Fokker–Planck equation, that is, concentrations with fluctuations. Recent advances in system biology modeling of cellular biochemical reaction networks have been based on both approaches, but their consistency has been seriously questioned (McAdams and Arkin, 1999). Understanding the mathematical origin of the disagreement between the stochastic jump processes and its diffusion processes approximation is essential in developing a sound stochastic modeling framework of cellular biochemical reaction systems. From this perspective, the Keizer's paradox remains to be investigated for the more general stochastic kinetic models.

3. Analysis of the stochastic model: Time to extinction and quasistationary state

Should we interpret the inconsistency between the long-time behaviors of the stochastic and deterministic models simply as a "weakness of the stochastic approach" (Keizer, 1987), or something more insightful? We realize that the ODE in Eq. (2) can be rewritten in the form

$$\frac{\mathrm{d}x}{\mathrm{d}t} = rx\left(1 - \frac{x}{K}\right), \tag{12}$$

where $r = k_1 a - k_2$ and $K = \frac{k_1 a - k_2}{k_{-1}}$. This equation is commonly known in the field of population biology as the logistic growth model, with growth rate r and carrying capacity K. As our previous analysis indicated, there are two steady states: the unstable equilibrium $x_1^* = 0$ and the stable equilibrium $x_2^* = K$.

Population biologists have also been interested in the problem of population extinction. To address this problem, the stochastic version of the logistic population growth (Nasell, 2001) in terms of a birth and death process (Taylor and Karlin, 1998) has been studied (Allen, 2003). The mathematical formulation is very similar, but not identical to our Eq. (9). A general birth and death process has its forward Kolmogorov differential equations in the form

$$\frac{\mathrm{d}p}{\mathrm{d}t} = Qp(t), \tag{13}$$

where $p(t) = (p_0(t), p_1(t), \ldots, p_N(t))^{\mathrm{T}}$ and Q is called a generator matrix:

$$Q = \begin{pmatrix} -\lambda_0 & \mu_1 & 0 & \ldots \\ \lambda_0 & -\lambda_1 - \mu_1 & \mu_2 & \ldots \\ 0 & \lambda_1 & -\lambda_2 - \mu_2 & \ldots \\ \vdots & \vdots & \vdots & \ldots \end{pmatrix}, \tag{14}$$

where λ_i and μ_i are the birth and death rates at state i, respectively. For the present problem, let the initial condition be $X(0) = m$ so that $p_m(0) = 1$ and $p_i(0) = 0$ for $i \neq m$.

The birth and death models with Eq. (13) have a unique stationary distribution (Taylor and Karlin, 1998):

$$\pi_i = \frac{\lambda_0 \lambda_1 \cdots \lambda_{i-1}}{\mu_1 \mu_2 \cdots \mu_i} \pi_0, \qquad i = 1, 2, \ldots, \tag{15}$$

where

$$\pi_0 = \left(1 + \sum_{i=1}^{\infty}\right)^{-1}, \quad \theta_i = \frac{\lambda_0 \lambda_1 \cdots \lambda_{i-1}}{\mu_1 \mu_2 \cdots \mu_i} \tag{16}$$

For our chemical kinetic model, Eq. (9), the birth and death rates are

$$\lambda_i = \hat{k}_1 n_a i. \tag{17}$$

and

$$\mu_i = \hat{k}_{-1} i(i-1) + \hat{k}_2 i, \tag{18}$$

respectively. Hence, there is the inevitability of extinction since $\lambda_0 = 0$.

In our reaction system (1), the "birth" rate λ_n is a linear function of population size n because the bimolecular forward reaction $A + X \to 2X$ involves the A molecules with a fixed number n_a. The "death" rate μ_n is a quadratic function of the population size because the backward reaction involves the reaction of two X

molecules. As in the deterministic model, this quadratic term causes the stochastic model to be similar to a logistic process. Since our $\lambda_0 = 0$, this dictates $\pi_i = 0$ for all $i \neq 0$. Thus, the unique stationary probability is

$$\tilde{\pi} = (1, 0, \ldots, 0)^{\mathrm{T}}. \tag{19}$$

This is what we have previously concluded in Section 2.

3.1. Time to extinction and its expectation

The stochastic model predicts extinction of X molecules as the long-term behavior. How long will it take when the number of molecules is very large? Would it be possible that a nonzero quasisteady state exists in a more reasonable time? This is the focus of this paper. The following theorem gives the expectation of the (random) time it takes for a birth and death process to reach its extinction (Allen, 2003, pp. 240–241):

Theorem 1. Suppose $X(t)$, $t \geq 0$, is a continuous time, discrete space birth and death chain with $X(0) = m \geq 1$ satisfying $\lambda_0 = \mu_0 = 0$ and $\lambda_i > 0$ and $\mu_i > 0$ for $i = 1, 2, \ldots, N$. The expected time until extinction, τ_m, satisfies

$$\tau_m = \begin{cases} \frac{1}{\mu_1} + \sum_{i=2}^{\infty} \frac{\lambda_1 \cdots \lambda_{i-1}}{\mu_1 \cdots \mu_i} & m = 1 \\ \tau_1 + \sum_{s=1}^{m-1} \left[\frac{\mu_1 \cdots \mu_s}{\lambda_1 \cdots \lambda_s} \sum_{i=s+1}^{\infty} \frac{\lambda_1 \cdots \lambda_{i-1}}{\mu_1 \cdots \mu_i} \right] & m = 2, 3, \ldots, N. \end{cases} \tag{20}$$

Note we are guaranteed convergence of the infinite series through the ratio test because the λ_i terms are linear and the μ_i terms are quadratic. However, a more compact analytic expression for time to extinction is difficult to obtain. In Nasell (2001), it is shown that for the stochastic logistic model studied on a finite domain (i.e. $n \in \{0, 1, 2, \ldots, N\}$), the time to extinction grows exponentially with the size of the domain (the parameter N). In our case, the domain size is infinite, and our "size" parameter is the system volume, V. There is strong numerical evidence that the time to extinction in this case grows exponentially with volume (see below). An analytical result will hopefully be obtained in future work.

When numerically computing the time to extinction and the quasistationary steady state in the section below, we must truncate the infinite sum at some finite value, N. Because the value of τ_1 may change drastically depending on the size of N, it is necessary to use a sufficiently large value in order to obtain accurate results. The choice can be made based on a knowledge of how τ_1 depends upon N, as illustrated here. Using parameter values of $k_1 = k_2 = k_{-1} = .55$, and $n_a = 100$, the expected time to extinction was calculated on Matlab for various state space sizes, $N = 1$ to $N = 300$. The deterministic, stable steady state for these parameters is $x_2^* = 99$.

Figure 3 is a plot of τ_1 against N. For values of N below the deterministic steady state, the expected time to extinction is relatively low. For values of N at and above

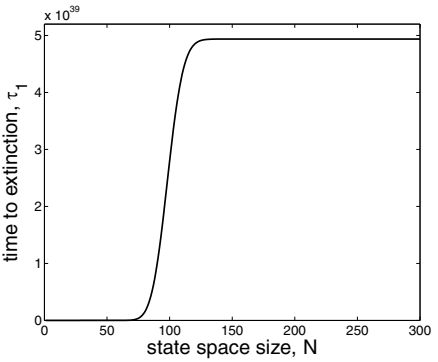

Fig. 3 Plot of τ_1 for fixed parameters $k_1 = k_{-1} = k_2 = 0.55$ and $n_a = 100$, and various state space sizes, N. Note that the expected time to extinction quickly rises to $O(10^{39})$ after $N = x_2^*$.

$N = x_2^* = 99$, τ_1 increases very quickly toward a constant value of almost 5×10^{39}. Thus, we require that N be "sufficiently larger" than the deterministic steady state, a concept which is quantified later in this paper (see Eq. 30).

Using a state space of $N = 300$ (and the same reaction rates as before), we can calculate values of τ_1 numerically for different volume sizes. Figure 4 shows a semi-logarithm plot of the system volume, V, versus the log of the time to extinction, $\log(\tau_1)$. The straight line shows an exponential relationship between volume size and time to extinction.

3.2. Quasistationary state and conditional probability

The steady-state distribution of the stochastic model does not make a good prediction of the behavior of the reaction (1). This is because the expected time to extinction is very long. Hence, it helps to consider an approximately stationary distribution in a reasonable time, if possible. Since the rate of extinction is very small, one is naturally interested in whether there exists a "quasistationary" probability

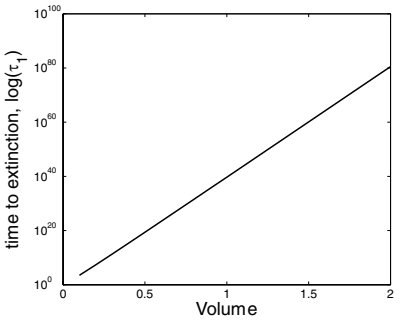

Fig. 4 Plot of volume size V against the logarithm of the time to extinction, $\log(\tau_1)$. As the system volume increases, the time to extinction explodes exponentially.

distribution of $X(t)$, given that there is still some X in the system. This is defined as

$$q_n(t) = P\{X(t) = n | X(t) > 0\} = \frac{p_n(t)}{1 - p_0(t)}, \qquad n = 1, 2, \ldots. \tag{21}$$

The steady-state distribution for the sequence $q_n(t)$ is called the quasistationary steady state of $p_n(t)$. The explicit distribution cannot be determined analytically because of the nonlinear nature of the model (Nasell, 2001). However, it can be approximated by using the assumption $\mu_1 = 0$, thereby eliminating the possibility of extinction.

Under the assumption $\mu_1 = 0$, the forward Kolmogorov differential equations are reduced by one dimension, yielding the system

$$\frac{\mathrm{d}q}{\mathrm{d}t} = \hat{Q}q(t), \tag{22}$$

where $q(t) = (q_1(t), q_2(t), \ldots, q_N(t))^{\mathrm{T}}$ and $\hat{Q}$ is the matrix Q (Eq. 14) with the first row and column removed:

$$\hat{Q} = \begin{pmatrix} -\lambda_1 & \mu_2 & 0 & \ldots \\ \lambda_1 & -\lambda_2 - \mu_2 & \mu_3 & \ldots \\ 0 & \vdots & \ldots & \vdots \end{pmatrix}. \tag{23}$$

Again, the steady state satisfies an infinite array of simultaneous equations described in Eq. (15). Stated recursively, the steady state is given by

$$\hat{\pi}_{i+1} = \frac{\lambda_i}{\mu_{i+1}}\hat{\pi}_i \quad i = 1, 2, \ldots \tag{24}$$

with the added restriction that the $\hat{\pi}_i$ must sum to 1 (Eq. 16).

To compute the quasistationary steady state numerically, we first assume that the state space is of a finite size, N. The steady state can then be found by solving the linear system

$$A\hat{\pi} = b \tag{25}$$

where A is the $N \times N$ matrix

$$\begin{pmatrix} 1 & 1 & \cdots & 1 & 1 \\ \frac{\lambda_1}{\mu_2} & -1 & 0 & \cdots & 0 \\ 0 & \frac{\lambda_2}{\mu_3} & -1 & \cdots & 0 \\ \vdots & 0 & \frac{\lambda_3}{\mu_4} & -1 & 0 \\ 0 & \cdots & 0 & \frac{\lambda_{N-1}}{\mu_N} & -1 \end{pmatrix} \tag{26}$$

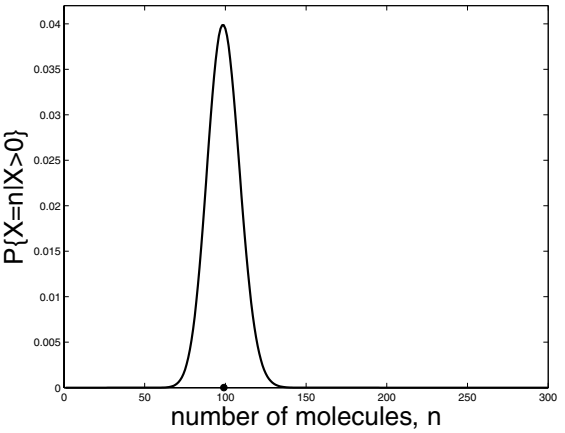

Fig. 5 Quasistationary distribution of the reaction using parameters $k_1 = k_{-1} = k_2 = 0.55$, $n_a = 100$, and $N = 300$.

and $\hat{\pi}$ and b are the $N \times 1$ vectors, $\hat{\pi} = (\hat{\pi}_1, \hat{\pi}_2, \ldots, \hat{\pi}_N)^{\mathrm{T}}$ and $b = (1, 0, \ldots, 0)^{\mathrm{T}}$. The matrix A has N pivots and is therefore nonsingular, which guarantees a unique quasistationary steady state. The steady state can be expressed as

$$\hat{\pi} = A^{-1} b. \tag{27}$$

The calculation of A^{-1} can become tedious as N becomes large. However, the limiting behavior of the distribution $\hat{\pi}$ can be observed using a state space sufficiently larger than the deterministic steady state, as illustrated by numerical examples. Figure 5 shows the quasistationary distribution of the reaction using the same parameter values as Fig. 3, with state space $N = 300$.

The mean of this distribution can then be expressed as

$$E\{X\} = \sum_{i=1}^{N} i\hat{\pi}_i = cA^{-1}b, \tag{28}$$

where c is the $1 \times N$ vector $c = (1, 2, \ldots, N)$.

As N becomes large, the distribution $\hat{\pi}$ approaches a discrete normal distribution. The mean of the distribution, n^*, is very close to the predicted nonzero deterministic steady state x^*. However, even for very large N, the mean is always slightly lower than x^*. The variance can be approximated by the expression (Nasell, 2001):

$$\sigma^2 = \frac{\lambda_{n^*}}{\mu'_{n^*} - \lambda'_{n^*}}, \tag{29}$$

where μ'_n and λ'_n are the derivatives of μ_n and λ_n with respect to n, respectively.

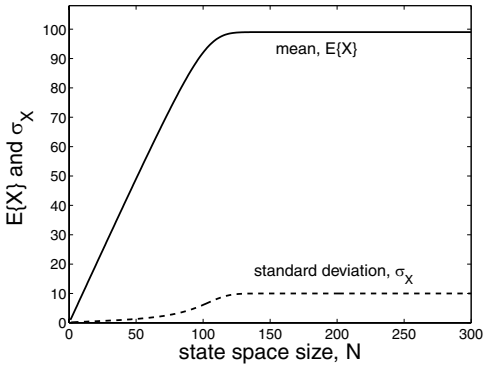

Fig. 6 Mean and variance of the quasistationary steady state for the same parameter values as Fig. 3.

This approximately normal behavior occurs only when the state space N is sufficiently larger than the deterministic mean. To quantify this, we calculate how far the normal distribution stretches to the right, up to a certain tolerance. If we consider the end of the distribution to occur when $P(X = n) < \delta$, then the minimum size N required is

$$N = n^* + \sqrt{-2\sigma^2 \ln(\delta\sigma\sqrt{2\pi})}. \tag{30}$$

Using the value $\delta = 10^{-5}$ with the data from Fig. 5, we require that the state space N be at least 140 to get an approximately normal distribution. From Fig. 5, we see that $N = 140$ would be a sufficiently large interval to contain the bulk of the distribution.

The cut-off value of N given in Eq. (30) also serves as a good truncation value in calculating the infinite sum associated with the time to extinction, τ_1. The components $\hat{\pi}_i$ of the quasistationary steady state defined in Eq. (24) are actually the terms of the sum in τ_i (see Eq. (20)) scaled by $\hat{\pi}_1$. Thus, Fig. 5 also gives us an idea of how quickly the infinite sum will converge, and the value N is a reasonable number of terms to use.

Figure 6 illustrates the behavior of the mean and standard deviation for increasing values of N, using the same parameters before, $k_1 = k_{-1} = k_2 = 0.55$ and $n_a = 100$. Again, the deterministic steady state is $x^* = 99$, and both the mean and variance become relatively constant as the state space size surpasses the deterministic steady state.

3.3. First passage time to quasistationary state

We have shown that the expected time to extinction for a system starting with one molecule is a very large number. However, given that extinction has not occurred, we have shown the system exhibits a quasistationary steady state. For comparison, we now consider how long it takes the system to reach this state, starting with one molecule.

In a birth and death process, the time it takes for the system to go from one state to another is called the first passage time. The expectation of the first passage time from state a to state b can be expressed as $E(T_{b,a})$. It can be calculated by adding up the expected times for each one-state jump:

$$E(T_{b,a}) = E(T_{a+1,a}) + E(T_{a+2,a+1}) + \cdots + E(T_{b,b-1}). \tag{31}$$

To estimate the expected first passage time (from the initial condition of 1 molecule) to the quasistationary steady state, we look at $E(T_{x^*,1})$ where x^* is the mean of the quasistationary steady state distribution, which can be calculated by Eq. (28). The mean of this distribution is rounded off to the nearest integer so that

$$E(T_{x^*,1}) = \sum_{i=2}^{x^*} E(T_{i,i-1}). \tag{32}$$

The interevent time for a birth and death process is exponential with a mean of $\frac{1}{\lambda_i+\mu_i}$ (Allen, 2003), so that the probability of going from i to $i+1$ is $\frac{\lambda_i}{\lambda_i+\mu_i}$ and the probability of going from i to $i-1$ is $\frac{\mu_i}{\lambda_i+\mu_i}$. This gives the recursive relation

$$E(T_{i+1,i}) = \frac{1}{\lambda_i} + \frac{\mu_i}{\lambda_i} E(T_{i,i-1}), \tag{33}$$

which can be used in Eq. (32) to calculate the expected time for the number of molecules to reach the quasistationary steady state.

Using the same parameter values as in the previous figures, we have that

$$E(T_{99,1}) = .1309.$$

Comparing this to the expected time to extinction, which is $O(10^{39})$, we know the quasistationary steady state will be reached almost immediately. Using Eq. (5) from the deterministic model, we also have

$$t(.073, 1) = .1309.$$

The amount of time it takes the stochastic model to reach its quasistationary steady state is about the same amount of time it takes the deterministic model to reach 92.7% of its steady state, starting from the same initial condition (one molecule).

3.4. Spectral analysis of the generator matrix

At the beginning of this section, we calculated the first passage time from $n = 1$ to $n = 0$, referred to as the time to extinction. Because 0 is an absorbing (steady) state, once it is reached, the system remains in that state. However, in general the first passage time only tells us how long it takes to reach a state, not how long the system remains in that state. Through a spectral analysis of the generator matrix

Q (see Eq. 14), we will show that in addition to being reached quickly, the quasistationary state is the dominant state over a long but finite time scale.

The stochastic formulation of our original problem (i.e. before the quasistationary analysis) is

$$\frac{\mathrm{d}p}{\mathrm{d}t} = Qp(t), \tag{34}$$

where Q is the generator matrix. If we include the initial condition $p(0)$, the exact solution can be written as

$$p(t) = p(0)\exp(Qt), \tag{35}$$

where $\exp(Qt)$ refers to the matrix exponential,

$$\exp(Qt) = 1 + (Qt) + \frac{(Qt)^2}{2!} + \frac{(Qt)^3}{3!} + \cdots. \tag{36}$$

This solution can also be expressed in terms of a linear combination of the eigenvectors:

$$p(t) = c_1 \mathrm{e}^{\lambda_1 t} v_1 + c_2 \mathrm{e}^{\lambda_2 t} v_2 + \cdots, \tag{37}$$

where the λ_i are the eigenvalues of Q with corresponding eigenvectors v_i, and the c_i are determined by the initial condition.

Because all the entries of Q have the property that $0 \leq Q_{ij} \leq 1$ and $\Sigma_{i=1}^{\infty} Q_{ij} = 0$, we know from Chung (1967) that the largest eigenvalue of Q is 0, and that it is unique. All other eigenvalues are negative and real. For our problem, since the birth rate $\lambda_0 = 0$ (not to be confused with the eigenvalues λ_i), the eigenvector corresponding to the zero eigenvalue is $v_1 = (1, 0, 0, 0, \ldots)$. Since this is the only nonnegative eigenvalue, this eigenvector is the only one which remains in the limit $t \to \infty$. This is also the steady-state vector found in Eq. (19).

If we again truncate the problem so that Q has finite dimension, we can calculate the eigenvectors and eigenvalues for Q using the same parameter values as before. A list of the first few largest eigenvalues can be found in Table 1. Note that the

Table 1 List of the first few largest eigenvalues of Q, using the same parameters as in Fig. 5

Rank	Value	Rank	Value
1	0	8	−152.5511
2	-8.6413×10^{-12}	9	−197.7168
3	−53.3026	10	−198.2536
5	−104.2117	11	−238.0277
7	−152.5036	12	−243.1988

Note that double eigenvalues are not listed twice, making the rank row discontinuous.

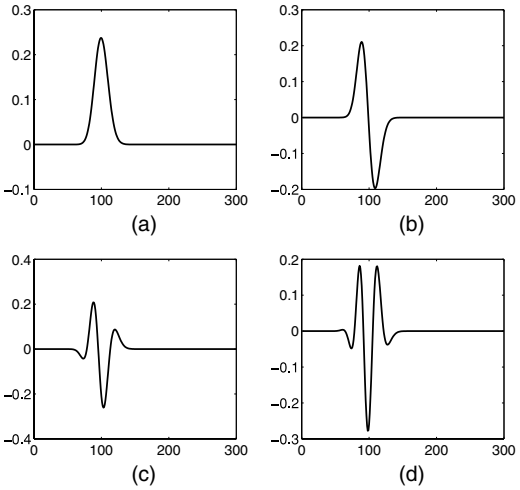

Fig. 7 Eigenvectors corresponding to eigenvalues: (a) -8.6413×10^{-12}, (b) -53.3026, (c) -152.5036, (d) -243.1988.

second largest eigenvalue is many orders of magnitude larger than the third. This implies that after a long but finite time, only the first and second eigenvectors will persist. Figure 7 displays the eigenvectors for some of the eigenvalues in Table 1. The eigenvectors have an interesting sign-switching property which is discussed in Courant and Hilbert (1952). Most importantly, note the striking similarity of the second eigenvector to the quasistationary steady-state in Fig. 5.

By calculating first passage times, we were able to show that the system will reach the mean of the quasistationary steady state almost instantly, while the time to extinction is exponentially large. Also, through an analysis of the spectrum (eigenvalues) and eigenvectors of the generator matrix Q, we have shown that over a long but finite time period, the system remains in the quasistationary steady state. However, as time continues to infinity, the influence of this state will slowly decline, leaving only the stationary steady-state of extinction.

4. Open systems, irreversibility and the origin of complexity

We now turn our attention to the significance of the assumptions made in the simple model (1). We note that the reaction $X \rightarrow C$ is assumed to be irreversible. This can be accomplished in an open biochemical system in which the C is constantly removed from the system while A is constantly supplied to the system. This turns out to be essential for the complex behavior, that is, the disagreement between the deterministic and stochastic models.

4.1. The uniqueness and global stability of deterministic equilibrium steady state

If a biochemical reaction system is in a closed vessel, then the long-time steady state is a chemical equilibrium with zero flux in each and every reaction (Qian and

Reluga, 2005). It is also important to note that in such a system, no reaction can be irreversible, since no matter how small the k_{-2} is in the reaction

$$X \underset{k_{-2}}{\overset{k_2}{\rightleftharpoons}} C,$$

the concentration of C ultimately will build up such that $k_{-2}[C]^{\text{eq}} = k_2[X]^{\text{eq}}$, where $[X]^{\text{eq}}$ and $[C]^{\text{eq}}$ are equilibrium concentrations for the species X and C, respectively.

One of the important characteristics of a closed biochemical reaction system is that it has a unique, globally stable equilibrium point (fixed point) for the ODE system based on the Law of Mass Action. It is also interesting to note that the equilibrium steady-state distribution from the stochastic model has a unique maximum which coincides with the fixed point. Using the reaction in Eq. (1) as an example, including the k_{-2} reaction, the equilibrium concentrations satisfy

$$\frac{[X]^{\text{eq}}}{[A]^{\text{eq}}} = \frac{k_1}{k_{-1}}, \quad \frac{[C]^{\text{eq}}}{[X]^{\text{eq}}} = \frac{k_2}{k_{-2}}, \tag{38}$$

and the total number of molecules in the system $[X] + [A] + [C]$ is a constant in the closed system. It is easy then to show that

$$L([X], [A], [C]) \equiv [X] \ln \frac{[X]}{[X]^{\text{eq}}} + [A] \ln \frac{[A]}{[A]^{\text{eq}}} + [C] \ln \frac{[C]}{[C]^{\text{eq}}}, \tag{39}$$

is a global Lyapunov function for the ODE system.

The concepts of equilibrium and nonequilibrium steady states are chemical concepts rather than mathematical ones. Briefly, an equilibrium steady state (ESS) is one in which there is no reaction flux. All reactions are balanced between their forward and backward reactions, also known as detailed balance. This implies that one can not model a system with equilibrium steady state with irreversible reactions. It can also be shown that any closed system necessarily tends to an ESS. In the contrary, if a system is coupled to its environment, then it tends to a nonequilibrium steady state (NESS) with fluxes in reactions, balanced through sources and sinks.

For a closed system, both the deterministic model with Law of Mass Action and the stochastic model based on Gillespie's approach will yield the same, stable steady state. The possibility of divergence between the steady state of the ODE system and that of stochastic model is due to multiple nonnegative fixed points.

5. Conclusion

Through a thorough analysis of the logistic stochastic model, we find that both deterministic and stochastic methods of modeling the autocatalytic reaction (1) predict a nonzero stationary concentration after a short amount of time. However, when considering the behavior as time becomes very large, the two models begin

to disagree. The deterministic model never moves from its nonzero steady state, whereas the stochastic model predicts eventual extinction of the X molecule.

The relationship between these two models was established by Kurtz in a general theory (Kurtz, 1971, 1972) in which a stochastic Markov chain model for a general chemical reaction is studied alongside its deterministic counterpart. The solution to the master equations is denoted by $X^V(t)$, where V is the volume of the system, and the initial condition (in the thermodynamic limit) is

$$\lim_{V\to\infty} V^{-1}X^V(0) = x_0.$$

The solution to the initial value problem of the corresponding ODE model is denoted as $X(t, x_0)$. Kurtz has shown that the relationship between these two solutions is

$$\lim_{V\to\infty} P\{\sup_{s\le t} V^{-1}X^V(s) - X(s, x_0) > \epsilon\} = 0$$

for every t and $\epsilon > 0$.

The reaction studied in this paper illustrates the subtlety of this statement. Our analysis of the two models by solving for the steady states made the implicit assumption that $\lim t \to \infty$ had been taken. However, the conclusion of Kurtz's paper does not allow us to exchange this limit with $\lim V \to \infty$ (i.e. $N \to \infty$, where the concentration (N/V) is held constant), since the solutions were proven to converge pointwise but not uniformly. Therefore, when looking at the steady state solutions of both equations we cannot expect them to agree. This work seems to suggest the breakdown of uniform convergence near the unstable fixed point of the ODE model.

This leads us to the question of what the long-term behavior of such a reaction actually is. In Section 4, it was shown that an open system will tend toward a nonequilibrium steady state, such as the quasistationary steady state in our example. Even in this balanced state, the master equations allow for a slow extinction of the X molecule because there is still a small probability of the second reaction occurring. Since the deterministic model does not account for random fluctuations in the system, it ignores this possibility after a long time and therefore excludes the long-term behavior shown in the stochastic model.

Although the master equations are a more difficult and time consuming alternative to traditional ODE models of chemical reactions, they provide a richer characterization of the systems dynamics. "The stochastic model is not an alternative to the deterministic kinetics, it is a more complete kinetic description which is capable of modeling reactions with and without fluctuations" (Qian et al., 2002).

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