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Global asymptotic stability of the size distribution in probabilistic models of the cell cycle

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Abstract. Probabilistic models of the cell cycle maintain that cell generation time is a random variable given by some distribution function, and that the probability of cell division per unit time is a function only of cell age (and not, for instance, of cell size). Given the probability density, $f(t)$, for time spent in the random compartment of the cell cycle, we derive a recursion relation for $\psi_n(x)$, the probability density for cell size at birth in a sample of cells in generation n. For the case of exponential growth of cells, the recursion relation has no steady-state solution. For the case of linear cell growth, we show that there exists a unique, globally asymptotically stable, steady-state birth size distribution, $\psi_*(x)$. For the special case of the transition probability model, we display $\psi_*(x)$ explicitly.

Key words: Cell cycle — size distribution — generation time distribution — transition probability model

I. Introduction

Under the right conditions cells from all types of organisms will begin to proliferate. Before long, cell number is increasing exponentially, $N = N_0 e^{kt}$, and all other extensive properties of the cell culture (such as protein content, DNA content, membrane surface area) are increasing at the same specific growth rate, k. Though the macroscopic variables describing the culture behave deterministically, the properties of individual cells show wide variations around the culture means. For instance, the doubling time for cell number is fixed at $\ln 2/k$, but the generation time (i.e., the time transpiring between birth and division) of individual cells is typically distributed with standard deviation/mean $\simeq 20\%$. Similarly, the average mass of a cell is a well-defined, constant property of the culture, but the "momentary" mass of indivdual cells at a certain stage in the cycle (e.g. birth) is typically

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distributed with standard deviation/mean$\simeq 10\%$. Despite the variability of cell generation time and momentary size, the distributions are constant properties of the culture, and if growth conditions are changed, the culture will rapidly approach new, characteristic distributions of generation time and momentary size. The stability of these distributions is the subject of this paper.

The origin of diversity in generation time and cell size of individual cells has long been attributed to probabilistic events in the cell division cycle. These stochastic events are postulated to be completed with certain probabilities per unit time, and upon completion of a certain number of events (anywhere from one to hundreds) the cell is irrevocably committed to division (after a fixed time lag). In this view the generation time of a cell can be written as $T = T_A + T_B$, where T_A is a random variable, the time necessary to complete the probabilistic events, and T_B is a constant, the fixed time lag. The distribution of generation time is

$$\hat{f}(T) = \begin{cases} 0, & T < T_B \\ f(T - T_B), & T \geqslant T_B \end{cases} \tag{1}$$

where

$$\int_{T_1}^{T_2} f(t)\, dt = \operatorname{Prob}\{T_1 < T_A < T_2\}.$$

The form of $f(t)$ depends on the specific hypotheses concerning the random events in the cell cycle. We shall assume that

$$f(t) > 0, \qquad 0 < t < \infty. \tag{2}$$

This assumption is true of the distributions of T_A commonly seen in the literature (see e.g. Hannsgen et al., 1985).

In Sect. II we derive a recursion relation for distribution of cell mass at birth in a sample of cells in generation n, given the distribution in generation $n-1$. In Sect. III we treat the case of exponential growth, where no nontrivial steady state exists. In Sect. IV we treat the case of linear growth, using a theorem of Lasota and Yorke to prove the existence of a unique, asymptotically stable, limiting size distribution. For the Smith-Martin model we display this distribution explicitly in Sect. V.

II. Recursion relation

We assume that individual cells of size x grow according to the growth law

$$dx/dt = V(x). \tag{3}$$

If $x(0) = x_0 =$ size at $t = 0$, then we denote the solution of (3) by $x = X(t, x_0) =$ size at $t \geqslant 0$. Furthermore, we assume that cells divide exactly in half. Thus, at time $t = T =$ generation time, a "mother" cell of size $X(T, x_0)$ divides into two daughters of equal size $X(T, x_0)/2$.

Suppose we start, at $t = 0$, with a sample of newborn cells of various sizes. The distribution of birth size in this sample is denoted $\psi_0(x)$, where the subscript signifies the zeroth generation of cells. Let us follow these newborn cells until each one divides. The sample of daughter cells at birth defines a new distribution of birth sizes, $\psi_1(x)$, the distribution in the first generation. Continuing in this fashion, we define a sequence of birth size distribution functions, $\psi_n(x)$, $n = 0, 1, 2, \ldots$ By definition of a distribution function, $\int_{u_1}^{u_2} \psi_n(x)\,dx$ is the probability that a cell of the nth generation is born with size between u_1 and $u_2 (u_1 < u_2)$. Obviously,

$$\psi_n(x) \geqslant 0 \text{ for all } x \geqslant 0, \text{ and } \int_0^\infty \psi_n(x)\,dx = 1. \tag{4}$$

We intend to relate $\psi_{n+1}(x)$ to $\psi_n(x)$ and to show that in certain cases $\psi_n(x)$ approaches a unique limit, $\psi_*(x)$, as $n \to \infty$.

Let x_0 = birth size of a cell of the zeroth generation and x_1 = birth size of this cell's daughters in the first generation. Then, if T = generation time of the cell under consideration, $\text{Prob}\{x_1 \geqslant x \mid x_0 = y\} = \text{Prob}\{T \geqslant \tau(x, y)\}$ where $\tau(x, y)$ is given by the condition $X(\tau, y) = 2x$. We assume that $\tau(x, y)$ is sufficiently well-behaved for our purposes in what follows. For the specific examples treated later, $\tau(x, y)$ is an analytic function in both x and y, $x, y \in (0, \infty)$. From (1) and (2),

$$\text{Prob}\{T \geqslant \tau\} = \text{Prob}\{T_A \geqslant \tau - T_B\} = \begin{cases} 1, \text{if } \tau < T_B \\ F(\tau - T_B), \text{if } \tau \geqslant T_B, \end{cases} \tag{5}$$

where

$$F(t) = \int_t^\infty f(s)\,ds. \tag{6}$$

Now

$$\begin{aligned} \text{Prob}\{x_1 \geqslant x\} &= \int_0^\infty \text{Prob}\{x_1 \geqslant x \mid x_0 = y\}\psi_0(y)\,dy \\ &= \int_0^{Y(x)} F(\tau(x, y) - T_B)\psi_0(y)\,dy + \int_{Y(x)}^\infty \psi_0(y)\,dy \end{aligned} \tag{7}$$

where $Y(x)$ is the largest possible size at birth consistent with producing daughters of size x at birth, i.e.,

$$X(T_B, Y(x)) = 2x. \tag{8}$$

Now $\text{Prob}\{x_1 \geqslant x\} = \int_x^\infty \psi_1(y)\,dy$, so

$$\psi_1(x) = \int_0^{Y(x)} f(\tau(x, y) - T_B)\frac{\partial}{\partial x}\tau(x, y)\psi_0(y)\,dy. \tag{9}$$

In general,

$$\psi_{n+1}(x) = \int_0^\infty K(x, y)\psi_n(y)\,dy \tag{10}$$

where

$$K(x, y) = \begin{cases} 0 \text{ if } y > Y(x), \\ f(\tau(x, y) - T_B)\partial\tau/\partial x, \text{ if } 0 < y \leqslant Y(x). \end{cases} \tag{11}$$

The kernel, $K(x, y)$, of the integral equation (10) is positive semi-definite and norm-preserving, i.e. $\int_0^\infty \psi_{n+1}(x)\,dx = \int_0^\infty \psi_n(y)\,dy$, because

$$\int_0^\infty K(x, y)\,dx = \int_{X(T_B,y)/2}^\infty f(\tau(x, y) - T_B)\,\partial\tau/\partial x\,dx$$
$$= \int_{T_B}^\infty f(\tau - T_B)\,d\tau = \int_0^\infty f(s)\,ds = 1.$$

Thus, $\psi_{n+1}(x)$ will be a probability density, satisfying (4), if $\psi_n(x)$ is a probability density.

III. Exponential growth

If $V(x) = kx$ in (3), then $X(t, y) = y\,e^{kt}$, $\tau(x, y) = k^{-1}\ln(2x/y)$, and $Y(x) = 2x\,e^{-kT_B}$. Thus

$$K(x, y) = \begin{cases} 0, \text{if } y > 2x\,e^{-kT_B} \\ f(k^{-1}\ln(2x/y) - T_B)/kx, \text{if } 0 < y < 2x\,e^{-kT_B}, \end{cases} \tag{12}$$

and

$$\psi_{n+1}(x) = \frac{1}{kx}\int_0^{2xe^{-kT_B}} f\left(\frac{1}{k}\ln\left(\frac{2x}{y}\right) - T_B\right)\psi_n(y)\,dy. \tag{13}$$

By the line of reasoning in Hannsgen, Tyson and Watson (1985), it can be shown that (13) has no steady state solution $\psi_{n+1}(x) = \psi_n(x) \equiv \psi_*(x)$ satisfying (4). By different methods Trucco and Bell (1970) have proved more generally that, for growth laws of the type $X(t, x_0) = x_0 g(t)$ with $g(t) > 0$ and $g(0) = 1$, there does not exist an asymptotically stable birth-size distribution for probabilistic (age-dependent) models of the cell cycle; on the contrary, as the population proceeds from generation to generation the variance of the size distribution increases without bound.

IV. Linear growth

If $V(x) = \tilde{k}$ in (3), then $X(t, y) = y + \tilde{k}t$, $\tau(x, y) = (2x - y)/\tilde{k}$, and $Y(x) = 2x - \tilde{k}T_B$. Thus,

$$K(x, y) = \begin{cases} 0, \text{if } y > 2x - \tilde{k}T_B \\ (2/\tilde{k})f((2x - y - \tilde{k}T_B)/\tilde{k}), \text{if } 0 < y < 2x - \tilde{k}T_B \end{cases} \tag{14}$$

and

$$\psi_{n+1}(x) = (2/\tilde{k})\int_0^{2x-\tilde{k}T_B} f((2x - y - \tilde{k}T_B)/\tilde{k})\psi_n(y)\,dy. \tag{15}$$

We wish to show that the size distribution $\psi_n(x)$ converges to some unique, steady state distribution $\psi_*(x)$ as $n \to \infty$. To this end we introduce the following definitions and theorems (Lasota and Mackey, 1984). Let D be the set of all real-valued functions on $[0, \infty)$ which satisfy

$$\psi(x) \geq 0 \text{ and } \int_0^\infty \psi(x)\, dx = 1. \tag{16}$$

Let $K(x, y)$ be a function defined on $x, y \in [0, \infty)$ satisfying

$$K(x, y) \geq 0, \text{ and } \int_0^\infty K(x, y)\, dx = 1 \text{ for all } y. \tag{17}$$

Let the operator P: $L^1 \to L^1$ be defined by

$$P\psi(x) = \int_0^\infty K(x, y)\psi(y)\, dy. \tag{18}$$

A function h will be called a *lower bound function* of P if, for every $\psi \in D$, there exists an integer $n_0(\psi)$ such that

$$P^n\psi(x) \geq h(x),\ n \geq n_0(\psi). \tag{19}$$

$h(x)$ is called *nontrivial* if

$$h(x) \geq 0 \text{ and } \int_0^\infty h(x)\, dx > 0. \tag{20}$$

Theorem 1. (Lasota and Yorke, 1982; see Lasota and Mackey, 1984). *If, for a kernel* $K(x, y)$ *satisfying* (17), *there is a nontrivial lower bound function for the operator* (18), *then the equation*

$$\psi(x) = \int_0^\infty K(x, y)\psi(y)\, dy \tag{21}$$

has a unique solution $\psi_* \in D$, *and moreover for any other* $\psi \in D$

$$\lim_{n\to\infty} \int_0^\infty |P^n\psi(x) - \psi_*(x)|\, dx = 0. \tag{22}$$

Theorem 2. (Lasota and Mackey, 1984). *Let* $K(x, y)$ *satisfy* (17). *If*

$$\int_0^\infty xK(x, y)\, dx \leq \gamma y + \delta,\ y \geq 0, \tag{23}$$

for some nonnegative constants γ *and* δ, $\gamma < 1$, *and if*

$$\int_0^\infty \inf_{y\in[0, a]} K(x, y)\, dx > 0, \tag{24}$$

for some $a > \delta/(1-\gamma)$, *then there exists a nontrivial lower bound function for the operator* (18).

Thus, to prove existence and global asymptotic stability of the steady-state solution, $\psi_*(x)$, of (15), we must verify (23) and (24). First,

$$\int_0^\infty xK(x,y)\,dx = \int_{(y+\tilde{k}T_B)/2}^\infty (2x/\tilde{k})f((2x-y-\tilde{k}T_B)/\tilde{k})\,dx$$
$$=(y/2)+(\tilde{k}/2)\left(T_B+\int_0^\infty tf(t)\,dt\right)$$
$$=(y+\tilde{k}\langle T\rangle)/2,$$

where $\langle T\rangle$ = mean generation time. Thus (28) holds with $\gamma=1/2$, $\delta=\tilde{k}\langle T\rangle/2$. Next, choose $a=\tilde{k}(\langle T\rangle+\varepsilon)$ for some $\varepsilon>0$. For $x<\tilde{k}(\langle T\rangle+T_B+\varepsilon)/2$, $\inf_{y\in[0,a]}K(x,y)=0$, but for $x>\tilde{k}(\langle T\rangle+T_B+\varepsilon)/2$,

$$\inf_{y\in[0,a]}K(x,y)=\inf_{y\in[0,a]}(2/\tilde{k})f((2x-\tilde{k}T_B-y)/\tilde{k})>0,$$

where the inequality follows from our assumption (2) concerning $f(t)$. This implies (24). It should be obvious that the upper limit on t in (2) need not be ∞. If $f(t)=0$ for $t>T_A^{\max}$, then (29) still holds as long as $T_A^{\max}>T_A^{\text{ave}}+T_B$, where $T_A^{\text{ave}}=\int_0^\infty tf(t)\,dt$.

V. Transition probability model

We have demonstrated the existence and global asymptotic stability of a steady-state birth-size distribution function for probabilistic models with linear cell growth. For some special choices of $f(t)$ we can write down the solution, $\psi_*(x)$, explicitly. For example, Smith and Martin (1973) proposed that

$$f(t)=p\,e^{-pt} \tag{25}$$

where p is some constant (the probability per unit time that a cell in A-phase will make a transition to B-phase). This assumption is usually referred to as the "transition probability model."

Without any loss of generality, we can take $T_B=1$ and $\tilde{k}=1$. In this case, the steady-state solution of Eq. (15), with $f(t)$ given by (25), satisfies

$$\psi_*(x)=2p\,e^{-p(2x-1)}\int_1^{2x-1} e^{py}\psi_*(y)\,dy. \tag{26}$$

The lower limit in Eq. (26) is $y=1$ because, in the steady state, no cells are born with size less than 1. To see why, suppose there were a cell, taken from a steady-state culture, with birth size $u_0<1$. This cell would give rise to daughters of birth size $u_1\geq(u_0+1)/2>u_0$. Since any cell of birth size <1 would give rise to daughters larger than itself, such a cell could not have been taken from a steady-state culture.

A nontrivial, normalized solution of (26) is (Hannsgen, et al. 1985)

$$\psi_*(x)=(p/N)\sum_{n=0}^\infty(-1)^n c_n 2^{n+1}\,e^{-2^{n+1}p(x-1)},\ x\geq 1 \tag{27}$$

where

$$c_0 = 1,\ c_n = [1 \cdot 3 \cdot 7 \cdots (2^n - 1)]^{-1} (n = 1, 2, 3, \ldots) \tag{28}$$

and

$$N = \sum_{n=0}^{\infty} (-1)^n c_n \simeq 0.288788. \tag{29}$$

Equation (27) can be verified by substituting into (26), integrating term-by-term, and using the identity (Hannsgen, et al. 1985, Appendix)

$$\sum_{n=1}^{\infty} (-2)^n c_n = -1. \tag{30}$$

From the treatment in Hannsgen, et al. (1985) one can, in a similar fashion, obtain an explicit solution for $\psi_*(x)$ if $f(t)$ is given by the more realistic two-transitions model of Brooks, Bennett and Smith (1980). We shall not display the solution here.

The analysis presented here assumes that the fundamentally discrete histogram of cell birth sizes in a finite population can be approximated by a continuous probability density $\psi_*(x)$. To see how our analytical formula for $\psi_*(x)$ might compare with a birth-size histogram from a finite cell population, we have simulated an ideal population by Monte-Carlo methods. In particular, 500 cells were allowed to grow and divide according to our assumptions of linear growth and exponentially distributed waiting times, T_A. At division one daughter was removed from the culture, so that there were always 500 cells in each generation.

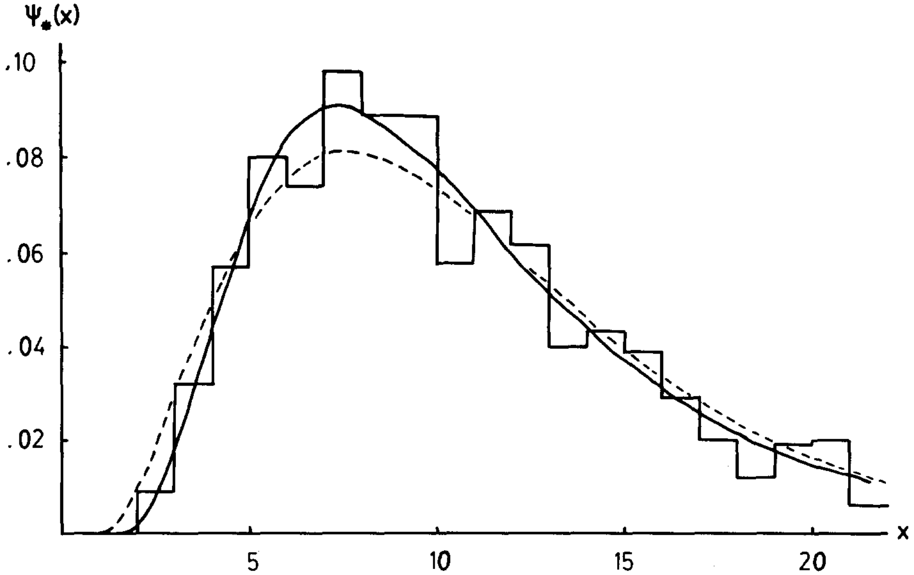

Fig. 1. Birth size distribution for the transition probability model with linear cell growth. Monte-Carlo simulations (*histogram*), analytic solution (*solid line*), and Pearson Type III distribution (*dashed line*) with the same mean and variance as the analytic solution. Parameter values: $T_B = 1$, $\tilde{k} = 1$, $p = 0.1$

From an initial arbitrary distribution of birth sizes of the 500 cells, the culture was simulated for 20 generations by which time the steady-state size distribution was attained. In repeated simulations, the population always approached (within 5-10 generations) the size distribution predicted by (27), as judged by the observed and expected mean and variance of the distribution. A histogram of birth sizes of 500 cells in generation 20 from a typical simulation is plotted in Fig. 1, in comparison with the analytical probability density function (27) and an approximation to the analytical solution provided by a Pearson Type III distribution (Hannsgen, et al. 1985). Figure 1 illustrates the kind of agreement one can expect to find between theoretical density functions and experimental histograms in the best case where (1) the model accurately reflects the laws of cell growth and division and (2) a large number of cells are used to construct the histogram.

References

1. Brooks, R. F., Bennett, D. C., Smith, J. A.: Mammalian cell cycles need two random transitions. Cell **19**, 493-504 (1980)
2. Hannsgen, K. B., Tyson, J. J., Watson, L. T.: Steady-state size distributions in probabilistic models of the cell division cycle. SIAM J. Appl. Math., in press (1985)
3. Lasota, A., Mackey, M. C.: Globally asymptotic properties of proliferating cell populations. **19**, 43-62 (1984)
4. Lasota, A., Yorke, J. A.: Exact dynamical systems and the Frobenius-Perron operator. Trans. Am. Math. Soc. **273**, 375-384 (1982)
5. Smith, J. A., Martin, L.: Do cells cycle? Proc. Natn. Acad. Sci. USA **70**, 1263-1267 (1973).
6. Trucco, E., Bell, G. I.: A note on the dispersionless growth law for single cells. Bull. Math. Biophys. **32**, 475-483 (1970)

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