STEADY-STATE SIZE DISTRIBUTIONS IN PROBABILISTIC MODELS OF THE CELL DIVISION CYCLE

by

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Abstract. A model for the steady-state size distribution in an exponentially growing population of single cells is derived and studied. The model incorporates a general growth law for individual cells and a probability density for interdivision times. A uniqueness theorem is proved, and it is shown that no solution exists when individual cells grow exponentially. For linear growth, infinite series solutions are found in two specific cases. Statistical data are obtained for these solutions, and comparisons are made with the results of some numerical simulations and with a known experimental result.

1. Introduction. We study a mathematical model for the steady-state size distribution in an exponentially growing population of single cells. The model incorporates a general growth law for individual cells and a probability distribution for interdivision times.

Mathematically, the basic model is an integral equation. In certain special cases it reduces to a functional differential equation of advanced type which may be of independent interest.

We derive the model in Sec. 2 and show that it admits at most one solution. When individual cells grow exponentially, there is no solution (Sec. 3); but in Sec. 4 we obtain infinite series solutions in two specific cases where cells grow linearly. In Sec. 5 we obtain statistical data for these solutions. Finally, in Sec. 6, we compare our analytical solutions with the results of some numerical simulations and with an experimental result of Schaechter, Maaløe, and Kjeldgaard [12]. A connection with some identities for basic hypergeometric series is noted in the Appendix.

2. The equation. Microorganisms (and some cells isolated from higher plants and animals, including man) are frequently grown as large cultures of single

cells. Under suitable nutritional conditions the cells grow continuously and divide asynchronously. The population as a whole increases exponentially in all its macroscopic properties: $G(t) = G_o e^{kt}$, where G = cell number, protein content, RNA content, DNA content, etc., and k = specific growth rate. Under these conditions the culture is said to be in a steady-state of balanced growth. Not only is the average cell size (e.g., protein/cell number) constant in time, but also there exists an asymptotically stable, time-independent distribution of cell sizes in the culture. Any model for the control of growth and division in a cell culture must account for the existence of a stable steady-state distribution of cell sizes.

In such cultures of simple cells, cell generation times (T = time from one division to the next) are not constant but follow a skewed distribution. Over the years several 'probabilistic' models of progress through the cell cycle have been proposed to explain this skewed distribution of cell generation times. In the earliest model, Rahn [10] proposed that a cell divides as soon as some fixed number g of events have occured; the events occur independently in any order and with the same constant probability p per unit time. These assumptions imply that e^{-pT} follows a beta distribution:

$$f(T) = gpe^{-pT}(1-e^{-pT})^{g-1},$$

where f(T)dT is the probability that a cell's generation time lies between T and T+dT. Later Kendall [6] proposed that the g events must occur in a specified sequence, in which case pT follows a gamma distribution:

$$f(T) = pe^{-pT}(pT)^{g-1}/(g-1)!.$$

In the early 1960's, Kubitschek [7] showed that, for the most part, cell generation times follow closely a reciprocal-normal distribution, and he pointed out that this would result if cell growth rates (1/T) are normally distributed. He attributed the normal distribution of growth rates to a random distribution of rate-limiting substances between sister cells at division. A fourth model, which has received much attention recently, is the 'transition probability' model of Smith and Martin [13]. They pointed out that a major component of the variability of cell generation times could be attributed to a single random event occuring with constant probability p per unit time. In their simplest model, cells must wait for completion of this random event (called the A-phase of the cell cycle, of variable duration T_A); then cells enter a deterministic phase leading to division (called the B-phase of the cell cycle, of constant duration T_B). In this model, the probability density for cell generation times is

$$f(T) = pe^{-p(T-T_B)}, \quad T \geq T_B.$$

This function is glaringly inconsistent with observed distributions of cell generation times, so the simple model has been extended by Brooks, Bennett and Smith [4] to include two random events in a fixed order, in which case

$$f(T) = pq[e^{-p(T-T_B)} - e^{-q(T-T_B)}]/(q-p), \quad T \geq T_B,$$

where p and q are the transition probabilities for the two random events.

In light of this variety of opinions among cell biologists about the distribution of cell generation times, we shall initially adopt a general point of view. We assume only that the total cell generation time is given by a random variable T with cumulative distribution function

(2.1) $\Pr\{T>t\} = F(t) = \int_t^\infty f(s)ds,$

where $f(t)$, the probability density for cell generation time, is nonnegative and piecewise continuous on each finite subinterval of $[0,\infty)$ with $\int_0^\infty f(t)dt = 1$. These assumptions cover all reasonable 'probabilistic' models of progress through the cell cycle. They are not consistent with 'size control' models, in which there is feedback between cell size and the timing of cell division. We discuss size control models in a separate publication [15].

Notice in (2.1) that our 'cumulative' distribution function, $F(t)$, decreases monotonically from 1 to 0 as t increases from 0 to ∞. This convention is opposite to the one normally adopted for cumulative distributions, but it conforms with the biological literature where $F(t)$, as we define it, is known as the 'alpha-curve' [13].

At time T after its birth, a cell divides into exactly r daughters of equal size. The case r=2 corresponds to binary fission. The case $r=2^n$ corresponds to division of a mother cell into 2^n daughters, which is not uncommon among microorganisms.

We must specify how individual cells grow. Experimental observation is inconclusive [8], so we consider a general growth law; later we shall consider exponential and linear growth in detail. We assume that a cell of mass $x_1 > 0$ at time t_1 will reach mass $x = M(t-t_1,x_1)$ if it grows to time $t > t_1$. Here $M(t,x)$ is a nonnegative, continuously differentiable function on $\{t,x \geq 0\}$ with $M > 0$, $\partial M/\partial t > 0$, $\partial M/\partial x \geq M_o > 0$ $(t > 0,\ x > 0)$,

(2.2) $M(0,x) = x \quad (x \geq 0),$

and $M(t_1+t_2,x) = M(t_1,M(t_2,x))$. Then define $m(t,x)$ for $t < 0$, $x \geq 0$ by

$m(t,x) = 0$, if $M(-t,y) > x$ for all $y > 0$,

$M(-t,m(t,x)) = x$, otherwise.

Then m represents the minimum size required for a newborn cell at time t to grow to size at least x at time 0. Note that $0 \leq m(t,x) < x$. The implicit function theorem shows that

(2.3) $$0 < \frac{\partial m}{\partial x}(t,x) \leq \frac{1}{M_o},\quad 0 < \frac{\partial m}{\partial t}(t,x) = \frac{\partial M/\partial t}{\partial M/\partial x}\Big|_{(-t,m(t,x))}$$

and both partials are continuous, provided we restrict t and x to the set where $m > 0$. Since $M(t,x)$ is finite,

(2.4) $$m(t,x) \to \infty \qquad (x \to \infty,\ -\infty < t < 0).$$

PROPOSITION 2.1. $\lim_{t\to-\infty} m(t,x) = 0$.

Proof. Since (2.3) holds, m increases with t. Suppose $0 < \mu < \lim_{t\to-\infty} m(t,x)$. Then $\tilde{\mu} = \lim_{t\to\infty} M(t,\mu) < \lim_{t\to\infty} M(t,m(-t,x)) = x < \infty$. Thus, $M(t,\mu)$ approaches a finite limit, $\tilde{\mu}$, from below as $t \to \infty$. Furthermore, $M(1,\tilde{\mu}) > \tilde{\mu}$, so by continuity of M, $M(1,\lambda) > \tilde{\mu}$ when $\tilde{\mu}-\lambda$ is sufficiently small. For large t, $\tilde{\mu} - M(t,\mu)$ is arbitrarily small, so $\tilde{\mu} < M(1,M(t,\mu)) = M(t+1,\mu)$. Thus $M(t,\mu) > \tilde{\mu}$ for t large, which contradicts our previous conclusion. Thus, no such μ exists, and the proof is complete.

Note finally that m(t,x) is continuous in x for all x. We need only check points (t,x_o) where $M(-t,0) = x_o = x_o(t)$. At such a point, $M(-t,y) > x_o$ if $y > 0$, so $m(t,x) = 0$ $(0 \leq x \leq x_o)$. On the other hand

$$M(-t, \lim_{x\to x_o+} m(t,x)) = \lim_{x\to x_o+} M(-t,m(t,x))$$

$$= x_o = M(-t,0).$$

Since $\partial M/\partial x > 0$ $(x > 0)$, we must have $\lim_{x \to x_0+} m(t,x) = 0$.

What is the distribution of cell sizes in such a model? First, we must be more specific about what we mean by a cell-size distribution. There are many different ways to express the distribution of sizes in a population of cells, depending on when size is measured and the sample from which the random selection of cells is made. We define $\bar{\Phi}$ to be the cumulative distribution function for size at division in a sample of dividing cells; thus

(2.5) $\bar{\Phi}(x)N$ = the number of cells with mass greater than x at division in a sample of N dividing cells.

(Notice that our 'cumulative' distribution function decreases monotonically from 1 to 0 as x increases from 0 to ∞.) Similarly, Ψ denotes the cumulative distribution function for size at birth in a sample of newborn cells. Our assumption about the division of cells into r daughters implies

(2.6) $\Psi(x) = \bar{\Phi}(rx)$ $(x \geq 0)$.

We assume that

(2.7) $\bar{\Phi}$ is a nonincreasing function on $[0, \infty)$, with

(2.8) $\bar{\Phi}(0) = 1$,

(2.9) $\lim_{x \to \infty} \bar{\Phi}(x) = 0$

(2.10) $\lim_{y \to x+} \bar{\Phi}(y) = \bar{\Phi}(x)$ $(0 \leq x < \infty)$;

these conditions are consistent with our interpretation of $\bar{\Phi}$ in (2.5).

We can now derive the basic equation for Φ. Suppose we have $N(t) = N_o e^{kt}$ cells at time t. Fix $\tau > 0$, and consider the set $S(\tau)$ of all cells which divide in the time interval $[0,\tau)$. Since the population grows by $N_o(e^{k\tau}-1)$ cells during this time interval, and since each division increases the population by r-1 cells, there are $(r-1)^{-1}N_o(e^{k\tau}-1)$ cells in $S(\tau)$. Then the number $S(x,\tau)$ of cells in $S(\tau)$ which are born before time $t = 0$ and have mass $> x$ at $t = 0$ satisfies

$$S(x,\tau) \leq (r-1)^{-1}N_o(e^{k\tau}-1)\Phi(x) \qquad (0 \leq x < \infty). \tag{2.11}$$

If a cell belongs to $S(\tau)$ but is born during $[0,\tau)$, then its division time is no greater than τ. Since only $r(r-1)^{-1}N_o(e^{k\tau}-1)$ cells are born during $[0,\tau)$, and since (2.1) holds, the number of such cells cannot exceed

$$r(r-1)^{-1}N_o(e^{k\tau}-1)[F(0)-F(\tau)] = o(\tau) \qquad (\tau \to 0+).$$

On the other hand, if a cell in $S(\tau)$ is born before t=0 and has division size greater than $M(\tau,x)$, then it has size greater than x at t=0. We conclude that

$$S(x,\tau) \geq (r-1)^{-1}N_o(e^{k\tau}-1)\Phi(M(\tau,x)) + o(\tau) \qquad (\tau \to 0+,\ 0 \leq x < \infty). \tag{2.12}$$

Now combine (2.11) and (2.12), divide by τ, and let $\tau \to 0+$. Since (2.2) and (2.10) hold, the result is

$$\lim_{\tau \to 0+} S(x,\tau)/\tau = k(r-1)^{-1}N_o\Phi(x). \tag{2.13}$$

Now fix $\tau > 0$ again, and note that the cells counted by $S(x,\tau)$ are born at time $-\sigma < 0$ at a rate of $r(r-1)^{-1}kN_o e^{-k\sigma}\,\Psi(m(-\sigma,x))\int_\sigma^{\sigma+\tau} f(s)ds$ per unit time (σ). Since (2.6) holds,

$$\lim_{\tau \to 0+} S(x,\tau)/\tau = \lim_{\tau \to 0+} \tau^{-1}\int_o^\infty r(r-1)^{-1}kN_o\, e^{-k\sigma}\,\Phi(rm(-\sigma,x))\int_\sigma^{\sigma+\tau} f(s)ds\, d\sigma$$

$$= r(r-1)^{-1}kN_o \int_o^\infty e^{-k\sigma}\, \bar{\phi}(rm(-\sigma,x))\, f(\sigma)d\sigma.$$

By (2.13), this means that

$$\bar{\phi}(x) = r \int_o^\infty e^{-k\sigma}\, \bar{\phi}(rm(-\sigma,x))\, f(\sigma)d\sigma. \tag{2.14}$$

Equation (2.14), subject to (2.7) through (2.10), is the basic mathematical problem to be solved.

While $r=2^n$ in the cell model, one may take r to be any number greater than 1 in the analysis of (2.14).

By (2.8) and (2.14),

$$1 = r \int_o^\infty e^{-k\sigma}\, f(\sigma)d\sigma. \tag{2.15}$$

This is a consistency condition through which f and r determine k uniquely.

Formal differentiation of (2.14) yields

$$\phi(x) = r^2 \int_o^\infty e^{-k\sigma}\, \phi(rm(-\sigma,x))\, \partial m(-\sigma,x)/\partial x\, f(\sigma)d\sigma, \tag{2.16}$$

with $\phi(x) = -\partial\bar{\phi}/\partial x$. (Notice that $\phi(x)$ is the probability density associated with the cumulative distribution $\bar{\phi}(x)$.) By a <u>solution</u> of (2.16), we mean a function ϕ in the class L^1 of Lebesgue integrable functions on $(0,\infty)$ such that (2.16) holds almost everywhere (a.e.). For such a solution, set

$$\bar{\phi}(x) = \int_x^\infty \phi(y)dy. \tag{2.17}$$

Then integration of (2.16), together with a change in the order of integration and a change of variable, shows that $\bar{\phi}$ satisfies (2.14). Moreover, this justifies the differentiation from (2.14) to (2.16) (a.e.), provided $\bar{\phi}$ in (2.14) is absolutely continuous with derivative $-\phi$.

For this absolutely continuous case, our basic problem, (2.14), (2.7)-(2.10), is equivalent to (2.16), subject to

(2.7′) $\phi(x) \geq 0$ a.e.

(2.8′) $\int_0^\infty \phi(x)dx = 1$

(2.9) and (2.10) are automatically satisfied when $\phi \varepsilon L^1$.

For (2.16), (2.8′), we will need the following uniqueness and positivity result.

THEOREM 1. *Let f be as described following (2.1), and suppose*

(2.18) $f(t) > 0$ $(t_1 \leq t < \infty)$, *for some* $t_1 \geq 0$.

Let m be as above, let $r > 1$, *and let k satisfy (2.15). Then there is at most one function* ϕ *in* L^1 *which satisfies (2.16) and (2.8′). Any such* ϕ *satisfies (2.7′).*

Proof. Let $\phi \varepsilon L^1$ satisfy (2.16). By (2.16), (2.15), (2.4), (2.3),

$$\int_0^\infty |\phi(x)|dx = r^2 \int_0^\infty \left| \int_0^\infty e^{-k\sigma}\, \phi(rm(-\sigma,x))\, \partial m(-\sigma,x)/\partial x\, f(\sigma)d\sigma \right| dx$$

$$\leq r^2 \int_0^\infty e^{-k\sigma} \left[\int_0^\infty |\phi(rm(-\sigma,x))|\, \partial m(-\sigma,x)/\partial x\, dx \right] f(\sigma)d\sigma$$

$$= r \int_0^\infty e^{-k\sigma} \left[\int_0^\infty |\phi(y)|dy \right] f(\sigma)d\sigma = \int_0^\infty |\phi(x)|dx.$$

Thus the inequality in the chain must be an equality, so

$$\left| \int_0^\infty e^{-k\sigma}\, \phi(rm(-\sigma,x))\, \partial m(-\sigma,x)/\partial x\, f(\sigma)d\sigma \right|$$

$$= \int_0^\infty e^{-k\sigma} |\phi(rm(-\sigma,x))|\, \partial m(-\sigma,x)/\partial x\, f(\sigma)d\sigma$$

for almost every x. By (2.18), it follows that for almost every x, $\phi(rm(-\sigma,x))$ does not change sign (except for a set of measure 0) on $\{\sigma: t_1 \leq \sigma < \infty\}$. Then by Proposition 2.1, $\phi(y)$ does not change sign for

$$0 = r \lim_{\sigma\to\infty} m(-\sigma,x) < y \leq r\, m(-t_1,x);$$

since $\partial m(t,x)/\partial t$ exists and is continuous (see (2.3)), the exceptional set still has measure zero. From (2.4), we conclude that $\phi(y)$ is of one sign (or zero) almost everywhere on $\{0 < y < \infty\}$. For (2.8') to hold, the sign must be positive, so (2.7') holds. If ϕ_1 and ϕ_2 are solutions of (2.16), (2.8'), then $\tilde{\phi} \equiv \phi_1 - \phi_2$ satisfies (2.16); by the argument above, $\tilde{\phi}$ does not change sign (a.e.). Since both ϕ_1 and ϕ_2 satisfy (2.8'), we must have $\phi_1 = \phi_2$ (a.e.). This proves Theorem 1.

Next we examine some more specific models.

3. The case of exponential growth. Suppose $M(t,x) = xe^{\tilde{k}t}$ $(t,x \geq 0)$ with $\tilde{k} > 0$, so that $m(t,x) = xe^{\tilde{k}t}$ $(-t,x > 0)$.

Then (2.14) becomes

$$(3.1) \qquad \bar{\mathfrak{b}}(x) = r \int_0^\infty e^{-k\sigma}\, \bar{\mathfrak{b}}(rxe^{-\tilde{k}\sigma})\, f(\sigma)d\sigma.$$

THEOREM 2. Problem (3.1), (2.7), (2.8), (2.9), (2.10) has no solution.

Therefore, there does not exist a stable, time-independent size distribution $\bar{\mathfrak{b}}$ for probabilistic models of the cell cycle if cells grow exponentially. We conjecture that the time-dependent distribution flattens and approaches a trivial solution $\bar{\mathfrak{b}}(x) =$ constant of (3.1); that is, as the culture ages, some cells get arbitrarily small and others get arbitrarily large (see Table 1). The only way to avoid this conclusion is to include feedback from cell size to division time in the model. This idea is pursued in [15].

Proof of Theorem 2. We use Mellin transforms. Suppose Φ is a solution. Assume first that Φ is absolutely continuous with derivative $\Phi'(x) = -\phi(x)$ a.e., so that (2.16) holds; that is,

$$\phi(x) = r^2 \int_0^\infty e^{-(k+\tilde{k})\sigma}\, \phi(rxe^{-\tilde{k}\sigma})\, f(\sigma)d\sigma \qquad \text{a.e.} \tag{3.2}$$

Multiply (3.2) by $x^{-i\eta}$ $(-\infty < \eta < \infty)$ and integrate to obtain

$$\begin{aligned} \tilde{\phi}(\eta) &\equiv \int_0^\infty \phi(x)x^{-i\eta}dx \\ &= r^2\int_0^\infty e^{-(k+\tilde{k})\sigma}\, [\int_0^\infty x^{-i\eta}\phi(rxe^{-\tilde{k}\sigma})dx]\, f(\sigma)d\sigma \\ &= r^{i\eta}\, B(\tilde{k}\eta)\, \tilde{\phi}(\eta), \end{aligned} \tag{3.3}$$

where

$$B(\eta) = r \int_0^\infty e^{-k\sigma}\, f(\sigma)\, e^{-i\eta\sigma}\, d\sigma \tag{3.4}$$

is the Fourier transform of $b(\sigma) = re^{-k\sigma}f(\sigma)$ $(0 < \sigma < \infty)$. The analogue of (3.3) for the general case of (3.1) is

$$\Phi^*(\eta) = r^{i\eta}\, B(\tilde{k}\eta)\, \Phi^*(\eta) \qquad (-\infty < \eta < \infty), \tag{3.5}$$

where

$$\Phi^*(\eta) = \int_0^\infty x^{-i\eta}\, d\Phi(x). \tag{3.6}$$

To justify (3.5), choose a sequence $\{\Phi_n\}$ of absolutely continuous, nonincreasing functions with

$$\Phi_n(0) = 1, \quad \lim_{x\to\infty} \Phi_n(x) = 0 \tag{3.7}$$

and $\Phi_n(x) \uparrow \Phi(x)$ $(n \to \infty,\ 0 < x < \infty)$; since Φ is nonincreasing and (2.10) holds, this is possible. By a convergence theorem for probability measures [5],

$$\int_0^\infty g(x)\, d\mathfrak{B}_n(x) \to \int_0^\infty g(x)\, d\mathfrak{B}(x) \qquad (n \to \infty)$$

for every bounded continuous function g on $(0, \infty)$. In particular,

$$\mathfrak{B}^*(\eta) = \lim_{n\to\infty} \mathfrak{B}_n^*(\eta) = -\lim_{n\to\infty} \widetilde{(\mathfrak{B}_n')}(\eta) \qquad (-\infty < \eta < \infty). \tag{3.8}$$

In addition, the functions

$$H_n(x) = r \int_0^\infty e^{-k\sigma}\, \mathfrak{B}_n(rxe^{-\tilde{k}\sigma})\, f(\sigma)d\sigma$$

are absolutely continuous, nonincreasing (in x), and nondecreasing (in n) to the limit

$$H(x) = r \int_0^\infty e^{-k\sigma}\, \mathfrak{B}(rxe^{-\tilde{k}\sigma})\, f(\sigma)d\sigma,$$

with $H_n(0) = H(0) = 1$, $\lim_{x\to\infty} H_n(x) = \lim_{x\to\infty} H(x) = 0$. Thus the convergence theorem for probability measures shows that

$$\lim_{n\to\infty} H_n^*(\eta) = H^*(\eta) = r \int_0^\infty x^{-i\eta}\, d_x\Big[\int_0^\infty e^{-k\sigma}\mathfrak{B}(rxe^{-\tilde{k}\sigma}) f(\sigma)d\sigma\Big]. \tag{3.9}$$

But since H_n is absolutely continuous,

$$H_n^*(\eta) = -r^2\int_0^\infty x^{-i\eta}\Big[\int_0^\infty e^{-(k+\tilde{k})\sigma)}\mathfrak{B}_n'(rxe^{-\tilde{k}\sigma}) f(\sigma)d\sigma\Big]dx,$$

as in the derivation of (2.16), so, as with (3.3),

$$H_n^*(\eta) = -r^{i\eta}\, B(\tilde{k}\eta)\, \tilde{\mathfrak{B}}_n'(\eta) \qquad (-\infty < \eta < \infty). \tag{3.10}$$

Equation (3.1) shows that $\mathfrak{B} = H$, so (3.8), (3.9), and (3.10) yield (3.5).

Now $\mathfrak{B}^*(\eta)$ is continuous and not identically zero. (Note that $\mathfrak{B}^*$ is the Fourier-Stieltjes transform of the nontrivial finite measure $d[\mathfrak{B}(e^t)]$ on $\{-\infty < t < \infty\}$.) Thus $\mathfrak{B}^*(\eta)$ is nonzero on some open interval (η_1, η_2); by (3.5),

(3.11) $\quad B(\tilde{k}\eta) - r^{-i\eta} = 0 \qquad (\eta_1 < \eta < \eta_2).$

But $B(\tilde{k}\eta)-r^{-i\eta}$ is analytic in $\{\text{Im } \eta < 0\}$ and bounded and continuous in $\Pi \equiv \{\text{Im } \eta \leq 0\}$. By a basic result for bounded analytic functions in a disk or half-plane [11], identity (3.11) extends to all of Π, and in particular to the set of all real η. But by the Riemann-Lebesgue Lemma, $\lim_{\eta \to +\infty} B(\tilde{k}\eta) = 0$, while $|r^{-i\eta}| = 1$ $(-\infty < \eta < \infty)$, so we have a contradiction. This proves Theorem 2.

4. The case of linear growth - theory. We have just proven that there does not exist a stable time-independent size distribution for probabilistic models of the cell cycle under the assumption of exponential cell growth. Brooks [3] suggests that the problem lies not with the probabilistic passage of cells through the cycle but with the assumption of exponential growth. If cells grow linearly instead of exponentially, then

mass at division = mass at birth + $\tilde{k}T$.

The constant $\tilde{k}$ is now the growth rate - mass per unit time - for individual cells; it is not related in any way to k, the specific growth rate of the population as a whole.

We shall find infinite series solutions for (2.16), (2.7′), (2.8′) in two cases suggested by the transition probability model discussed at the beginning of Sec. 2. Here $T = T_A + T_B$, where T_B is a positive, deterministic constant, and $\Pr\{T_A > t\} = \int_t^\infty f_1(s)ds$, where either

(4.1) $\quad f_1(t) = pe^{-pt} \qquad (t > 0)$ (transition probability model)

or

(4.2) $\quad f_1(t) = \frac{pq}{q-p}(e^{-pt} - e^{-qt}) \qquad (t > 0)$ (two transition model),

with p and q positive and $p \neq q$. The probability density (4.1) portrays T_A as a single random event which occurs with probability p per unit time. Eq. (4.2) portrays T_A as a sequence of two such events which occur successively in a fixed order. Eq. (4.1) is the limiting case of (4.2) where $q \to \infty$, and our solution formulas can be obtained by taking this limit. Since (4.1) is substantially simpler, however, we shall work out this case in detail and outline the procedure for (4.2). The solution for the excepted case p=q ($f_1(t) = p^2te^{-pt}$) of (4.2) can be obtained directly or by letting $q \to p$ in our solution formulas. (See 'Remark on singular cases' below.)

For simplicity in what follows, we scale time and mass in such a way that $\tilde{k} = T_B = 1$. Then, in terms of (2.16),

$$f(t) = \begin{cases} 0 & (0 < t \leq 1) \\ f_1(t-1) & (1 < t < \infty). \end{cases}$$

Notice that under steady state conditions, there is a minimum birth size $x_m = 1/(r-1)$, and $\phi(x) = 0$ for $x < x_o \equiv r/(r-1)$. To prove this (independently of Theorem 1), suppose a cell is born with mass $u_o < x_m$. Then, after one generation, this cell will give rise to daughters of mass $\geq u_1 \equiv (u_o+1)/r$. If we iterate this process, we find that, in the j'th generation, all descendents of the original cell have birth masses $\geq$

$$u_j \equiv r^{-j}u_o + (1-r^{-j})/(r-1) > (1-r^{-j})/(r-1),$$

and this approaches x_o/r as $j \to \infty$.

Since $m(-\sigma,x) = x-\sigma$ if $0 < \sigma < x-x_m$ and $m(-\sigma,x) = 0$ if $\sigma > x-x_m$, our basic equation (2.16) becomes

$$\phi(x) = r^2\int_1^{x-x_m} e^{-k\sigma}\phi(r(x-\sigma))f_1(\sigma-1)d\sigma, \tag{4.3}$$

or, after the change of variable $y=r(x-\sigma)$,

$$(4.4) \qquad \phi(x) = r\, e^{-kx} \int_{x_o}^{r(x-1)} e^{ky/r}\, \phi(y)\, f_1(x-1-(y/r))\, dy.$$

a. Transition probability model. When (4.1) holds, (4.4) is

$$(4.4a) \qquad \phi(x) = pr\, e^{-(k+p)x}\, e^{p} \int_{x_o}^{r(x-1)} e^{(k+p)y/r}\, \phi(y)dy.$$

Differentiate (4.4a) to obtain

$$\phi'(x) = -(k+p)\phi(x) + r(pre^{-k})\phi(r(x-1)).$$

But by (2.15), $pre^{-k} = k+p$, so we get

$$(4.5) \qquad \phi'(x) = -(k+p)\phi(x) + r(k+p)\phi(r(x-1)) \qquad (x_o < x < \infty).$$

Eq. (4.4a) also implies

$$(4.6) \qquad \phi(x_o) = 0.$$

Conversely, any solution of (4.5) and (4.6) will solve (4.4a); to see this, multiply (4.5) by $\exp[(k+p)x]$ and integrate.

A solution of (4.5) is

$$(4.7) \qquad \phi_o(x) = \sum_{n=o}^{\infty} (-1)^n\, c_n(r)\, r^n\, e^{-(k+p)r^n(x-x_o)},$$

where

$$c_o(r) = 1,\ c_n(r) = \frac{1}{(r-1)(r^2-1)\ldots(r^n-1)} \qquad (n=1,2,\ldots).$$

This series and its derivatives to all orders converge absolutely and uniformly on $\{x_o \le x < \infty\}$. Term-by-term integration shows that $\int_{x_o}^{\infty} |\phi_o(x)|\, dx < \infty$ and

$$(4.8) \qquad (k+p)\int_{x_o}^{\infty} \phi_o(x)dx = \sum_{n=0}^{\infty} (-1)^n c_n(r) \equiv N(r).$$

Approximate values for N(r) are given in Fig. 1.

By (4.8),

$$(4.9) \qquad \phi_o(x) \sim e^{-(k+p)(x-x_o)} \qquad (x \to \infty);$$

in particular, $\phi_o(x) \not\equiv 0$ and $\lim_{x\to\infty} \phi(x) = 0$. Integrating (4.5) from x_o to ∞ and changing variables, we see that

$$\phi_o(x_o) = -\int_{x_o}^{\infty} \phi_o'(x)dx = 0.$$

Thus (4.6) holds for ϕ_o, so ϕ_o satisfies (4.4a). By Theorem 1 and (4.8), $N(r) > 0$ and

$$(4.10) \qquad \phi(x) = \phi(x;r,p) = (k+p)\phi_o(x)/N(r)$$

is the unique solution of (2.16), (2.7′), (2.8′) in this case. The cumulative distribution function, $\Phi(x)$, derived from (4.10), is plotted in Fig. 2 for r = 2, 4 and 8.

b. Two transition model. Now let (4.2) hold. Then (4.4) can be written

$$(4.4b) \qquad \phi(x) = pq(q-p)^{-1} re^{p} e^{-(k+p)x} \int_{x_o}^{r(x-1)} e^{(k+p)y/r} \phi(y)dy$$

$$- pq(q-p)^{-1} re^{q} e^{-(k+q)} \int_{x_o}^{r(x-1)} e^{(k+q)y/r} \phi(y)dy \equiv \phi_1(x)+\phi_2(x).$$

Differentiate (4.4b) to obtain

$$(4.11) \qquad \phi'(x) = -k\phi(x) - p\phi_1(x) - q\phi_2(x).$$

Eliminating ϕ_1 from (4.4b), (4.11), we obtain

$$\phi(x) + (k+p)\phi'(x) = (p-q)\phi_2(x).$$

Another differentiation, using the definition of ϕ_2, shows that

$$\phi''(x) + [(k+p)+(k+q)]\phi'(x) + (k+p)(k+q)\phi(x) = pqr^2e^{-k}\phi(r(x-1)).$$

But (2.15) now says that $pqre^{-k} = (k+p)(k+q)$, so our equation is

(4.12) $$\phi''(x) + [(k+p)+(k+q)]\phi'(x) + (k+p)(k+q)\phi(x) = r(k+p)(k+q)\phi(r(x-1)).$$

Solutions of (4.12) are

(4.13) $$\alpha\phi_1(x;r,p,q) + \beta\phi_1(x;r,q,p),$$

where α and β are arbitrary constants and

(4.14) $$\phi_1(x;r,p,q) = \sum_{n=o}^{\infty} (-1)^n\, c_n(r,p,q)\, r^n\, e^{-r^n(k+p)(x-x_o)},$$

with

(4.15) $$c_o(r,p,q) = 1, \quad \frac{c_n(r,p,q)}{c_{n-1}(r,p,q)} = \frac{1}{r^n-1} \cdot \frac{1}{1-r^n\,\frac{k+p}{k+q}},$$

except in the singular cases where

$$1 = r^{n'}(k+p)/(k+q)$$

for some $n' \geq 1$ (recall that $p \neq q$). We discuss these singular cases in a remark below and ignore them for now.

The series (4.14) for ϕ_1 and its derivatives to all orders converge absolutely and uniformly on $\{x_o \leq x < \infty\}$. In particular, denote by ϕ^o any solution (4.13) where α and β are chosen so that $\phi^o(x_o) = 0$. Integrating (4.12) from x_o to ∞, we see that $(\phi^o)'(x_o) = 0$ as well. These initial

conditions enable us to reverse the steps leading from (4.4b) to (4.12). Since $p \neq q$, (4.13) shows that $\phi^o(x) \neq 0$ for large x, so $\phi^o \not\equiv 0$. By Theorem 1, there is a number $N = N(r,p,q) \neq 0$ such that $\phi(x) = \phi_o(x)/N$ is the unique solution of (2.16), (2.7′), (2.8′).

Remark on singular cases. When

$$(k+q) = r^{n'}(k+p), \text{ for some } n' = 0,1,2,\ldots$$

(including the excepted cases p=q, n′ = 0) the form of the series changes. The solution $\phi_1(x;r,q,p)$ of (4.12) remains valid (unless p=q), but there is a second solution of the form

$$\tilde{\phi}_1(x;r,p,q) =$$

$$\sum_{n=n'}^{\infty} (-1)^n \tilde{c}_n r^{n(x-x_o)} e^{-(k+p)r^n(x-x_o)} + \sum_{n=0}^{\infty} (-1)^n \tilde{d}_n r^n e^{-(k+p)r^n(x-x_o)}.$$

We shall not pursue the details.

5. The case of linear growth - moments. To compare our results with observations of cell size distributions in cell cultures experiencing balanced growth, we would like to know the moments of the size distribution $\phi(x)$. The moments can be obtained most easily by transforming (4.4) with the Laplace transformation

$$(5.1) \qquad L\{y\}(s) = \int_{x_o}^{\infty} y(x)e^{-s(x-x_o)}dx.$$

The result (obtained with a change in the order of integration, a change of variable, and use of the definition $x_o = r/(r-1)$) is

$$(5.2) \qquad L\{\phi\}(s) \equiv \tilde{\phi}(s) = \tilde{\phi}(s/r)\, re^{-k}\, \tilde{f}_1(s+k),$$

where $\tilde{f}_1(s) = \int_o^{\infty} e^{-st} f_1(t)dt$. By (2.15), this can also be written as

(5.3) $\tilde{\phi}(s) = \tilde{\phi}(s/r)\tilde{f}_1(s+k)/\tilde{f}_1(k).$

If ϕ is in L^1 and $\phi(x)$ decays exponentially (as in the examples of Sec. 4), then $\tilde{\phi}(s)$ is differentiable to all orders ($s \geq 0$) and

(5.4) $\tilde{\phi}^{(m)}(0) = (-1)^m \int_{x_o}^{\infty} (x-x_o)^m \phi(x)dx \qquad (m = 1,2,\ldots).$

Eq. (2.8′) implies that

(5.5) $\tilde{\phi}(0) = 1.$

For the higher derivatives, assuming that f_1 decays exponentially, we can differentiate (5.3) to obtain

(5.6) $(1-r^{-m})\ \tilde{f}_1(k)\ \tilde{\phi}^{(m)}(0) = \sum_{j=0}^{m-1} \binom{m}{j} r^{-j}\ \tilde{\phi}^{(j)}(0)\ \tilde{f}_1^{(m-j)}(k), \quad m = 1,2,\ldots .$

This enables us to evaluate the derivatives $\tilde{\phi}^{(m)}(0)$ inductively; using (5.4) we can then determine the moments of ϕ.

In practice, in the common case where $\tilde{f}_1$ is rational (e.g., if f_1 is a linear combination of exponentials), the work can be simplified by first multiplying (5.3) by the denominator of $\tilde{f}_1(s+k)$, so that only polynomials need be differentiated.

Specifically, in example (a) of Sec. 4, (5.3) becomes

(5.3a) $(k+p+s)\ \tilde{\phi}(s) = (k+p)\ \tilde{\phi}(s/r).$

Solving for the derivatives of $\tilde{\phi}$, one obtains

(5.7) $\int_{x_o}^{\infty} (x-x_o)^m \phi(x)dx = m!(k+p)^{-m}\ r^{m(m+1)/2}\ c_m(r) \qquad (m = 0,1,2,\ldots).$

The first three moments of ϕ are

(5.8) $\quad \text{mean} = \langle x \rangle = x_o[1 + (k+p)^{-1}] = r(r-1)^{-1}[1 + (k+p)^{-1}],$

(5.9) $\quad \text{variance} = \mu_2 = [x_o/(k+p)]^2(r-1)/(r+1) = (k+p)^{-2}r^2(r^2-1)^{-1},$

(5.10) $\quad \text{skewness} = \mu_3/\mu_2^{3/2} = 2(r-1)^{1/2}\ (r+1)^{3/2}\ (r^2+r+1)^{-1},$

where μ_n = n'th moment of $\phi(x)$ about the mean. The distribution ϕ is quite close to the Pearson Type III distribution [1]

(5.11) $\quad P(x;x_o,\beta,\rho) = y^{\rho-1}\ e^{-y}/\beta\Gamma(\rho),$

where

$y = (x-x_o)/\beta$, $\Gamma(\cdot)$ = gamma function

$\beta = r/(k+p)(r+1)$, $\rho = (r+1)/(r-1)$.

Distribution (5.11) has mean and variance given by (5.8) and (5.9) and a skewness of $2(r-1)^{1/2}(r+1)^{-1/2}$. The ratio of the skewness of P(x) to that of $\phi(x)$ is $(r^2+r+1)/(r^2+2r+1)$, which is never much different from 1. P(x) and $\phi(x)$ are compared in Fig. 2 for r=2.

Similar computations can be carried out for the three other models discussed at the beginning of Sec. 2. The results are collected in Table 2.

6. Discussion

The probabilistic cell cycle models described in this paper are easy to simulate by Monte-Carlo methods. Some results of simulations for the transition probability model are reported in Table 3. The numerical values for the mean and variance of the division mass distribution function, $\phi(x)$, compare excellently with the analytical results in Sec. 5. In Fig. 2 we compare the analytical result for the cumulative distribution function for the transition probability model,

$$(6.1) \qquad \bar{\Phi}(x) = N^{-1}(r) \sum_{n=0}^{\infty} (-1)^n c_n(r) e^{-(k+p)r^n(x-x_o)},$$

with the Monte-Carlo simulations, and we see more clearly the excellent agreement between theory and simulation.

We have shown for probabilistic models of the cell cycle that, if cell growth is exponential, then there is no steady-state size distribution. This is contrary to the facts for expanding cell cultures. On the other hand, a steady-state size distribution does exist for probabilistic models of the cell cycle, if cells grow linearly (i.e., at constant rate per unit time, independent of cell size), and our Monte-Carlo simulations indicate that this steady-state size distribution is stable.

However, the assumption of linear growth does not accord with a well-known relation between average cell size and population doubling time in bacteria. In Salmonella typhimurium Schaechter, Maaløe and Kjeldaard [12] found that

$$(6.2) \qquad \ln\langle x\rangle_\lambda = \mu + \nu k$$

where $\langle x\rangle_\lambda$ is the average cell mass (protein content) in a sample of extant cells, k is the specific growth rate of culture (i.e., doublings per hour), and

μ and ν are experimentally determined constants. A simple relation between $\langle x \rangle_\lambda$ and k can be derived from the Collins-Richmond equation [9]

$$(6.3) \qquad \lambda(x)V(x) = k(r-1)^{-1} \int_x^\infty [(r-1)\lambda(y) + \phi(y) - r\psi(y)]dy$$

where $\lambda(x)dx$ = probability that the present size of an extant cell lies between x and x+dx, $\psi(x) = -\Psi'(x)$ = probability density for birth mass in a sample of newborn cells, and V(x) = rate of growth of a cell of size x. (The Collins-Richmond equation is just a statement of the conservation of cells for populations in steady state expansion.) If individual cells grow linearly, then $V(x) = \tilde{k}$ = constant, and, on integrating (6.3) from x = 0 to ∞, we obtain (after changing the order of integration and substituting $r\phi(rx)$ for $\psi(x)$)

$$(6.4) \qquad \tilde{k} = k \langle x \rangle_\lambda .$$

In order that (6.4) and (6.2) be consistent, we must insist that

$$(6.5) \qquad \tilde{k} = \gamma k e^{\nu k}$$

where $\gamma = e^\mu$ = constant. There is no obvious reason why $\tilde{k}$ should depend on k in just this way.

This discrepancy, along with other problems, indicates that probabilistic models with linear growth are not adequate descriptions of the rules of cell growth and division [14]. It appears that there must be some correlation between cell size and the probability of cell division [15].

BIBLIOGRAPHY

1. M. Abramowitz and I. A. Stegun, Handbook of Mathematical Functions, Dover, New York (1965) p. 930.

2. W. N. Bailey, Generalized Hypergeometric Functions, Cambridge Univ. Press, London (1935) Sec. 8.2 (6).

3. R. F. Brooks, Variability in the cell cycle and the control of proliferation. In: The Cell Cycle, Edit. by P. C. L. John, Cambridge Univ. Press, London (1981) pp. 35-61.

4. R. F. Brooks, D. C. Bennett and J. A. Smith, Mammalian cell cycles need two random transitions, Cell 19 (1980) pp. 493-504.

5. K. L. Chung, A Course in Probability Theory, Harcourt, Brace, and World, Inc., New York (1968) Thm. 4.4.4.

6. D. G. Kendall, On the role of variable generation time in the development of a stochastic birth process, Biometrika 35 (1948) pp. 316-330.

7. H. E. Kubitschek, The distribution of cell generation times, Cell Tiss. Kinet. 4 (1971) pp. 113-122.

8. J. M. Mitchison, The Biology of the Cell Cycle, Cambridge Univ. Press, London (1971) chapter 7.

9. P. R. Painter and A. G. Marr, Mathematics of microbial populations, Ann. Rev. Microbiol. 22 (1968) pp. 519-548.

10. O. Rahn, A chemical explanation of the variability of the growth rate, J. Gen. Physiol. 15 (1932) pp. 257-277.

11. W. Rudin, Real and Complex Analysis, Second Ed., McGraw-Hill, New York (1972) Thm. 11.22.

12. M. Schaechter, O. Maaløe and N. O. Kjeldgaard, Dependency on medium and temperature of cell size and chemical composition during balanced growth of Salmonella typhimurium, J. Gen. Microbiol., 19 (1958) pp. 592-606.

13. J. A. Smith and L. Martin, Do cells cycle? Proc. Natn. Acad. Sci. USA 70 (1973) pp. 1263-1267.

14. J. J. Tyson and K. B. Hannsgen, Cell size homeostasis and the transition probability model (in preparation).

15. J. J. Tyson and K. B. Hannsgen, The distribution of cell generation times: a model incorporating size control and random transitions (in preparation).

Appendix - Some interesting series. The formula (4.7) leads to several known identities from the theory of basic hypergeometric series. In this appendix, set k+p=1 in (4.7). Since ϕ_o satisfies (4.5) and (4.6), $\phi_o^{(m)}(x_o) = 0$ (m = 0,1,2,...). Differentiating (4.7) m times, we find that

$$\phi_o^{(m)}(x) = \sum_{n=0}^{\infty} (-1)^{n+m} c_n(r)\, r^{(1+m)n}\, e^{-r^n(x-x_o)},$$

so

$$0 = (-1)^m \phi_o^{(m)}(x_o) = \sum_{n=o}^{\infty} (-1)^n c_n(r)\, r^{n(m+1)}$$

$$= 1 + \sum_{n=1}^{\infty} (-1)^n c_n(r)\, r^{n(m+1)}.$$

Let $q = 1/r$. Then

$$(A1) \qquad 1 + \sum_{n=1}^{\infty} c_n(q)\, q^{-mn}\, q^{n(n-1)/2} = 0.$$

Bailey [2], gives

$$(A2) \qquad 1 + \sum_{n=1}^{\infty} c_n(q)\, z^n\, q^{n(n-1)/2} = \prod_{n=0}^{\infty} (1-zq^n)$$

for $|q| < 1$, $|z| < 1$; with $z = q^{-m}$ (m = 0,1,2,...), (A2) reduces to (A1). Notice that the identity (A2) is valid for $|z| \geq 1$, $|q| < 1$, since both sides are entire in z.

Next we compute the moments of $\phi_o(x)$ about $x=x_o$ by integrating the series expansion (4.7) term by term:

$$(A3) \quad \int_{x_o}^{\infty} (x-x_o)^m \phi_o(x)dx = m![1 + \sum_{n=1}^{\infty} (-1)^n c_n(r) r^{-mn}].$$

Comparing (5.7) to (A3), and using (4.8) and (4.10), we see that

$$1 + \sum_{n=1}^{\infty} (-1)^n c_n(r) r^{-mn} = r^{m(m+1)/2}\, c_m(r)\Big[1 + \sum_{n=1}^{\infty} (-1)^n c_n(r)\Big],$$

or, with $q = 1/r$,

$$\text{(A4)} \qquad 1 + \sum_{n=1}^{\infty} c_n(q)\, q^{n(n-1)/2}\, q^{n(m+1)}$$

$$= (-1)^m c_m(q)\Big[1 + \sum_{n=1}^{\infty} c_n(q) q^{n(n+1)/2}\Big].$$

In (A2), set $z = q^{m+1}$; the left side of (A4) is equal to $(1-q^{m+1})(1-q^{m+2})\ldots$.

Next, set $z=q$ in (A2); the right side of (A4) is also equal to

$(1-q^{m+1})(1-q^{m+2})\ldots$.

Table 1

Increase in the mean and standard deviation of cell size for the transition probability model, assuming exponential cell growth.

Generation Number	Mean	Standard Deviation	Minimum	Maximum
0	2	0	2.0	2×10^0
10	15	147	0.2	5×10^3
20	34	151	0.06	4×10^3
30	95	391	0.04	7×10^3
40	317	1640	0.01	3×10^4
50	1497	15955	0.009	5×10^5

A population of 1000 cells was started out with all cells of mass = 1 at birth. Next 1000 random numbers, representing T_A for each cell, were generated according to an exponential distribution with mean = 1 (i.e., p = 1). The generation time of each cell was taken to be $T_A + 1$ (i.e. $T_B = 1$), and the mass of each cell at division was calculated as birth mass times $\exp\{\tilde{k}(T_A + 1)\}$, where $\tilde{k}$ = specific growth rate for individual cells. If a steady-state exists, this parameter must be the same as the specific growth rate for the population as a whole [15]. Thus we set $\tilde{k} = k$, where k, according to Eq. [10] in Smith and Martin [13], is given by the root of the transcendental equation

$$p + k - 2p \exp(-kT_B) = 0.$$

In our case ($p = T_B = 1$), we find $\tilde{k} = k = 0.375$, which corresponds to a population doubling-time of 1.85, whereas the mean generation time is $T_B + p^{-1} = 2$. After calculating the mass of each cell at division, we retained one daughter (of half the mother-cell mass), thus generating a new population of 1000 newborn cells of known birth masses. This process was repeated 50 times, and at division in each generation were calculated the mean cell mass, the standard deviation of cell masses, the minimum cell mass and the maximum cell mass. Though this algorithm does <u>not</u> produce the mean and variance of $\phi(x,t)$ =

time-dependent probability density for mass at division in a sample of dividing cells, it does illustrate clearly the continual broadening of the distribution of cell sizes in the probabilistic model with exponential cell growth.

Table 2

The mean and variance of the division-mass distribution function for four different probabilistic models of the cell cycle.

Model	$f_1(t)$	Mean	Variance
Transition Prob.	pe^{-pt}	$x_o\left[1 + \frac{1}{k+p}\right]$	$x_o^2 \frac{r-1}{r+1} \frac{1}{(k+p)^2}$
Two Transition	$\frac{pq}{p-q}(e^{-pt}-e^{-qt})$	$x_o\left[1 + \frac{1}{k+p} + \frac{1}{k+q}\right]$	$x_o^2 \frac{r-1}{r+1}\left[\frac{1}{(k+p)^2} + \frac{1}{(k+q)^2}\right]$
Kendall's	$\frac{p}{(g-1)!}(pt)^{g-1} e^{-pt}$	$x_o\left[1 + \frac{g}{k+p}\right]$	$x_o^2 \frac{r-1}{r+1} \frac{g}{(k+p)^2}$
Rahn's	$gpe^{-pt}(1-e^{-pt})^{g-1}$	$x_o\left[1 + \frac{1}{k+p} + \frac{1}{k+2p} + \ldots + \frac{1}{k+gp}\right]$	$x_o^2 \frac{r-1}{r+1}\left[\frac{1}{(k+p)^2} + \frac{1}{(k+2p)^2} + \ldots + \frac{1}{(k+gp)^2}\right]$

Table 3

Mean and variance of the steady-state division-mass distribution function, as predicted by equations (5.8) and (5.9) and (in parentheses) as calculated from Monte-Carlo simulations.

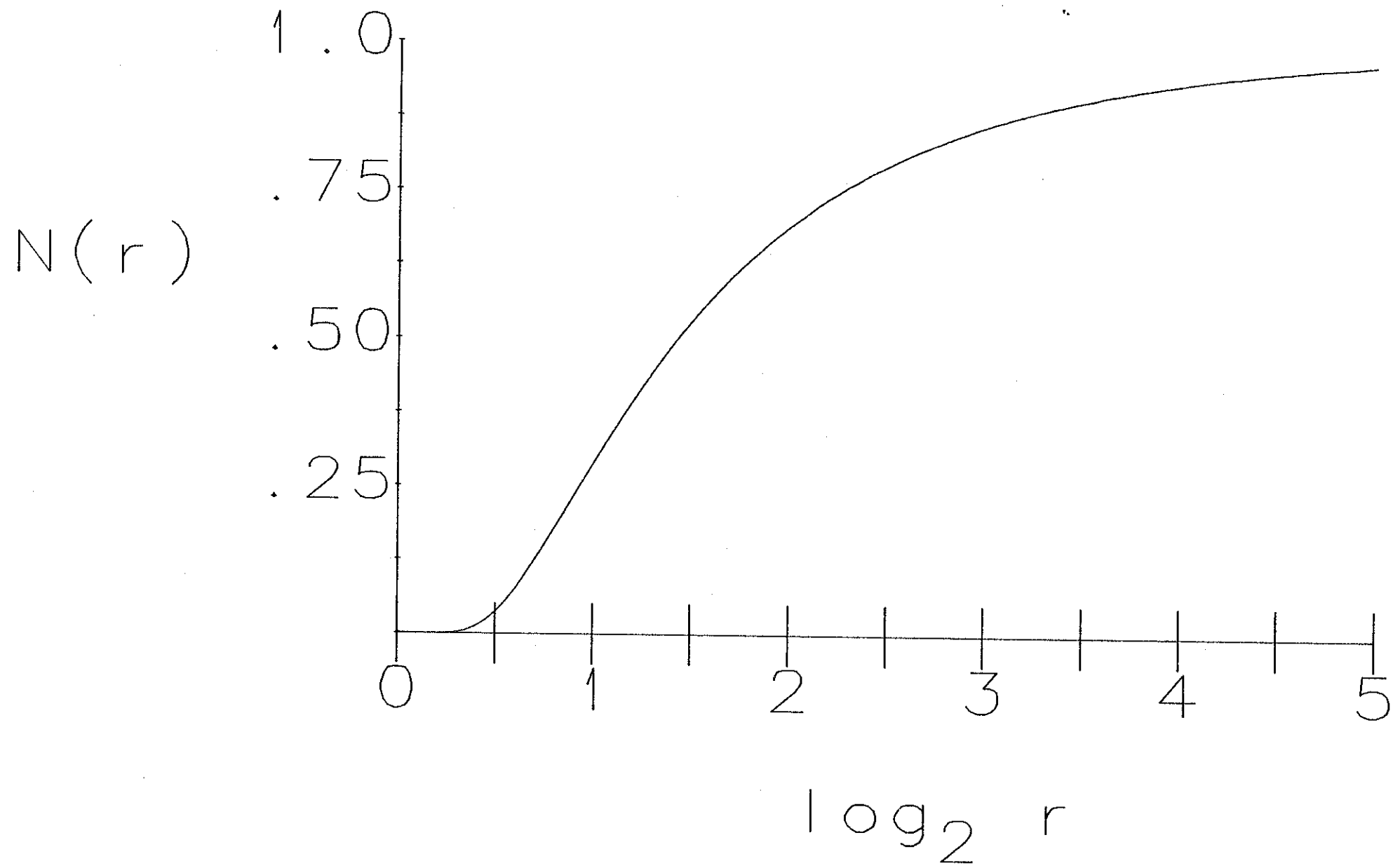
	r = 2	r = 4	r = 8
p = 2	2.807 (2.811) 0.217 (0.218)	1.780 (1.801) 0.120 (0.166)	1.468 (1.496) 0.082 (0.073)
p = 1	3.455 (3.428) 0.705 (0.763)	2.074 (2.074) 0.330 (0.329)	1.648 (1.598) 0.198 (0.114)
p = .5	4.610 (4.627) 2.271 (2.400)	2.547 (2.690) 0.883 (0.890)	1.911 (1.899) 0.459 (0.526)

In each box we report the mean and variance predicted by equations (5.8) and (5.9) and, in parentheses, the same quantities calculated from a simulated mass distribution. The simulation was carried out as follows. At time t = 0 a cell of arbitrary birth-mass, μ_o, was created. The generation time, T_o, of this cell was calculated by choosing a random number, T_A, from an exponential distribution with mean p^{-1} and adding $T_B = 1$, i.e., $T_o = T_A + 1$. The mass at division was calculated by adding $\tilde{k}T_o$ (with $\tilde{k}$=1) to the mass at birth. Thus at time t = T_o we had r newborn cells of identical birth-masses, $\mu_i = (\mu_o + T_o)/r$, i = 1,2,..., r. The generation times of these cells, T_i (i = 1,2, ..., r) were then calculated by choosing r random numbers from an exponential distribution (mean = p^{-1}) and adding 1 to each. For each of the r cells, the times of division, t = $T_o + T_i$, and the masses at division, $M_i = \mu_i + T_i$, were recorded. The calculation continued in this fashion, allowing the cell population to expand

exponentially in time until there were 4000-8000 cells (i.e., 12 or 13 population doublings). At regular intervals of elapsed time a sample of dividing cells was chosen by searching for all cells which divided in a time window, chosen small enough so that no cell could divide twice in the time window but large enough so that about 25% of the total current population would be sampled. From this sample of dividing cells, the mean and variance of the division masses were calculated. By the 12th population doubling the division-mass distribution had reached steady-state, as judged by the relative constancy of the mean and variance.

Figure 1. N(r), as defined by (4.8). N(r) is needed to normalize the division-mass probability density function (4.10) for the transition probability model.

Figure 2. The division-mass cumulative distribution function for the transition probability model with linear growth. The solid lines are graphs of equation (6.1) for p = 0.5 and r = 2,4,8. The dashed line is a graph of the cumulative Pearson Type III distribution for p = 0.5, r = 2, k = 0.266; i.e., $\mathbb{P}_p(x) = e^{-y}(y^2/2 + y + 1)$, y = (x-2)/0.8700. The solid circles are the observed distributions of division masses in the Monte-Carlo simulations of the model presented in Table 3 (last row). The open circles report a second simulation of the case r=4.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.75
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N(r)
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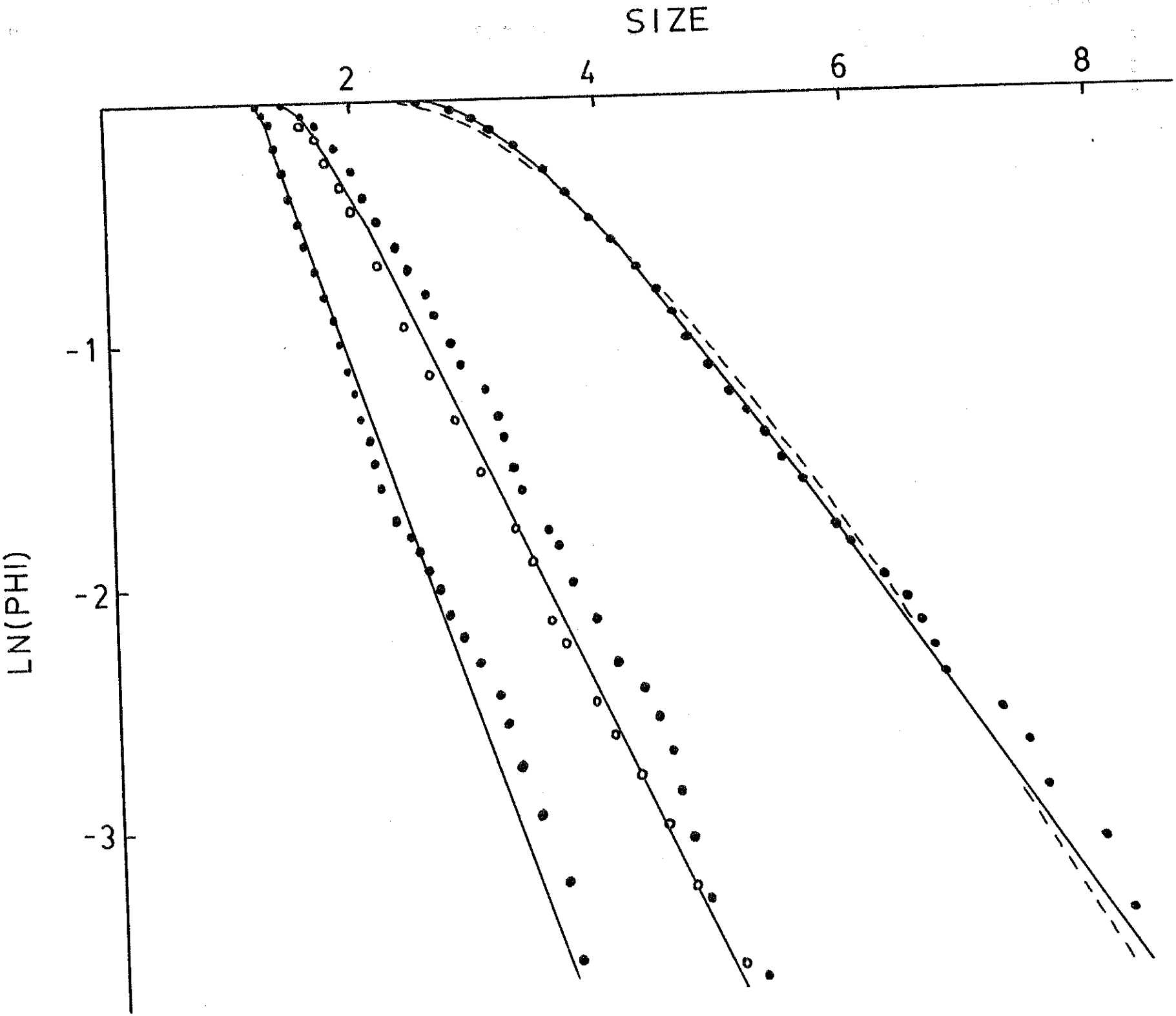
SIZE
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