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Failure of programmed cell death and differentiation as causes of tumors: Some simple mathematical models

(tumorigenesis/apoptosis)

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ABSTRACT **Most models of tumorigenesis assume that the tumor grows by increased cell division. In these models, it is generally supposed that daughter cells behave as do their parents, and cell numbers have clear potential for exponential growth. We have constructed simple mathematical models of tumorigenesis through failure of programmed cell death (PCD) or differentiation. These models do not assume that descendant cells behave as their parents do. The models predict that exponential growth in cell numbers does sometimes occur, usually when stem cells fail to die or differentiate. At other times, exponential growth does not occur: instead, the number of cells in the population reaches a new, higher equilibrium. This behavior is predicted when fully differentiated cells fail to undergo PCD. When cells of intermediate differentiation fail to die or to differentiate further, the values of growth parameters determine whether growth is exponential or leads to a new equilibrium. The predictions of the model are sensitive to small differences in growth parameters. Failure of PCD and differentiation, leading to a new equilibrium number of cells, may explain many aspects of tumor behavior—for example, early premalignant lesions such as cervical intraepithelial neoplasia, the fact that some tumors very rarely become malignant, the observation of plateaux in the growth of some solid tumors, and, finally, long lag phases of growth until mutations arise that eventually result in exponential growth.**

The control of the cell cycle and cell death are frequently deranged in tumors (1). The most frequently mutated locus in tumors, *TP53* (2), is centrally involved in pathways leading to programmed cell death (PCD) (3–5). Failure of PCD may be important in causing the development of a tumor and in determining its response to antitumor therapy (6). The precise role of PCD in tumorigenesis is, however, still unclear. There are several ways in which the failure of PCD might lead to or promote tumor growth. One such way may be to give cells the equivalent of a replicative advantage, whereby failure to die is effectively the same as more rapid cell division. Alternatively, the failure of PCD may lead to an increase in the intrinsic mutation rate by permitting cells to live on into senescence and be exposed to mutagens or acquire more spontaneous mutations. Failure of PCD can be regarded as a special case of failure of cell differentiation, which may be of general importance in tumorigenesis.

Basic models of tumorigenesis assume that the tumor grows by increased cell division (7–10). A single mutant cell replicates at a maximum rate of 2^g (where g is the number of divisions or "generations" for the mutant). More generally, the rate of replication in the tumor is $(1 + w)^g$ per generation, where w is the selective advantage of the mutant (relative to a mean nonincreasing population of normal cells). All descendants of the original mutant cell simply replicate at that rate. Models of tumorigenesis by failure of PCD or differentiation cannot be assumed to occur via the same straightforward mathematical pathway as tumorigenesis by increased cell division. The reason for this is fundamental to models that incorporate cell death and differentiation: even for the purpose of simplification, it cannot be assumed that descendant cells behave as their parents do. For example, a mutation occurring in a stem cell may have no effect until the cell is fully differentiated and about to undergo PCD. By that time, the cell may have divided several times or not at all, depending on its function, or may even have risked PCD at another stage in its development. Consequently, it is not clear that tumor growth will necessarily be exponential, if it occurs at all. When cells differentiate and die a planned death, the effect of any mutation and its importance in tumorigenesis are likely to vary, depending on when and where it occurs and has its effects.

This study sets up simple mathematical models of tumorigenesis by failure of PCD in particular and by failure of differentiation in general. The models aim to demonstrate how tumor growth proceeds under these circumstances and compare it with potentially exponential growth under tumorigenesis simply by increased rates of cell division.

The Model

General Features. The model is based on a simple system of cell differentiation, resembling a colonic crypt (11), but applicable generally (Table 1). A self-renewing population of stem cells (F_0) is normally fixed at number N_0. The stem cells give rise to a cell population of intermediate differentiation (F_1) with number N_1, where N_1 is dependent on the number of cells coming through from F_0 and on the number lost to the population of fully differentiated cells (F_2, with number N_2). Cells in the F_2 stage may undergo PCD to form a nominal population of dead cells (F_3).

The number of cells in any F_n ($n = 0, 1, 2, 3$) depends on the following variables: (*i*) the number of cells in F_{n-1} ($1 \leq n \leq 3$); (*ii*) the rate of division of cells in the F_{n-1}; (*iii*) the probability that cells in F_{n-1} differentiate into F_n cells, rather than remain in F_{n-1} or die; (*iv*) the rate of division of cells in F_n; and (*v*) the probability that cells in F_n differentiate into cells in F_{n+1} or die, rather than remain in F*n*.

We denote the time for one cell division to occur in population F_n as t_n (where, for convenience, values are normally measured relative to baseline t_0). The number of cells after G divisions is denoted by $N_n(G)$. After each cell in F_n divides, it may (*i*) die, (*ii*) differentiate to form a F_{n+1} cell, or (*iii*) renew itself. These proportions are denoted in the F_0 by α_1, α_2, and α_3, respectively. β_1, β_2, and β_3 are the respective proportions for F_1. γ is the probability of a cell in the F_2 population dying (that is, passing through to the F_3 stage per unit time).

The following restrictions apply to the values of α, β, and γ: (*i*) $\alpha_1 + \alpha_2 + \alpha_3 = 1$; $\beta_1 + \beta_2 + \beta_3 = 1$; (*ii*) α_1, α_2, α_3, β_1, β_2,

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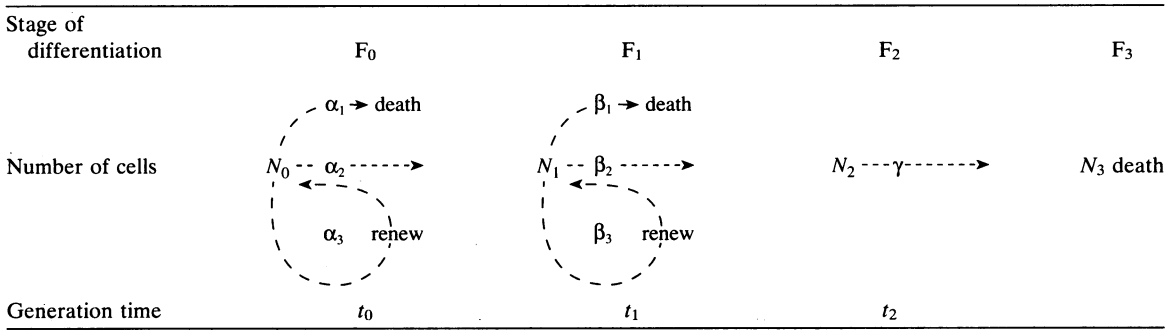
Abbreviation: PCD, programmed cell death.

Table 1. Model of cell division, differentiation, and death



F_0, F_1, and F_2 represent stem cells, semidifferentiated cells, and fully differentiated cells, respectively.

β_3, $\gamma \geq 0$; (*iii*) α_3 (normally) = 1/2; (*iv*) β_2 (normally) $\leq 1/2$ (owing to differences among t values); and (*v*) $\gamma \leq 1$.

Results

Model 1: Normal Cell Division. Cell numbers in successive generations in F_0 are related according to

$$N_0(G+1) = 2\alpha_3 N_0(G). \quad [1]$$

At equilibrium,

$$N_0(G+1) = N_0(G),$$
$$\Rightarrow 2\alpha_3 N_0(G) = N_0(G),$$
$$\Rightarrow \alpha_3 = 0.5. \quad [2]$$

There is, as expected, a unique point of equilibrium at which the population of stem cells exactly renews itself. If α_3 rises above or falls below 0.5, N_0 respectively increases or decreases exponentially.

For F_1,

$$N_1(G+1) = 2\beta_3 N_1(G)t_0/t_1 + 2\alpha_2 N_0(G), \quad [3]$$

and at equilibrium

$$N_1(G+1) = N_1(G),$$
$$\Rightarrow N_1(G) = 2\beta_3 N_1(G)t_0/t_1 + 2\alpha_2 N_0(G),$$
$$\Rightarrow N_1(G) = \frac{2\alpha_2 N_0(G)}{[1 - (2\beta_3 t_0/t_1)]}. \quad [4]$$

Unlike F_0, therefore, there are multiple equilibria depending on $N_0(G)$, α_2, β_3, t_0, and t_1. No equilibrium exists when N_0 is not at equilibrium or when $2\beta_3 t_0/t_1 > 1$. In the latter case, N_1 increases exponentially because of self-renewal of F_1 cells above the number required simply to maintain their steady state.

For F_2,

$$N_2(G+1) = 2\beta_2 N_1(G)t_0/t_1 + N_2[1 - (\gamma t_0/t_2)], \quad [5]$$

and at equilibrium

$$N_2(G+1) = N_2(G),$$
$$\Rightarrow N_2(G) = 2\beta_2 N_1(G)t_0/t_1 + N_2[1 - \gamma(t_0/t_2)],$$
$$\Rightarrow N_2(G) = \frac{2\beta_2 N_1(G)t_2/t_1}{\gamma}. \quad [6]$$

As for F_1, there exist multiple equilibria depending on $N_1(G)$, β_2, γ, t_2, and t_1. However, when N_1 is at equilibrium, so is N_2.

The simple results illustrate the increased complexity of behavior that accompanies models that consider cell differentiation and PCD. Parameters of replication are constrained within limits for the cell population to be at equilibrium. The limits for stem cells are restrictive but are less so for partially or fully differentiated cells. We now analyze the case in which a mutation has altered the proportions of cells differentiating, dying, or renewing themselves in order to determine the effects on tumorigenesis. The models deliberately do not specify in which cells the mutation occurs or the locus at which it might occur.

Model 2: Change in γ, Proportion of Differentiated Cells Undergoing Programmed Death. γ only affects the value of N_2. It is assumed for the purposes of the model that a mutation causes γ to change by value δ, where $(0 \leq \gamma + \delta \leq 1)$. This mutation may have occurred in the F_2 population itself but would be unlikely to have a large effect, since only one cell and its descendants are affected. It is more likely that the mutation has occurred in the F_0 or F_1 population but only has an effect on F_2 cells.

After the mutation has occurred,

$$N_2(G+1) = 2\beta_2 N_1(G)t_0/t_1 + N_2(G)[1 - (\gamma + \delta)t_0/t_2]. \quad [7]$$

At equilibrium

$$N_2(G+1) = N_2(G),$$
$$\Rightarrow N_2(G) = 2\beta_2 N_1(G)t_0/t_1 + N_2(G)[1 - (\gamma + \delta)t_0/t_2],$$
$$\Rightarrow N_2(G) = \frac{2\beta_2 N_1(G)t_2/t_1}{(\gamma + \delta)}. \quad [8]$$

Therefore, a change in the proportion of differentiated cells undergoing apoptosis does not lead to exponential tumor growth but rather to a new equilibrium. When $\delta < 0$, $N_2(G)$ will be larger than before, and when $\delta > 0$, $N_2(G)$ will be smaller than before. Clearly, if $\gamma + \delta \approx 0$, N_2 can be very large at equilibrium. N_1 and N_0 populations are always unchanged. Fig. 1 shows how $N_2(G)$ depends on δ for representative values of β_2, N_1, t_2, t_1, and γ.

Model 3: Change in β_1, the Proportion of Semidifferentiated Cells Undergoing Programmed Death. Here, it is assumed a mutation occurs in an F_0 or F_1 cell that causes β_1 to be reduced by an amount δ $(0 < \delta < \beta_1)$. The cells that fail to die are partitioned between β_2 and β_3 relative to their original values. Here,

$$N_1(G+1) = 2\beta_3[1 + \delta/(\beta_2 + \beta_3)]N_1(G)t_0/t_1 + 2\alpha_2 N_0(G), \quad [9]$$

and at equilibrium

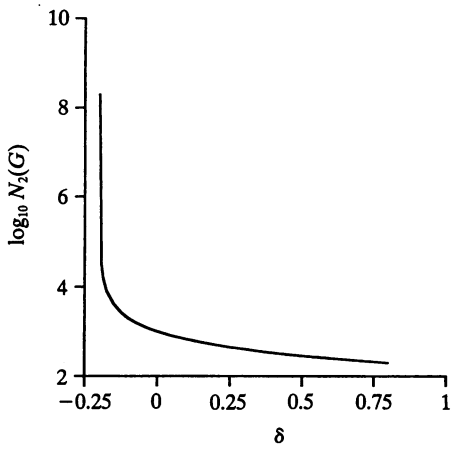


FIG. 1. Model 1: dependence of N_2 at equilibrium on δ. α_1 is set at 0.1, α_2 is 0.4, α_3 is 0.5, β_1 is 0.275, β_2 is 0.5, β_3 is 0.225, γ is 0.2, t_0 is 2, t_1 is 1, and t_2 is 0.5. Under normal growth, the equilibrium value of N_0 is 50, that of N_1 is 400, and that of N_2 is 1000. These values and those used in the other figure have been chosen for illustrative purposes.

$$N_1(G) = 2\beta_3[1 + \delta/(\beta_2 + \beta_3)]N_1(G)t_0/t_1 + 2\alpha_2 N_0(G),$$

$$\Rightarrow N_1(G) = \frac{2\alpha_2 N_0(G)}{1 - 2\beta_3(t_0/t_1)[1 + \delta/(\beta_2 + \beta_3)]}. \quad [10]$$

There is no equilibrium when

$$2\beta_3(t_0/t_1)[1 + \delta/(\beta_2 + \beta_3)] > 1,$$

$$\Rightarrow 2\beta_3(t_0/t_1) + 2\delta\beta_3(t_0/t_1)/(\beta_2 + \beta_3) > 1,$$

$$\Rightarrow \delta > \frac{\{1 - 2\beta_3(t_0/t_1)\}\{\beta_2 + \beta_3\}}{2\beta_3(t_0/t_1)}. \quad [11]$$

If this condition is fulfilled, cell numbers in the F_1 population undergo exponential growth, but they do not do so otherwise. In the model of normal cell differentiation, the condition $2\beta_3 t_0/t_1 > 1$ must hold for the population not to reach equilibrium. Therefore, the tendency to nonequilibrium is made more likely by the term $2\delta\beta_3(t_0/t_1)/(\beta_2 + \beta_3)$ (when δ is positive). Again, however, failure of PCD does not necessarily lead to exponential tumor growth. Thus, when δ is less than the limit given in Eq. 11, $N_1(G)$ simply approaches a new, higher equilibrium. At each such stage, however, the probability that a decrease in β_1 caused by a new mutation then leads to exponential growth of F_1 cells (and hence of the tumor overall) must increase.

N_0 is unchanged here. For F_2 in this case, however,

$$N_2(G + 1) = 2\beta_2[1 + \delta/(\beta_2 + \beta_3)]N_1(G)t_0/t_1 + (1 - \gamma t_0/t_2)N_2(G), \quad [12]$$

and at equilibrium

$$N_2(G) = 2\beta_2[1 + \delta/(\beta_2 + \beta_3)]N_1(G)t_0/t_1 + (1 - \gamma t_0/t_2)N_2(G),$$

$$\Rightarrow N_2(G)[1 - (1 - \gamma t_0/t_2)] = 2\beta_2[1 + \delta/(\beta_2 + \beta_3)]N_1(G)t_0/t_1,$$

$$\Rightarrow N_2(G) = \frac{2\beta_2[1 + \delta/(\beta_2 + \beta_3)]N_1(G)t_2}{\gamma t_1}. \quad [13]$$

That is, the existence of an equilibrium depends solely on whether $N_1(G)$ is at equilibrium. N_2 is always increased, however. It will increase exponentially when N_1 is also increasing exponentially. Fig. 2 shows both the approach of N_1 and N_2 to equilibrium and the exponential growth for N_1 and N_2 (with different values of δ and appropriate values of β_2, β_3, N_1, t_2, t_1, and γ).

Model 4: Change in α1, Proportion of Stem Cells Undergoing Programmed Death. Here, it is assumed that α_1 is reduced by an amount $\delta(0 < \delta < \alpha_1)$ and that the cells that fail to die are partitioned between α_2 and α_3. This model, which considers stem cells, provides a comparison with the previous model, which considered semidifferentiated cells. In successive generations

$$N_0(G + 1) = 2\alpha_3[1 + \delta/(\alpha_2 + \alpha_3)]N_0(G), \quad [14]$$

and at equilibrium

$$N_0(G) = 2\alpha_3[1 + \delta/(\alpha_2 + \alpha_3)]N_0(G),$$

$$\Rightarrow 2\alpha_3[1 + \delta/(\alpha_2 + \alpha_3)] = 1,$$

$$\Rightarrow \delta = (\alpha_2 + \alpha_3)[(1/2\alpha_3) - 1]. \quad [15]$$

There is no equilibrium when these special conditions are not met ($\delta = 0$, $\alpha_3 = 0.5$). Otherwise, there is exponential growth in cell numbers in all F_n. N_1 and N_2 rise exponentially according to their normal dependence on N_0.

Model 5: Change in α_2, Proportion of Stem Cells Undergoing Differentiation, Relative to α_3. A mutation causes α_2 to be reduced or increased by an amount δ $(-\alpha_2 < \delta < 1 - \alpha_2)$. The cells that fail to die are added to α_3. Then

$$N_0(G + 1) = 2(\alpha_3 + \delta)N_0(G), \quad [16]$$

and at equilibrium

$$N_0(G) = 2(\alpha_3 + \delta)N_0(G). \quad [17]$$

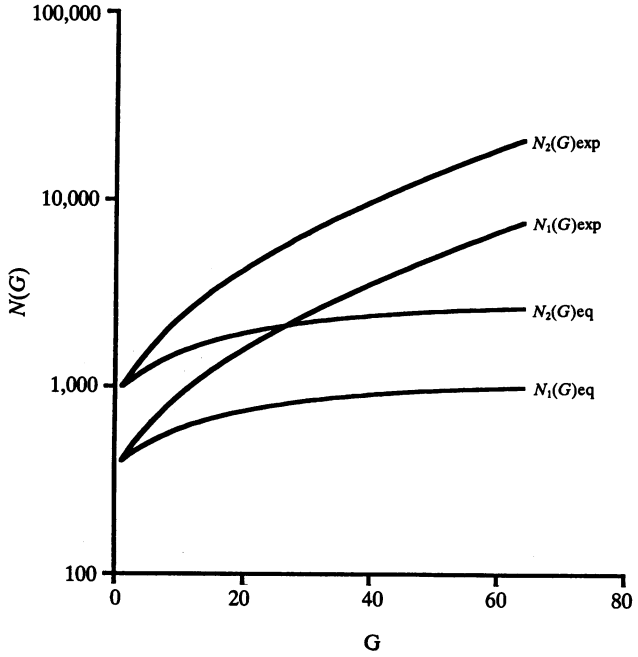


FIG. 2. Model 3: approach of $N_1(G)$ and $N_2(G)$ to a higher equilibrium or exponential growth. Values of α_1, α_2, α_3, β_1, β_2, β_3, γ, t_0, t_1, and t_2 are as given in Fig. 1. δ in this model is set at 0.05 (see text) when equilibrium occurs [$N_1(G)$eq, $N_2(G)$eq] and at 0.1 when there is exponential growth [$N_1(G)$exp, $N_2(G)$exp]. Once δ exceeds 0.08056 in this model, exponential growth occurs.

Clearly, there is no equilibrium (unless $\delta = 0$), and N_0 always rises or falls. Simulation shows that, with $\delta < 0$, there is a transient rise in N_1 and N_2, followed by a decline to zero as the stem cell population is exhausted. In reality, a single mutation may affect only the cell in which it occurs. Hence, δ may become zero once all of the progeny of that cell are dead. With $\delta > 0$, there is a transient fall in N_1 and N_2, followed by an exponential rise.

Model 6: Change in β_2, Proportion of Semidifferentiated Cells Undergoing Differentiation, Relative to β_3. A mutation causes β_2 to be reduced or increased by an amount δ ($-\beta_2 < \delta < 1 - \beta_2$). The cells that fail to die are then added to β_3, and so

$$N_1(G + 1) = 2(\beta_3 + \delta)N_1(G)t_0/t_1 + 2\alpha_2 N_0(G), \quad [18]$$

and at equilibrium

$$N_1(G) = \frac{2\alpha_2 N_0(G)}{[1 - 2(\beta_3 + \delta)t_0/t_1]}. \quad [19]$$

There is no equilibrium and there is exponential growth in cell numbers when $2(\beta_3 + \delta)t_0/t_1 > 1$, again setting lower limits on δ for exponential growth, as in the case of a decrease in the proportion, β_1, of cells undergoing PCD (Model **4**). N_0 does not change. N_2 changes with changes in N_1 as it would in a normal population. Hence, the model shows that the effects on semidifferentiated cells of changing β_2 are again to make equilibrium less likely, but not necessarily to abolish it.

Model 7: Effects of Proliferative Advantage on Cell Differentiation Models. For the sake of completeness, we shall consider what happens when a proliferative advantage is superimposed on the normal model of cell differentiation and PCD.

Assume first that cells in the F_1 population gain a proliferative advantage w and still differentiate into F_2 cells. Then

$$N_2(G + 1) = 2\beta_2(1 + w)N_1(G)t_0/t_1 + (1 - \gamma t_0/t_2)N_2(G), \quad [20]$$

and at equilibrium

$$N_2(G) = \frac{2\beta_2(1 + w)N_1(G)t_2}{\gamma t_1}. \quad [21]$$

Hence, this proliferative advantage does not overcome apoptosis to cause exponential tumor growth, although it does increase the number of cells at equilibrium by $(1 + w)$, perhaps a small quantity. Similarly, if cells in F_0 gained the proliferative advantage w' and differentiated into F1 cells,

$$N_1(G + 1) = 2\beta_3 N_1(G)t_0/t_1 + 2\alpha_2 N_0(G)(1 + w'), \quad [22]$$

and at equilibrium

$$N_1(G) = \frac{2\alpha_2 N_0(G)(1 + w')}{1 - 2\beta_3 t_0/t_1}. \quad [23]$$

Hence, the conditions for equilibrium are unchanged from the normal population, although the number of cells at any time is increased by $(1 + w')$. Without PCD and differentiation, cell numbers in each generation would have been given by $(1 + w')^G$.

Consider now the situation in which the cells with the proliferative advantage do not differentiate. For the F_0 population,

$$N_0(G + 1) = 2\alpha_3(1 + w'')N_0(G). \quad [24]$$

There is no equilibrium, and the situation is formally very similar to Eq. **16** in Model **5**, above with exponential growth. The effects are nearly identical, with differences only in the rate of tumor growth. Similarly, for F_1, when an F_1 cell with a proliferative advantage does not differentiate,

$$N_1(G + 1) = 2\beta_3(1 + w''')N_1(G)t_0/t_1 + 2\alpha_2 N_0(G), \quad [25]$$

and at equilibrium

$$N_1(G) = \frac{2\alpha_2 N_0(G)}{[1 - 2\beta_3(1 + w''')t_0/t_1]}, \quad [26]$$

and there is no equilibrium when $2\beta_3(1 + w''')t_0/t_1 > 1$.

Hence, the effects on semidifferentiated cells are again to make equilibrium less likely but not necessarily to abolish it. The situation is formally almost identical to Eqs. **18** and **19** in Model **6** above.

The models in this section show that cell proliferation that might otherwise have been considered to lead to tumorigenesis may not do so under a situation where cells differentiate and undergo PCD. In some cases, however, proliferation may substitute for the failure of PCD or differentiation in leading to tumor growth.

Conclusions

The failure of PCD or differentiation is sometimes sufficient but is not necessary for tumorigenesis. When stem cells (F_0 here) fail to undergo PCD or to differentiate in the models above, exponential growth in cell numbers occurs. This result is intuitive: extra stem cells produce both more differentiated cells and more stem cells, which in turn produce yet more stem cells and so on. This situation is formally very similar to that of tumorigenesis via stem cell proliferation, which also can result in an exponential growth in cell numbers. These are powerful mechanisms for causing tumors to develop.

If, however, a mutation causes semidifferentiated cells (F_1 in the models) to fail to undergo PCD or to differentiate further, exponential tumor growth does not always result. Sometimes, the cell population reaches an equilibrium at higher numbers than normal; sometimes, exponential growth occurs. As long as the F_0 is at equilibrium, whether the F_1 population reaches equilibrium or shows exponential growth depends solely on whether a particular function of the parameters of cell replication exceeds some threshold value (see above).

Since the F_1 population is potentially self-renewing like the stem cell F_0 population, why does exponential growth not always occur in the former when PCD or differentiation fails? The answer is that the semidifferentiated F_1 cell population is normally only partly self-renewing: many cells arise not just from the previous generation's F_1 but also from the F_0 population. In the F_0, any increase in the number of stem cells that subsequently remain as stem cells causes exponential growth because precisely 50% of the F_0 daughters self-renew under normal circumstances. When a mutation causes the F_1 to become more than self-renewing, exponential growth does occur, just as in the F_0. In the F_1, however, a mutation must raise the number of F_1 cells giving rise to more F_1 cells to >50% from a normal value some way <50%. Therefore, many mutations acting on the F_1 population may not have a large enough effect on their own to cause exponential growth of cell numbers.

When PCD of fully differentiated cells (here F_2) fails to occur, there is no exponential growth in cell numbers. A decrease in apoptosis in the F_2 population leads only to potentially linear growth, since the F_2 cells do not themselves proliferate. Although a large decrease in the proportion of cells dying can lead to a significant growth in cell numbers, this always leads to a new equilibrium (at which the linear rate of

increase in cell numbers is balanced by the numbers dying per generation). It has been assumed above that a fixed proportion of cells die per generation, since this is arguably the most realistic scenario. If a constant (or maximum) number of cells dies per generation, failure of PCD in the F_2 could lead to linear growth with no equilibrium.

How might the predicted exponential growth and growth to an equilibrium be reflected in observations of tumors? In both cases, cells gain a selective advantage over normal cells, the essential component of tumorigenesis. Either type of growth may therefore be causal in initiating or promoting tumor growth, no doubt in conjunction with other mutations. Mutations need not occur in, or have their effects on, stem cells for tumors to grow. It is tempting to suggest that growth to equilibrium, which clearly tends to be slower than exponential growth, is more likely to occur in benign lesions or premalignant states such as cervical intraepithelial neoplasia. Malignancies may be more likely to show the rapid, exponential growth. Perhaps the tendency of some tissues to produce benign tumors and others to produce malignant tumors reflects the relative susceptibilities of stem cells in each tissue to mutations affecting PCD and/or the replicative parameters of each tissue. An increase in cell numbers to a new equilibrium is also consistent with the plateaux of growth and lag phases observed in some solid tumors (12). Once equilibrium has been attained, there may be a delay until a new mutation occurs to cause further growth, whether exponential or to another equilibrium; but each such new state reached increases the chance that a further mutation, advantageous to tumorigenesis, will lead to exponential growth and to malignancy.

The interaction between failure of PCD and tumor growth via cell proliferation has been commented on by many workers (13–15). For example, a mutation may cause a cell to proliferate to an excessive degree and thereby initiate tumorigenesis. If, however, all of the daughters of the mutant cell undergo PCD, tumorigenesis will be rapidly aborted. It has been suggested, therefore, that a cell must acquire both a proliferation mutation and a mutation preventing PCD if a tumor is to develop. Two such mutations can undoubtedly give a cell a greater selective advantage than just one mutation—and hence will tend to be observed together in many tumors—but the models suggest that both mutations are not necessary for tumor growth.

There are two reasons for this. First, some models of the failure of PCD and differentiation are very similar to those of proliferation. A single mutation can both overcome PCD and lead to exponential growth in cell numbers. For example, the proportion of cells undergoing PCD in the stem cell (F_0) generation may fall and thereby lead, in effect, to a proliferative advantage for the cells involved. Therefore, failure of PCD may sometimes be sufficient for tumorigenesis: there is no need to invoke a separate proliferative advantage. Second, it is true that if all differentiated cells undergo PCD in each and every generation and the exponential growth of F_0 or F_1 cells does not continue indefinitely, two mutations are necessary. This is, however, an extreme case and unlikely to apply in reality, since apoptosis probably includes some stochastic element and is unlikely to apply uniformly to a population of differentiated cells.

The models have been deliberately imprecise as to the type of mutation that might lead to the differences proposed in the proportion of cells undergoing PCD or differentiation. While the roles of genes such as *TP53, MYC*, and *BCL2* in PCD are being discovered (5, 16–21), the pathways that lead to apoptosis are likely to be complex and varied. There is little virtue in incorporating such complexity into models of tumorigenesis. It is sufficient to ensure that the assumptions made in the models are not known to be unrealistic in any important feature. Further developments of the model will include simulations of stochastic effects and incorporation of successive mutational steps, with mutation rates and selective parameters chosen from appropriate distributions.

In summary, the models presented illustrate the possible roles played in tumorigenesis by the failure of PCD in particular and of differentiation in general. Perhaps most importantly, they show that the incorporation of differentiation and PCD into genetic models of tumor growth can have profound effects. Tumors may, for example, grow to some equilibrium rather than the continuing exponential growth that might have been expected. Which path the tumor follows depends on functions of growth parameters, on the values of those parameters and on the stage of differentiation of the cell that fails to apoptose or differentiate. Small changes in these parameters, causing them to exceed or fall below threshold values, can profoundly alter tumor behavior. We believe, therefore, that these models provide effective explanations for the development of benign tumors and premalignant growths as well as the stepwise, gradual growth of many tumors, with sometimes very long apparent lag phases.

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