Diversity of haematopoietic stem cell growth from a uniform population of cells

THE stochastic model for haematopoietic stem cell differentiation[1] with the addition of a maturation pathway and a 'resting' G_0 phase provides unifying explanations for three characteristics of the growth of colonies developing in the spleens of irradiated mice that have been injected with haematopoietic cells (Fig. 1).

First, it predicts that a homogeneous cell population can produce a high proportion of visible colonies that disappear between the 7th and 12th day of colony growth, with a nearly equal number of cells appearing during this interval; however, this requires the proviso that the self-renewal probability of colony-forming cells does not increase from the normal steady-state value until a few days after the irradiation and injection of haematopoietic cells. Second, the tiny transient erythroid colonies (devoid of colony-forming cells) observed in erythropoietically stimulated irradiated mice[2] can be predicted if there is a reduction in the threshold size for visibility, due to the increased contrast of more mature erythroid cells. Thus, these colonies may be produced by the same homogeneous population of multipotential cells producing the late-disappearing and late-appearing colonies, but such colonies are those which run out of stem cells early, 60% predicted theoretically[3], and so are not usually seen. Third, the late appearance of colonies (with higher colony-forming content) observed after injection of bone marrow cells from donor mice that have been treated with the anticancer agent 5-fluorouracil[4] can be predicted by a 15% reduction in the number of maturing cell generations.

Physical cell separation shows unambiguously that spleen colony-forming cells are heterogeneous, but does not argue forcibly for the existence of a separate subpopulation of committed spleen colony-forming cells comparable in number to the multipotential population. We therefore suggest that the experimental results of Magli *et al.*[5] fail to provide satisfactory evidence for the widely accepted claim "that the spleen colony method measures pluripotential stem cells only where macroscopic colonies are scored 11 days or later". Moreover, up to 80% of 10-day colonies contain colony-forming cells[6]. Thus, there is no compelling reason for scoring colonies later than 10 days, with the attendant increase in such technical problems as fewer countable colonies, linearity of colony counts with number of cells injected due to their larger size, possible migration from other sites, appearance of endogenous colonies and more frequent difficulty with animal survival. Moreover, these data do not necessarily require a reassessment of work carried out over the past 20 years using this important assay for haematopoietic stem cells.

This highly oversimplistic model, with quite uncontroversial assumptions, demonstrates that results previously explained by postulating separate stem cell populations or selective drug sensitivity can be explained elegantly by a homogeneous population responding in different ways. Unless the minimum number of assumptions required to explain particular experimental data is found, it is unlikely that we will acquire a better understanding of the nature and regulation of haematopoietic stem cells. Our unifying model reverses the tendency to postulate a new cell population each time a new type of experimental result is observed. Elsewhere it is shown that these conclusions support the specific mechanism for the regulation of the number of haematopoietic stem cells proposed previously[7], that they provide a straightforward explanation for the 'puzzle' concerning stem cell regeneration from cells with impaired self-renewal capacity[8] and that the equation[3] for calculating self-renewal has been misapplied.

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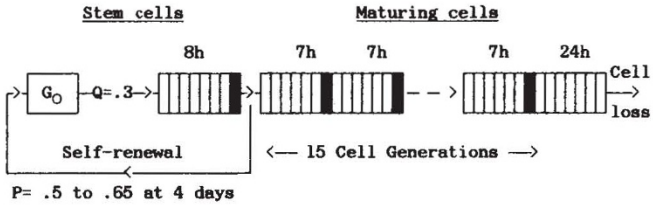
N. M. BLACKETT
E. NECAS
E. FRINDEL
Laboratoire de Cinétique Célulaire (U250), Institut Gustave Roussy, Villejuif, France



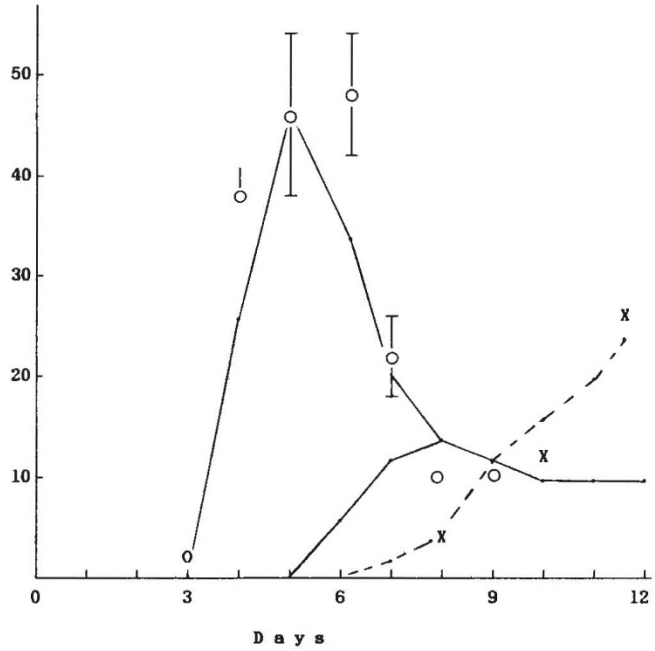


Fig. 1 Model with parameters predicting 40% loss of 7-day colonies and the number of colonies visible during growth. *a*, normal bone marrow (●); *b*, a reduction in the number of cell generations from 15 to 13 compared with experimental results, × ref. 4; *c*, a threefold reduction in the diameter threshold for colony visibility compared with experimental results, ○ ref. 2.

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ISCOVE *ET AL.* REPLY—Blackett, Necas and Frindel show that our observations[1] of transient and late-appearing spleen colonies can be simulated by assuming an initially homogeneous colony-forming cell population and making an *ad hoc* adjustment of self-renewal probability. They conclude that our data do not prove that only the later colonies arise from pluripotential cells capable of extensive proliferation, and therefore claim that "there is no compelling reason for scoring colonies later than 10 days . . .".