$\#773$

Hemopoiesis during Thiamphenicol Treatment. II. A Theoretical Analysis Shows Consistency of New Data with a Previously Hypothesized Model of Stem Cell Regulation

Markus Loeffler,[1] Birgit Bungart,[1] Henk Goris,[2] Stephan Schmitz,[1] and Willem Nijhof[2]

[1]Medical Clinic I, University of Cologne, LFI Ebene 02, Joseph Stelzmann-Strasse 9, D-5000 Cologne 41, Federal Republic of Germany; and [2]Laboratory of Physiological Chemistry, University of Groningen, Bloemsingel 10, 9712 KZ Groningen, The Netherlands

Abstract. In a recent theoretical model of stem cell regulation, specific quantitative assumptions were made about an in vivo feedback process from erythroid and granuloid precursor cell stages to the spleen colony-forming units (CFU-S), erythroid burst-forming units (BFU-E), and granulocyte–macrophage colony-forming units (CFU-GM). Utilizing specific effects of the antibiotic thiamphenicol (TAP), new experiments have been performed to challenge this model. Here these data are treated in an analysis that implies three steps. First, model assumptions about TAP toxicity are justified. The toxic TAP effects on erythroid and granuloid precursors are quantified as a continuous reduction of the normal amplification coefficient for CFU-E (down to $\frac{1}{250}$), proerythroblasts, basophilic erythroblasts, and proliferating granuloid precursors (down to $\frac{1}{4}$). Second, the original model predictions for the behavior of CFU-S, CFU-GM, and BFU-E are compared with the corresponding data. Third, discrepancies are discussed and it is demonstrated that adjustment of one single parameter resolves most of them. Thus one can quantiatively explain the experimental results for CFU-S, BFU-E, and CFU-GM by an activation of the regulatory process postulated: the decline in erythroid (and granuloid) cell numbers enhances the cycling of CFU-S while their self-renewal probability is reduced; consequently CFU-S numbers decline; as more cells differentiate towards BFU-E and CFU-GM per unit time the cell numbers of these cell stages increase. Thus the new data on stem cell behavior during TAP treatment support the hypothesis of a feedback from erythroid and granuloid precursors to the stem cells.

Key words: Hemopoietic stem cells — Regulation — Thiamphenicol — Mathematical model

Quantitative modeling of theoretical biological concepts can contribute to the understanding of complex biological systems for several reasons. First, the assumptions implied in the concept have to be stated very clearly in order to allow a translation into mathematical terms (and vice versa). Second, the contribution of individual parameters in a complex system becomes traceable. Third, one can examine whether the concept can give a consistent interpretation of different types of experiments that are otherwise not directly com-

Address reprint requests to: Dr. M. Loeffler, Medical Clinic I, University of Cologne, LFI Ebene 02, Joseph-Stelzmann-Strasse 9, D-5000 Cologne 41, FRG.

parable (different doses or manipulations). Fourth, specific predictions can be formulated that can be tested by new experiments. The logic of model testing implies that a fit of data to predictions does not prove the truth of the model but only supports further application. If a fit is not convincing one has to examine whether minor or major assumptions of the model have to be corrected. The latter implies rejection of the original model.

We have recently suggested a theoretical model of stem cell regulation in which specific quantitative assumptions were made about the feedback controls involved [1–5] (see Wichmann et al. [6] for review). The model scheme is summarized in Figure 1. The basic concept is that three interrelated feedback loops govern hemopoietic proliferation and differentiation (feedback I, II, and III).

Recent data suggested that thiamphenicol (TAP) is a suitable drug to challenge the feedback II hypothesis (feedback from erythroid and granuloid precursor stages to the stem cells) due to its effects on specific cell stages of the erythroid and granuloid lineages [7–9]. The essential assumptions on feedback II can be summarized as follows: two distinct parameters of the primitive hemopoietic cells are regulated — the cyclic activity (of spleen colony-forming units [CFU-S], erythroid burst-forming units [BFU-E], and granulocyte–macrophage colony-forming units [CFU-GM]) and the self-renewal (of CFU-S). The specific values of both parameters depend in a complicated (nonlinear) way on the actual cell numbers of CFU-S, granuloid (G), and erythroid (E) precursors. This concept of mutual interactions of CFU-S, E, and G numbers on cyclic activity (of CFU-S, BFU-E, and CFU-GM) and self-renewal (of CFU-S) leads to a consistent explanation of hemopoietic behavior under such different circumstances as acute and continuous irradiation (CFU-S, E, and G are all reduced), red cell transfusion and posthypoxia (E is reduced, but CFU-S and G slightly increase), and different kinds of anemia (E is increased, G slightly decreases, and CFU-S remains unchanged) (see *Discussion*) [2, 6].

On the basis of this concept it was predicted that TAP should lead to an increase in cyclic activity (of CFU-S, BFU-E, and CFU-GM), to a decrease in CFU-S number, and to an increased cell differentiation. To obtain an independent test for this model new and more comprehensive experiments were undertaken and reported by Goris et al. [10]. A part of these data serves the quantification of the toxic effects of TAP and is used as input to the model. Based on this, model

predictions are generated. They are compared with the remaining part of the data and the necessity of modifications of the model is discussed.

Methods

Standard model and definitions

The mathematical model used is a combination of a previous stem cell model [2] and a recent model of mature erythropoiesis including erythroid colony-forming units (CFU-E), erythroblasts, reticulocytes, and erythrocytes [3]. All technical details are given in these references. Therefore, only a brief summary and some definitions are given.

The mathematical description is based on the compartment concept. Each compartment refers to one biologically identifiable cell stage. It is characterized by a transit time "T," a cyclic activity "a," and either an amplification coefficient "Z" (in nonself-sustaining compartments) or a self-renewal probability "p." The compartments are linked by cell flows. Changes in a compartment size Y with time t are described by ordinary differential equations of the type:

$$d/dt\ Y = C^{in} \cdot Z - a \cdot Y/T \qquad (1)$$

with C^{in} representing the cell input rate. The amplification coefficient Z appears in the input term and the output is proportional to the compartment content (first order kinetics). This allows for simple numerical calculations but implies a biologically unrealistic random age structure within such a compartment. The model is designed to represent the "total" hemopoietic cell production of the marrow and spleen. The corresponding experimental values can be obtained by pooling the splenic and femoral cell numbers, assuming that one femur represents 6% of the whole bone marrow [2, vol II, Appendix 2].

Control processes. The essential model assumptions about the three interrelated feedback loops are: a) The CFU-S exert self control on their self-renewal and cyclic activity (feedback I). b) The E and G progenitors and precursors also have a regulatory impact on the self-renewal of CFU-S and on the cyclic activity of CFU-S, BFU-E, and CFU-GM (feedback II). c) Amplification in the highly proliferating erythroid cell stages (CFU-S, pro-, baso-, and polychromatic erythroblasts) is controlled by the demand for red blood cells that is mediated by erythropoietin (EPO) (feedback III).

Cyclic activity (proliferative fraction). In the model cyclic activity of a cell stage is defined as the fraction of cells in cell cycle. It is denoted by "a." Experimentally this parameter correlates (e.g., to the measurements) by the tritiated thymidine ([^{3}H]TdR) suicide technique. An experimental [^{3}H]TdR suicide of 60% is the maximum value frequently found and therefore corresponds to a = 1.0 [2, vol I, chapter 4; 10]. In the model variable cyclic activities are only assumed for the three cell stages CFU-S (denoted as a_S), BFU-E (a_{BE}), and CFU-GM (a_{CG}). Their normal steady state values are assumed to be 0.15, 0.33, and 0.33, respectively. This implies that the cyclic activity can be increased 6.6-fold for CFU-S and three-fold for BFU-E and CFU-GM. a_S, a_{BE}, and a_{CG} are monotonously decreasing functions of CFU-S (feedback I), and of E and G numbers (feedback II) [2, chapter 4].

Self-renewal probability. Self-renewal is defined as the property of CFU-S to maintain, after cell division, the same characteristics as the mother cells. It is quantified by the self-renewal probability "p." If all daughter cells keep the properties of the mother stem cells, p = 1. If all daughter cells change to a different cell type, p = 0. A steady state is characterized by p = 0.5. In constantly growing cell populations p can be quantified experimentally by reseeding assays [11, 12]. For dynamically changing growth situations no experimental assay is available. In the model p can vary between 0.6 and 0.4. It is assumed that p is a monotonously decreasing function of CFU-S (feedback I) and a monotonously increasing function of E and G numbers (feedback II) [2, chapter 4].

Amplification. Amplification is defined as the increase in cell numbers by cell division in transient (not self-sustaining) cell stages.
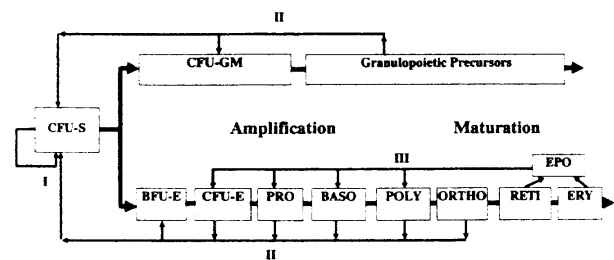
Fig. 1. Structure of the model. In this compartment model the different cell stages are connected by cell fluxes. Granulopoiesis and erythropoiesis descend from stem cells. Thrombopoiesis is neglected. The basic concept is that three interrelated feedback loops govern hemopoietic proliferation, amplification, and differentiation (see text). *Feedback I:* self control of stem cells. *Feedback II:* feedback from erythroid (E) and granuloid (G) precursor cell stages to the CFU-S, BFU-E, and CFU-GM. *Feedback III:* feedback from mature cell stages (reticulocytes and erythrocytes) to the proliferating erythroid cell stages (CFU-E, proerythroblasts, basophilic and polychromatic erythroblasts). As progenitor we define BFU-E, CFU-GM, CFU-E and as precursors we define the morphologically recognizable E and G cells.

Amplification of a lineage or a cell stage can be quantified by the amplification coefficient "Z," which is the number of descendants being produced per progeny (Z = output of cells/input of cells). Z can be related to the average number of cell divisions "n" taking place, thus $Z = 2^n$. For CFU-E and proliferating erythroblasts Z is not constant but EPO-dependent (feedback III).

Simulation of the effect of TAP

For model simulations of TAP effects three basic assumptions are made: first, TAP acts instantaneously. Second, TAP reduces the amplification coefficient Z of CFU-E, erythroblasts, and granuloid precursors by a certain factor (that has to be determined). Third, TAP equally affects young and old cells in a compartment. The last assumption is not in agreement with formula (1), where a manipulation of Z affects only the newly entering cells. To adjust for this, compartments are subdivided in N subcompartments Y_i each of the type given in formula (1) with a $Z_i = Z^{1/N}$ and $T_i = T/N$ and $Y = SUM\ (Y_i)$. For practical purposes N = 10 is used for each compartment affected by TAP. The numerical calculations are performed with the parameters of the standard average mouse model given previously [2,3].

Results

The analysis proceeds in three steps. First, one has to justify quantitative assumptions about TAP toxicity and the way to describe them in the standard model. Second, the predictions of the standard model are examined for cell stages on which TAP is not assumed to act directly. They are compared to the data. Third, requirements to modify the standard model are investigated.

Description and quantification of TAP toxicity

To estimate the degree of TAP toxicity on different cell stages, model simulations are used. Four model scenarios are shown in Figure 2 that differ in the reduction of the normal amplification coefficient of CFU-E, proerythroblasts, and basophilic and polychromatic erythroblasts. For CFU-E a reasonable match of model curves and experimental data can
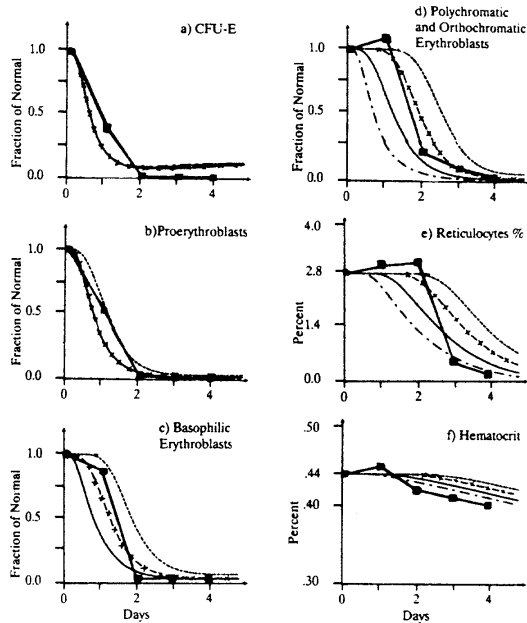
Fig. 2. Model scenarios to describe and quantify TAP toxicity on erythropoiesis. Curves of different model scenarios are compared with experimental data (■) for the "total" cell stage that is obtained by adding the bone marrow and spleen contents measured in Goris et al. [10]. Scenarios: A (– – –), B (– × – × –), C (———), and D (– · – · –). Model curves and experimental data are expressed as fraction of their respective normal values. In the model the toxicity of TAP on each cell stage is expressed in terms of reduction of the normal amplification coefficient to the corresponding fraction given in *Table 1*. The model scenarios A–D give almost identical curves for CFU-E, B–D for proerythroblasts, and C–D for basophilic erythroblasts.
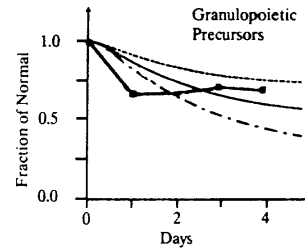

Fig. 3. Model scenarios to describe and quantify TAP toxicity on granulopiesis. In three scenarios a reduction of the normal amplification coefficient of granulopoietic precursors was assumed to a fraction of $\frac{1}{2}$ (– – –), $\frac{1}{4}$ (———), and $\frac{1}{16}$ (– · – · –). They are compared with data (■) for the total cell number (see *Fig. 2*).

Table 1. Fraction to which the normal amplification coefficient is reduced

Cell stage	Scenario			
	A	B	C	D
CFU-E	$\frac{1}{250}$	$\frac{1}{250}$	$\frac{1}{250}$	$\frac{1}{250}$
Proerythroblasts	—	$\frac{1}{4}$	$\frac{1}{4}$	$\frac{1}{4}$
Basophilic erythroblasts	—	—	$\frac{1}{4}$	$\frac{1}{4}$
Polychromatic erythroblasts	—	—	—	$\frac{1}{4}$

only be obtained if the amplification coefficient is reduced to about $\frac{1}{250}$ of the normal value for the entire 4-day period (Fig. 2a) (implying an effective amplification coefficient Z of $^{32}\!/_{250}$ = 0.128 [for a definition of Z see formula (1) in the *Methods* section]). Assuming a selective effect on the CFU-E alone implies that the subsequent compartments exhibit a sequential decline due to reduced cell influx (upper curves, scenario A, Fig. 2b–f). However, this scenario can be rejected because the more mature cell stages decline later in the model than actually observed (Fig. 2d–f). Likewise scenario D can be excluded, in which all erythroblast cell stages are assumed to be affected. The decline in mature cell stages of the model is much faster than actually observed (lowest curve, Fig. 2d and e). In scenario B (crossed lines) a toxic effect of TAP is assumed on the CFU-E and proerythroblasts with a reduction of Z to $\frac{1}{250}$ of normal at the CFU-E and to $\frac{1}{4}$ of normal at

the proerythroblast stage. In scenario C (full lines) an additional effect on basophilic erythroblasts is assumed. Both scenarios B and C are sufficient to explain the data of most cell stages except for the hematocrit, which in all model scenarios decreases too slowly.

A similar analysis can be undertaken to quantify and localize the toxicity of TAP on granulopoiesis. Three scenarios are shown in Figure 3. One can conclude that TAP reduces the amplification coefficient to about $\frac{1}{4}$–$\frac{1}{2}$ of normal at the level of morphologically recognizable granuloid precursors.

Based on this analysis, it is justified to assume in the subsequent model simulations that TAP is toxic to CFU-E, pro- and basophilic erythroblasts, and morphologically recognizable granuloid precursors, reducing their amplification coefficient to $\frac{1}{250}$, $\frac{1}{4}$, $\frac{1}{4}$, and $\frac{1}{4}$ of normal, respectively. Taken together the total erythroid amplification is reduced by a factor of 1000. No (negligible) toxic TAP effects are assumed for CFU-S, BFU-E, CFU-GM, and polychromatic erythroblasts.

Stem cell behavior under TAP treatment (standard model)

Having justified these assumptions about the TAP effect on specific erythroid and granuloid cell stages, the standard model predicts the behavior for CFU-S, BFU-E, and CFU-GM on the basis of feedback II. Figure 4 shows the corresponding model curves for the CFU-S, BFU-E, and CFU-GM. These curves originate in the following way: the TAP-induced depletion of the erythroid and granuloid precursors leads via feedback II to two effects. First, the feedback induces an activation of the cyclic activity, which increases in the model from 0.15 to 1.0 for CFU-S (Fig. 4c) and from 0.33 to 1.0 for BFU-E (a_{BE}) and CFU-GM (a_{CG}) (full curves, Fig. 4f). Second, the feedback causes a decrease of the self-renewal probability of the CFU-S (Fig. 4b). Both effects lead to an increased cell flux out of the CFU-S compartment towards differentiation. The CFU-S cell numbers decline (Fig. 4a) while an increased number of cells enters into the progenitor compartments per unit time, enlarging the cell flux through the BFU-E and CFU-GM about twofold. Due to the high turnover (high a_{BE} and a_{CG}) in these model compartments, this increased cell flux, however, is largely masked and only appears as a slight increase in BFU-E and CFU-GM numbers (full lines, Fig. 4d and e).

The comparison of the standard model curves (full lines)

with the experimental data shows a qualitatively correct pattern (decline in CFU-S, increase in BFU-E and CFU-GM, and increase in CFU-S cycling). But quantitatively, the increase of the BFU-E and the CFU-GM numbers is not sufficiently large compared with the experimental data (Fig. 4d and e). On the other hand Figure 4f shows a clear discrepancy between the cyclic activity of BFU-E and CFU-GM in the standard model (full line, increasing) and the actual [³H]TdR kill in the data (almost constant in the marrow and the spleen). In particular, the control values of untreated C57bl/6 animals already exhibit a higher cyclic activity than assumed in the standard average mouse model (day 0, Fig. 4f).

Stem cell behavior under TAP treatment (modified model)

Based on the experimental evidence provided by Goris et al. [10], it is justified to introduce a modification into the standard model with respect to the variability of the cyclic activity of BFU-E and CFU-GM. It has to be assumed that BFU-E and CFU-GM exhibit a constant and high cyclic activity (at least in this particular case). Therefore the values are chosen as $a_{BE} = a_{CG} = 0.66 = const$. This corresponds to the median 40% [³H]TdR kill found in marrow and spleen [10] (Fig. 4f). Using this as the only modification, the corresponding model curves (dashed lines) now show much clearer increases of the BFU-E and CFU-GM numbers (Fig. 4d and e) to about 200% of normal. This compares with the magnitude of change actually measured.

Thus, adjusting the standard model only for the one parameter of normal cyclic activity of BFU-E and CFU-GM (which was measured) generates a satisfactory explanation of CFU-S, BFU-E, and CFU-GM cell numbers and of CFU-S cycling under the application of TAP.

Discussion

It was the major objective of this experimental–theoretical cooperation to subject the model hypothesis of a feedback II to a test with a new experiment and to compare the data to the predictions.

The first part in the analysis concerns the quantification and description of TAP toxicity in model terms. It is concluded that TAP reduces the normal amplification process at the stage of CFU-E, proerythroblasts, basophilic erythroblasts, and morphologically recognizable granuloid precursors. Erythroid amplification in total appears to be reduced by a factor of at least 1000 continuously during the 4-day period. This implies that on average 1 cell entering a CFU-E produces only 1 erythrocyte instead of 1000 as normal. In granulopoiesis the amplification is reduced by a factor of about two to four.

In previous papers [7–9] it was suggested that a specific block at the BFU-E-to-CFU-E boundary is an important TAP effect. The present analysis confirms that the CFU-E is indeed the most immature erythroid cell stage affected. The reduction of the normal amplification coefficient to an effective Z below 1 (Z = 0.128) implies a strong effect not only on proliferation but also on differentiation that can be interpreted as a block (i.e., out of ten cells entering the
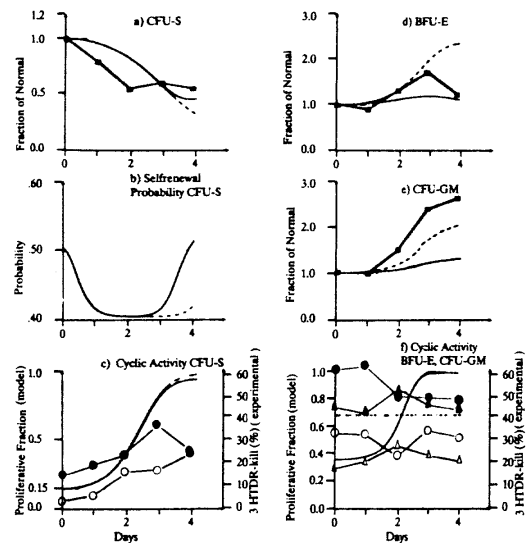

Fig. 4. Model analysis of the CFU-S, BFU-E, and CFU-GM behavior during 4 days of TAP treatment. Simulations are performed assuming scenario C for erythropoietic precursors and a reduction of amplification to ¼ for granulopoietic precursors (see full lines in Figs. 2 and 3). Full lines: standard model; Dashed lines: proliferative fraction of BFU-E and CFU-GM fixed at 0.66. They are compared to data for CFU-S, BFU-E, and CFU-GM numbers (■) for the "total" cell number (see Fig. 2.). For the [³H]TdR kills, experimental data are shown separately for the bone marrow CFU-S (●, c), BFU-E (●, f), CFU-GM (▲, f), and the spleen CFU-S (○, c), BFU-E (○, f), and CFU-GM (△, f).

CFU-E stage on average only one cell manages its way to the next stage). However, the analysis also shows that additional effects on erythroblast cell stages are necessary to explain the data satisfactorily.

Biologically, several mechanisms could be responsible for reduction of amplification: 1) there could be ineffective hemopoiesis with ongoing cell division but subsequent cell death; 2) there could be a block of differentiation and proliferation; and 3) there could be a deletion of some or all of the cell divisions with ongoing differentiation. At the present stage a decision between these possibilities is not possible. Therefore it is preferable to use the term "reduction of the amplification coefficient," which is neutral with respect to the biological mechanisms implied.

The second part in the analysis concerns the behavior of CFU-S, BFU-E, and CFU-GM. It is concluded that the data on CFU-S, BFU-E, and CFU-GM can be explained on the basis of a compensatory regulatory process acting from E and G cell stages to the more primitive cell stages (feedback II). The modification introduced to the standard model ($a_{BE} = a_{CG} = const.$) can be considered as a minor model modification because the general concept of a feedback II is not changed, in particular with respect to CFU-S regulation. It is very likely that the difference has to do with strain differences.

Comparing model predictions quantitatively with the data, one should bear in mind that at least three important simplifications have been made in the model, because appropriate data are not available. First, the model is designed for the total body hemopoiesis in mice. Therefore the model is

compared with the data pooled from bone marrow and spleen. This appears justified because marrow and spleen exhibit a similar pattern of reactions during TAP treatment with some minor quantitative differences. Model refinements considering the conditions in marrow and spleen separately are in preparation.

Second, the description of granulopoiesis is not very realistic, because no CSF effects are assumed to act on CFU-GM. It is possible that during the 4-day application of the antibiotic TAP a specific stimulation of CFU-GM growth by CSFs or other granulopoietic growth factor takes place. This could explain why the CFU-GM data are higher than predicted by the model curve. This could also explain why CFU-GM increase more than BFU-E in the data. At present it is difficult to make quantitative assumptions about the in vivo feedback system governing granulopoiesis. Further experimental research seems necessary to provide data for an appropriate modeling.

Third, one may question the description of CFU-S as a homogeneous self-renewing stem cell in the light of reports on stem cell hierarchies [13–15]. However, it should be kept in mind that the present model is designed to describe the behavior of erythromyeloid stem cells on a time scale of a few days. It is not very likely that the behavior of more primitive stem cells will play a considerable role in such short-term circumstances. In addition, it was pointed out that the present description can often be interpreted as an averaged description of a heterogeneous stem cell population [2, 6]. Nevertheless the future availability of data on mixed lineage colony-forming units (CFU-GEMM) and pre-CFU-S might make it possible to abandon the simplistic description of a homogeneous stem cell population.

It is certainly possible to imagine other ways to explain the data on CFU-S, BFU-E, and CFU-GM during TAP application than by feedback II activation. TAP could specifically stimulate amplification of BFU-E and CFU-GM, although this would be surprising with respect to the effects on more mature cells. Alternatively, maturation could be slowed down, leading to an accumulation of BFU-E and CFU-GM, but the constant cyclic activities argue against this. Changes in microenvironmental conditions have not been investigated and cannot be ruled out. Due to these possibilities it would certainly be unjustified to take the TAP data alone as sufficient evidence for the feedback II process. However, they supplement a series of other experiments that all fit into this concept, thus suggesting consistency and comprehensiveness. This is illustrated by four examples.

First, this feedback II concept explains that during continuous whole body irradiation peripheral hemopoiesis can be maintained at almost normal levels although stem cell numbers stay at low values. This is due to a massive stimulation of stem cells to produce more differentiating progeny [2, vol I, chapter 9].

Second, in the case of erythropoietic suppression by red cell transfusion as well as posthypoxic conditions, significant increases in CFU-S numbers can be attributed to a feedback II activation. At the beginning the reduction in erythroid precursors E has two effects: an increase in CFU-S cycling and a decrease of self-renewal. A wave of differentiation results, enlarging BFU-E and CFU-GM numbers (note that up to here the explanation for the TAP effects is identical). As

a consequence the number of G increases. Due to the strong role of G in feedback II this leads to a change of the self-renewal of CFU-S, which then start to increase. Thus, after several days CFU-S, BFU-E, and CFU-GM numbers are all elevated [2, vol II, chapters 2 and 4]. There is a specific difference between these circumstances and those found during TAP treatment. In the TAP experiments the number of G does not increase but remains below normal. This causes CFU-S to continue their decline.

Third, in the case of erythropoietic stimulation by hypoxia, the increase in E numbers leads to a suppression of CFU-S cyclic activity, resulting in a lower differentiation rate visible in a decline of BFU-E, CFU-GM, and G that is in agreement with the data [2, vol I, chapter 15].

As a final example, experiments with iron 55 need to be mentioned. The isotope is incorporated into heme-synthesizing cells and kills them. A decline in CFU-S numbers to 50% within 1 day was observed. This was interpreted by Reincke et al. [16] and Reincke and Cronkite [17] as evidence for an intramedullary feedback. A recent model analysis of their data qualitatively supported the conclusions, but also demonstrated that the CFU-S decline seen is more pronounced and more rapid than should be expected from the model. Reincke et al. [16] and Reincke and Cronkite [17] found about a 50% reduction in ortho- and polychromatic erythroblasts and no changes in granuloid precursors. The effects during TAP treatment are much more pronounced on erythroblasts and also affect granulopoiesis. Nevertheless, with regard to the different mode of action, the data on iron-55 and TAP application can be considered as supplementary.

The feedback II concept has gradually developed in the past and it is difficult to find its root. Lajtha and Schofield [18] and Lajtha [19] suggested a feedback from erythroid cells to the CFU-S. Reincke et al. [16] and Reincke and Cronkite [17] argued along similar lines based on the iron-55 experiments mentioned. Blackett and Botnick [20] argued for a regulatory impact of granulopoiesis on CFU-S based on data obtained after cyclophosphamide treatment and irradiation. Their hypothesis, however, lacked some precision [21]. Some authors described such feedback concepts in terms of mathematical models [22–24], but usually considered only one cell lineage, either erythropoiesis or granulopoiesis. Consequently we consider the proposed model concept as more comprehensive because it allows the description of the behavior for various combinations of CFU-S, E, and G numbers under different situations. The model assumptions on how CFU-S, E, and G numbers quantitatively influence cyclic activity and self-renewal in vivo have up to now resisted a series of tests and can therefore be recommended for further application.

Clearly a rigorous proof of feedback II requires a molecular basis of this process. Little is known at the moment about possible control factors and the role of the microenvironment under such circumstances, but it may be important to focus interest on this topic. A better understanding of feedback II may also be interesting because of its potential clinical implications. Because, at the moment, growth factors of feedback III (EPO, GM-CSF, and G-CSF) enter clinical practice, it is interesting to know whether manipulations of the precursor cell numbers by growth factors can indirectly influence stem cells.

References

1. Loeffler M, Wichmann HE (1980) A comprehensive mathematical model of stem cell proliferation which reproduces most of the published experimental results. Cell Tissue Kinet 13:543
2. Wichmann HE, Loeffler M (eds) (1985) Mathematical modeling of cell proliferation: stem cell regulation in hemopoiesis, vols I and II. Boca Raton FL: CRC Press
3. Loeffler M, Pantel K, Wulff H, Wichmann HE (1989) A mathematical model of erythropoiesis in mice and rats. Part 1: structure of the model. Cell Tissue Kinet (in press)
4. Wichmann HE, Loeffler M, Pantel K, Wulff H (1989) A mathematical model of erythropoiesis in mice and rats. Part 2: stimulated erythropoiesis. Cell Tissue Kinet (in press)
5. Wulff H, Wichmann HE, Pantel K, Loeffler M (1989) A mathematical model of erythropoiesis in mice and rats. Part 3: suppressed erythropoiesis. Cell Tissue Kinet (in press)
6. Wichman HE, Loeffler M, Schmitz S (1988) A concept of hemopoietic regulation and its biomathematical realisation. Blood Cells 14:411
7. Nijhof W, Wierenga PK, Kardaun S (1977) The effect of thiamphenicol on the production of immature red blood cells under anemic conditions. Br J Haematol 36:29
8. Nijhof W, Wierenga PK (1980) Thiamphenicol as an inhibitor of early red cell differentiation. Hoppe-Seyler's Z Physiol Chem 361:1371
9. Nijhof W, Wierenga PK (1982) The regeneration of stem cells after a bone marrow depression induced by thiamphenicol. Exp Hematol 10:36
10. Goris H, Loeffler M, Bungart B, Schmitz S, Nijhof W (1989) Hemopoiesis during thiamphenicol treatment. I. Stimulation of stem cells during eradication of intermediate cell stages. Exp Hematol 17:957–961
11. Siminovitch L, McCulloch EA, Till JE (1963) The distribution of colony forming cells among spleen colonies. J Cell Comp Physiol 62:327
12. Vogel H, Niewisch H, Matioli G (1968) The self renewal probability of hemopoietic stem cells. J Cell Physiol 72:221
13. Nakahata T, Ogawa M (1982) Identification in culture of a class of hemopoietic colony-forming units with extensive capabilities to self-renewal and generate multipotential hemopoietic colonies. Proc Natl Acad Sci USA 79:3843
14. Rosendahl M, Adam J (1984) Haemopoiesis by clonal succession? Blood Cells 10:473
15. Fujimori Y, Hara H, Taguchi T, Kitamura Y, Nagai K (1988) Clonal nature of murine hemopoietic blast cell colonies. Blut 57:169
16. Reincke U, Brookhoff D, Burlington N, Cronkite EP (1980) Forced differentiation of CFU-S by iron-55 erythrocytocide. Blood Cells 5:351
17. Reincke U, Cronkite EP (1985) Iron 55 experiments—experimental results: evidence for intramedullary stem cell regulation. In: Wichmann HE, Loeffler M (eds) Mathematical modeling of cell proliferation: stem cell regulation in hemopoiesis, vol II. Boca Raton FL: CRC Press, p 79
18. Lajtha LG, Schofield R (1971) Regulation of stem cell renewal and differentiation: possible significance in aging. Adv Geront Res 3:131
19. Lajtha LG (1971) Kinetics of the haemopoietic stem cells. Haematologia 5:355
20. Blackett NM, Botnick LE (1981) A regulatory mechanism for the number of pluropotential haemopoietic progenitor cells in mice. Blood Cells 7:417
21. Wichmann HE, Loeffler M (1982) A solution to the controversy on stem cell regulation [letter to the editor]. Blood Cells 8:461
22. Rubinow SI, Lobowitz LJ (1975) A mathematical model of neutrophil production and control in normal man. J Math Biol 1:187
23. Aarnaes E (1978) Mathematical model of the control of red blood cell production. In: Valleron AJ, MacDonald PDM (eds) Biomathematics and cell kinetics, 1st edn. Elsevier/North Holland Biomedical Press, p 309
24. Steinbach KH, Raffler H, Pabst G, Fliedner TM (1980) A mathematical model of canine granulocytopoiesis. J Math Biol 10:1