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Bifurcation and Bistability in a Model of Hematopoietic Regulation*

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Abstract. Stem cells and their relationship with mature tissues are of increasing interest in the biomedical sciences, but the dynamics of stem cell/tissue interactions are not well understood. We give a generic stem cell/tissue model and examine the dynamics of a specific case of this, namely, a four-compartment model of blood cell production and regulation. We apply the findings to cyclical neutropenia and periodic chronic myelogenous leukemia, two diseases of the blood production system. We track the position of the Hopf bifurcation believed to give rise to blood cell oscillations in these diseases. Results account for the variable success of granulocyte-colony stimulating factor, a common treatment for cyclical neutropenia, in reducing oscillations. The model displays bistability of periodic solutions, presenting the opportunity to stabilize the system through a temporary perturbation that induces switching between locally stable solutions. It is found that oscillations can be suppressed by properly timed pulses of increased amplification in the platelet line. The medical interpretation of this result is that temporary administration of thrombopoietin may suppress the oscillations. Though it is neutrophil oscillations that characterize both diseases clinically, and though it is probably a destabilization in the neutrophil line that initiates the oscillations, pulses in the neutrophil amplification do not effectively suppress oscillations.

Key words. mathematical biology, delay differential equations, bifurcation, coupled oscillations, stem cells

AMS subject classifications. 37G10, 34C14, 34C23, 37L15

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1. Introduction.

Stem cells are of great interest in the biomedical sciences due to their ability to develop into all of the cell types in the body. It has been suggested that therapies using pluripotent stem cells could present cures for currently incurable degenerative diseases such as Alzheimer's and diabetes, among others. Recently, new sources for stem cells in amniotic fluid [7] and fibroblast culture [24] have emerged; these findings could alleviate some of the political debate about the ethics of stem cell research. The spatial and temporal dynamics of stem cell proliferation and differentiation are not well understood, which presents the opportunity for mathematicians to contribute to an exciting and rapidly growing field with many open problems.

Stem cells, by definition, are pluripotential, and many stem cells (adult stem cells) are self-renewing. In other words, they can differentiate and then mature into diverse tissue types, and their population is able to sustain itself through proliferation. Consider a generic situation as follows: a small population of stem cells is capable of differentiating and then,

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through a series of cell divisions, giving rise to N distinct tissue types. The rate at which stem cells differentiate into a given cell type is a function of the existing populations of cells. Furthermore, it takes some time, typically a number of days, for the complete maturation process.

A generic model along these lines is

$$
\begin{aligned}
&\dot{q} = -kqh(q) + \mu q_\tau h(t_\tau) - q\sum_{i=1}^{N} h_i(X_1,\dots,X_n),\\
&\frac{\partial x_i}{\partial t} + \frac{\partial x_i}{\partial a} = f_i(x_1,\dots,x_n),\\
&x_i(0,t) = a_i q_{\tau_i} h_i(X_{1_{\tau_i}},\dots,X_{n_{\tau_i}}),\\
&x_i(a,0) = x_i^0(a),
\end{aligned}
\tag{1}
$$

where

$$X_i(t) = \int_0^\infty x_i(a,t)da.$$

Here, q represents quiescent stem cells, namely, those that are not undergoing mitosis (proliferation). They can enter a proliferative phase, at rate $kh(q)$; they then return a time τ later having been multiplied by some factor $\mu > k$. We use the notation $q_\tau \equiv q(t-\tau)$ for delays. They may also differentiate into tissue types $i = 1,\dots,N$. In (1) the regulation of stem cell differentiation into each tissue type is a function of the total tissue numbers X_i. The quiescent stem cells q therefore have a loss term at rate $h_i(X_1,\dots,X_N)$ for each tissue. The tissue populations themselves are given by $x_i(a,t)$, where a represents the time since maturation and t is time.

A salient feature of stem cell/tissue interactions is given in the first boundary condition of (1): the factors a_i represent "amplification" resulting from the many stages of cell division between the stem cell compartment and the fully mature tissues. The number of mature cells, X_i, is typically several orders of magnitude larger than the number of stem cells; i.e., the a_i are large. The tissue numbers are coupled to each other not only through their interactions (given by f_i) but by their delayed common origin in a small pool of stem cells.

The dynamics of models such as (1) are in general difficult to determine. Furthermore, the tissue population dynamics may depend not only on time and age, as given here, but on spatial properties as well. However, to the extent possible, it is desirable to understand to what extent the stem cell dynamics are independent of the mature tissues, the strength of effective coupling between tissues, the resilience of the system to increased loss of stem or tissue cells, and of course the steady state(s), periodic solutions, and bifurcation structure of the system.

Perhaps the simplest version of a model that includes stem cells as well as several fully differentiated tissues is the hematopoietic (blood-producing) system. Here, the tissue types are the circulating blood cells and the stem cells are the hematopoietic stem cells in the bone marrow. Since blood circulates through the body on a time scale of minutes, on time scales relevant to stem cell differentiation (which takes days) there are no spatial effects to speak of. Furthermore, the feedback loops by which blood cell numbers are regulated are comparatively

well known and simple, and we can model differentiation as $h_i(X_1, \ldots, X_n) = h_i(X_i)$ so that the rate of entry into each cell lineage is a function of mature numbers in that lineage only.

Though in this context it is relatively simple, the hematological system demonstrates interesting observed dynamics. In several hematopoietic diseases, blood cell numbers oscillate significantly, with the same period of oscillation occurring in the neutrophils (white blood cells), platelets, and sometimes reticulocytes (red blood cell precursors). In cyclical neutropenia (CN), neutrophil numbers reach dangerously low levels, oscillating with a period of 19–21 days in humans and 11–16 days in dogs. Longer periods of up to 50 days have been observed [15]. Platelet levels oscillate around their mean value with the same period. In periodic chronic myelogenous leukemia (PCML), leukocyte levels oscillate far above normal values with very long periods, ranging from 40–80 days [11].

Previous modeling efforts and traditional biological research have made progress in understanding the dynamics of these diseases, but the precise nature and origins of the oscillations remain disputed. This is in part because the dynamics of the hematopoietic stem cells have not been well characterized. These are located inside the bone marrow, and so are comparatively inaccessible. So, unlike the circulating blood cells, good time series data for the hematopoietic stem cells (HSCs) are unavailable.

Bernard, Bélair, and Mackey [1] presented a mathematical model that coupled the HSCs and circulating neutrophil population dynamics. Oscillations arose from a Hopf bifurcation in the HSC compartment, and were consistent with some, but not all, observed features of neutrophil oscillations in CN. Colijn and Mackey [5] presented a model of blood cell production that included the HSCs, neutrophils, platelets, and erythrocytes, and found parameters that were most important in fitting model simulations to data.

In section 2 we analyze a version of (1) analogous to that given in [5]. We perform bifurcation analysis (section 3) with respect to the parameters that are most critical in CN, and compare the hematopoietic stem cell compartment alone with the full model. In section 3.3, for a point in parameter space characteristic of treated CN, we find three locally stable solutions: two periodic branches and the steady state. This allows the exploration in section 4 of several methods to perturb the system from an oscillating branch to the steady-state branch.

2. The model.

A nondimensional model of the hematopoietic production system is given by

$$
\begin{aligned}
\frac{dq}{dt} &= -qb_1h_q(q) + b_1\mu_1q_1h_q(q_1) - q\left\{b_2h_n(n) + b_3h_p(p) + b_4h_r(r)\right\},\\
\frac{dn}{dt} &= -\gamma_n n + a_n b_2 q_{\tau_{nm}} h_n(n_{\tau_{nm}}),\\
\frac{dp}{dt} &= -\gamma_p p + a_p b_3\left\{q_{\tau_{pm}}h_p(p_{\tau_{pm}}) - \mu_3 q_{\tau_{psum}}h_p(p_{\tau_{psum}})\right\},\\
\frac{dr}{dt} &= -\gamma_r r + a_r b_4\left\{q_{\tau_{rm}}h_r(r_{\tau_{rm}}) - \mu_4 q_{\tau_{rsum}}h_r(r_{\tau_{rsum}})\right\},
\end{aligned}
\tag{2}
$$

where q, n, r, and p are nondimensional stem cells, neutrophils, erythrocytes, and platelets, respectively. Subscripts indicate delays: $q_1 = q(t-1)$ and so on.

The functions h_q, h_n, h_r, and h_p are Hill functions given by

$$h_q = \frac{1}{1+q^4}, \quad h_n = \frac{\theta_1}{\theta_1+n}, \quad h_r = \frac{1}{1+r^{6.96}}, \quad h_p = \frac{1}{1+p^{1.29}},$$

where the values of the exponents were determined by previous modeling efforts in the respective cell lines [22, 18, 1]. This model is a nondimensional version of that given in [6].

As in (1), quiescent pluripotential HSCs q can leave the quiescent compartment and enter the proliferating compartment (at a rate $b_1 h_q(q)$). After a proliferation time τ_s, which in these units is 1, they re-enter the quiescent compartment having been multiplied by 2 and lost at a rate γ_s during the time for which they were proliferating ($\mu_1 = 2e^{-\tau_s \gamma_s}$). Alternatively, they can differentiate into each of the three peripheral cell lines, at rates $h_n(n)$, $h_p(p)$, and $h_r(r)$. All of these rates are negative feedback functions. In each circulating compartment there is a random loss rate (γ_n, γ_r, γ_p). The platelets and erythrocytes have an additional loss due to senescence, resulting after integration over the maturation structure of these compartments, in losses with delays τ_{psum} and τ_{rsum}.

This model is a simplified version of (1) in several ways: the regulatory functions h_i are functions of only one tissue type, the coupling terms f_i between the tissue types are simply random loss terms with no intertissue interaction, and we have not explicitly included maturation structure. The resulting system, (2), is a set of four coupled delay-differential equations with six delays. In the appendix, a list of the parameter values and their dimensional counterparts is given in Table 1, along with a list and description of the delays in the system.

3. Bifurcation analysis.

The dynamics of the stem cell compartment have been found to be critical in modeling CN and PCML [17, 14, 1, 6]. The stem cell parameters are not well constrained by current observation, and indeed, even the structure of the first equations of (1) and (2) are simplifications of a much more complex (and not fully understood) set of dynamics. However, given the parameters that must change in order for the system to mimic observed disease data [6], we have the opportunity to use bifurcation analysis to better understand the relevant dynamics of the stem cells. The parameters of interest are the stem cell death rate, which is inversely related to μ_1, the rate of re-entry to the proliferative compartment b_1, and the amount of differentiation out of the hematopoietic stem cell compartment.

The neutrophil compartment (second equation of (2)) is also important in CN and PCML. Changes in the amplification a_n in the neutrophil line are necessary to mimic CN, along with changes in the stem cells. We wish to characterize the effects of changing a_n in the comprehensive model for two reasons: we believe that reduced a_n is the central cause of CN, and G-CSF, the most common treatment for CN, raises a_n, often to above its normal steady-state value.

3.1. The stem cell compartment.

One mechanism that has been suggested for the onset of oscillations in neutropenia [15, 1, 6] is that there is an increase in apoptosis in the neutrophil line which destabilizes the stem cell compartment. Apoptosis is preprogrammed cell death; in this hypothesis, increased apoptosis affects cells during the maturation phase and fewer cells reach maturity. The negative feedback $h_n(n)$ responds, creating a greater demand for stem cells to differentiate into the neutrophil lineage. The stem cell compartment begins to oscillate and the oscillations are subsequently observed in the neutrophils, platelets, and

reticulocytes. An alternative mechanism suggested by [20] is that there is a failure in the peripheral neutrophil regulation, i.e., in the n compartment.

To investigate this further, we study the effects of parameter changes on the stem cell compartment when it is decoupled from the peripheral cell lines. This decoupled model for the stem cells alone is given by

$$\frac{dq}{dt} = -\frac{b_1}{1+q^4}q + \mu_1 \frac{b_1}{1+q_1^4}q_1 - \delta q, \tag{3}$$

where δ is a constant summarizing the total differentiation out of the stem cell compartment and into the peripheral lines. If apoptosis were to increase in any of the circulating cell lines, the negative feedback would respond by increasing the differentiation out of the stem cell compartment, corresponding to an increase in δ.

Equation (3) has a unique nontrivial positive steady-state solution q^* given by

$$q^*(1+q^{*4}) = \frac{b_1(\mu_1 - 1)}{\delta}.$$

In the range of interest here, $q^* \sim 2$ or more, so that $q^{*4} >> q^*$ and we can write $q^* \sim (\frac{\lambda_1}{\delta})^{1/5}$, where $\lambda_1 = b_1(1-\mu_1)$. Equation (3) may be linearized about its steady state, with $z = q - q^*$, to give

$$\dot{z} = \alpha z - \Lambda z_1$$

with

$$\alpha = b_1 h'(q^*) - \delta \ \sim \ \delta \frac{5-\mu_1}{\mu_1 - 1}$$

and

$$\Lambda = b_1 h'(q^*) \ \sim \ \frac{4\delta}{\mu_1 - 1}.$$

Both α and Λ are positive. The characteristic equation for the eigenvalues σ is

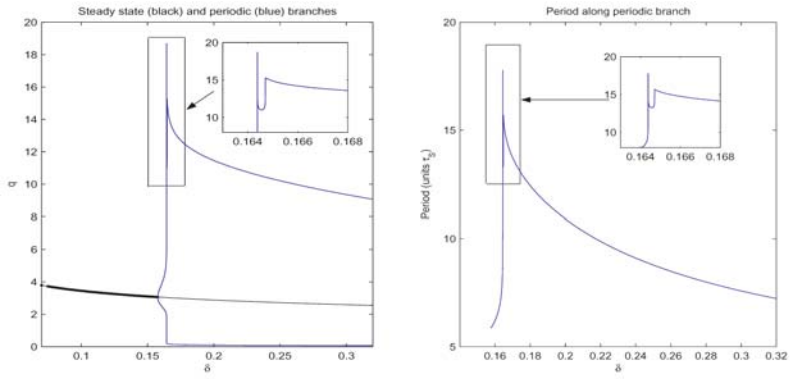
$$\sigma = \alpha - \Lambda e^{-\sigma}.$$

Letting $\sigma = i\omega$, the boundary at which instability occurs is defined by

$$\omega = \alpha \tan\omega, \quad \Lambda_1 = \frac{\omega}{\sin\omega},$$

and instability arises when $\Lambda > \Lambda_1$. As δ increases, α increases. For small α, the solution to $\omega = \alpha \tan\omega$ lies in the interval $[0, \pi/2)$, where $\Lambda_1 = \frac{\omega}{\sin\omega}$ is positive. For $\alpha > 1$, however, $\omega \in [\pi, 3\pi/2)$ and Λ_1 is negative. This ensures that $\Lambda > \Lambda_1$. We therefore expect a Hopf bifurcation when $\alpha = 1$, or $\delta = 1/6$ (when $\mu_1 = 1.6$, the normal value).

This confirms that an increase in apoptosis in one of the circulating cell lines can initiate oscillations in the stem cells. We compute the stability of the periodic solution using DDE-BIFTOOL [9, 10], and it is stable, though some of the Floquet multipliers are very close to 1 in magnitude, so that we may expect long transient approaches to the orbit. Note that α, Λ, and therefore the position of the Hopf bifurcation do not depend on b_1, the rate of re-entry to the proliferative compartment (see Figure 2). However, below a critical value of b_1 the



(a) q vs. δ (b) Period vs. δ

Figure 1. *Steady-state and periodic oscillations in the decoupled stem cell compartment. Where the steady-state branch is shown as a thick line (left side), it is stable, and where it is thin (right side), it is unstable. The maximum and minimum values of the oscillations in the quiescent (q) cells are shown for the resulting periodic branch* (a)*, and the period is shown in* (b).

stem cells cannot maintain a positive steady state and the trivial solution is the only solution to (3).

Figure 1 shows the steady-state branch of solutions (thick line: stable; thin line: unstable) and the loss of stability at the bifurcation $\delta \sim 1/6$. Compared to the steady-state value of $\delta = 1/8$, the change required to initiate oscillations is not large, and certainly not unrealistic biologically. To cause such a change in the differentiation function, the value of n need only decrease by a factor of 1/3 from the healthy value. The period along the branch is shown in Figure 1 in the right-hand plot and corresponds to the observed period of 13–50 days (4–16 units). Periods longer than those normally observed (> 10 units) occur only in a small part of the parameter space, in correspondence with observation.

We now turn to the parameters μ_1 and b_1, tracking the position of the Hopf bifurcation as b_1, δ, and μ_1 change. Figure 2 shows the results. In Figure 2(a), the "healthy" steady-state solution is in the upper stable region shown on the plot. As μ_1 decreases (the death rate in the stem cell compartment rises), oscillations begin, and then cease for values of approximately $\mu_1 < 1$. The steady-state solution in this lower region is the trivial solution, $q \equiv 0$, because if the death rate γ_S rises too much, the stem cells can no longer maintain their population. The colors shown in the oscillating regions of the plots in Figure 2 represent the period of oscillations.

Figure 2(b) shows the stability curve and periods of oscillation resulting from the Hopf bifurcation as δ increases. As the death rate in the stem cell compartment falls, μ_1 rises and the stem cells are better able to maintain a stable steady-state equilibrium in response to a need for more circulating cells (an increase in δ). In Figure 2(c), there is a threshold value of δ below which the system is always stable no matter what the value of b_1, but at higher values of δ, an increase in b_1 can destabilize the system and result in the initiation of oscillations. However, since the healthy value of b_1 (22.4) is much above this range, we conclude that oscillations are caused by an increase in δ rather than an increase in b_1.

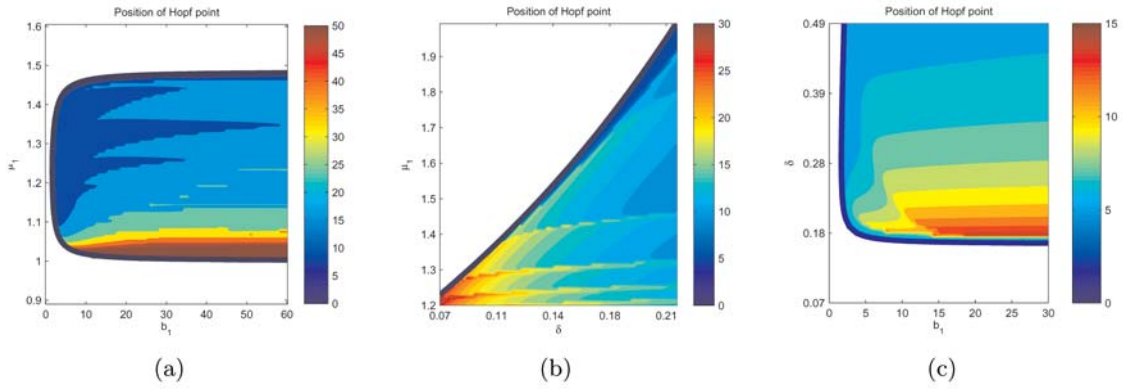


Figure 2. *Hopf bifurcation, position dependent on b_1, μ_1, and δ in the decoupled stem cell model of (3). The colored regions are the regions of Hopf instability, and their boundaries give the location of the bifurcation. The color indicates the period of oscillation.*

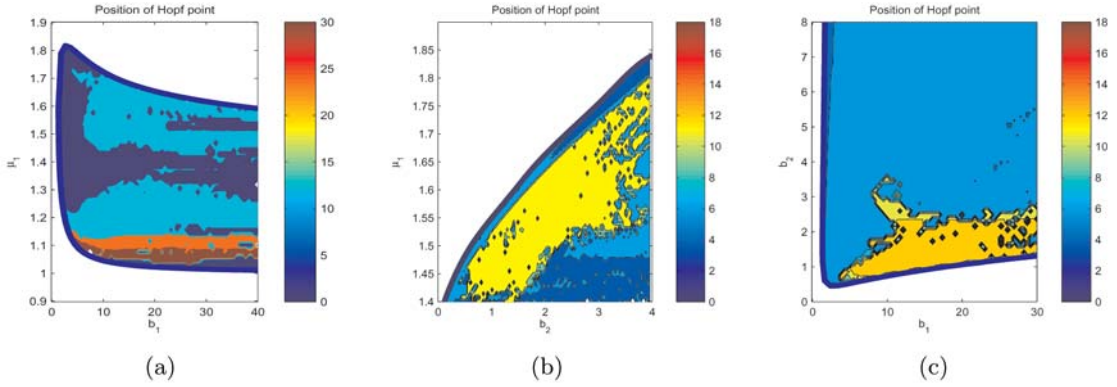


Figure 3. *Hopf bifurcation dependent on b_1, μ_1, and b_2 in the full model of (2). Thick solid lines are the location of the Hopf instability, colored regions are oscillatory, and the color indicates the period of oscillation.*

3.2. The full model: Hopf bifurcations. We now wish to find the corresponding Hopf bifurcation in the full model, if it exists, and compare its location to that in the stem cell compartment alone, under corresponding parameter changes. Due to the complexity of the model, this is done numerically. Also, δ is not constant in the full model; here, we can explicitly raise b_2 (increase output from the hematopoietic stem cells q) and/or decrease a_n. The bifurcation occurs as a_n decreases below $a_n = 21.5$, and is again supercritical. Figure 3 shows its position under the analogous parameter changes to Figure 2.

The qualitative behavior of the full model is similar to that of the stem cell compartment alone, and there are biological interpretations for the differences between the two. For example, in Figure 3(a) oscillations begin as μ_1 is decreased, and then give way to the trivial solution below $\mu_1 = 1$. In the full model there is a region in the upper-left portion of the plot where an increase in b_1 can stabilize oscillations: an increased rate of re-entry to the proliferative phase can compensate for increased stem cell apoptosis, unlike in Figure 2(a). In Figure 3(b), the concavity of the Hopf curve is reversed, but otherwise the Hopf position as μ_1 and b_2 are

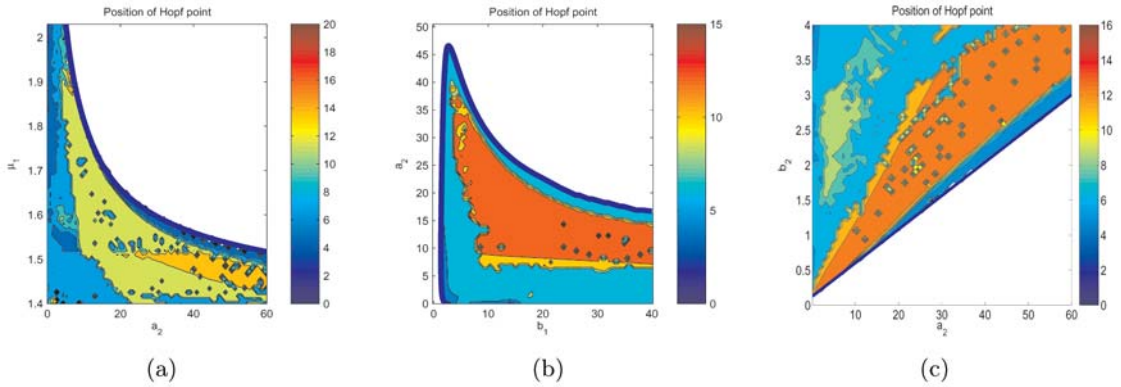


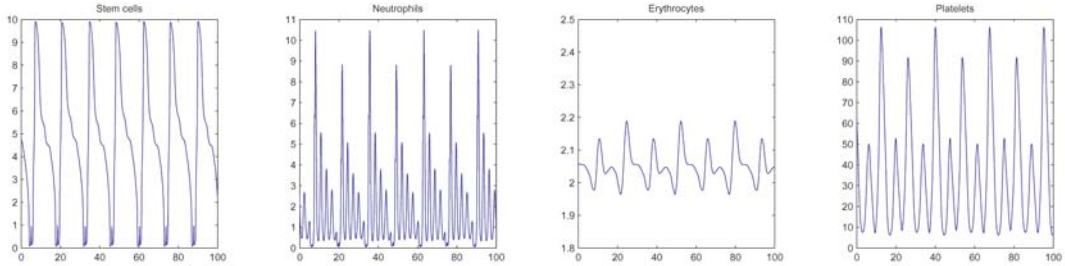
Figure 4. *Hopf position as a_n changes. Colored regions are oscillatory, and the color indicates the period of oscillation.*

changed is qualitatively the same. Figure 3(c) shows that unlike in the stem cell model alone, an increase in b_1 can again compensate for an increase in b_2 and stabilize oscillations. The full model is better able to recover from destabilizing changes than the stem cell model without the coupling to the peripheral cell lines.

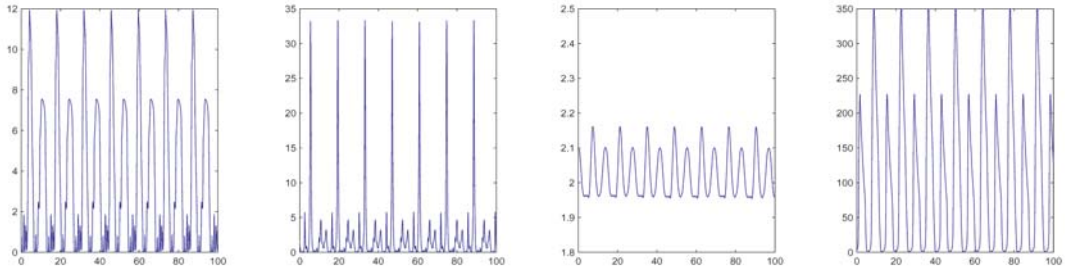
Decreasing b_2 and increasing μ_1, based on these results, are probably the two best methods of stabilizing existing oscillations. However, the most common treatment for CN is the administration of granulocyte-colony stimulating factor (G-CSF) [25, 13, 2, 19], which is known to increase neutrophil levels by reducing apoptosis in the neutrophil precursors [13], and is therefore modeled by an increase in the parameter a_n. We track the Hopf bifurcation with respect to a_n; the results are shown in Figure 4. In each plot of the figure it is clear that increasing a_n has the desired effect of stabilizing the oscillations, though significant increases may be necessary if μ_1 or b_1 is small. If they are too small, oscillations may not be stabilized at all.

In clinical data, G-CSF sometimes abolishes oscillations but may actually increase their amplitude [16]. The bifurcation analysis accounts for this. We would hypothesize that when G-CSF does not abolish oscillations, it is because there is not enough capacity for self-maintenance in the stem cells: their apoptosis rate is too high, and/or their proliferation rate is too low. Furthermore, the results shown in Figure 4(b) show that increasing b_1 may stabilize oscillations if a_2 is high enough; clinically, stem cell factor would raise b_1 and is, in fact, sometimes used to treat CN.

In this section, we have found that oscillations begin if the death rate in the stem cell compartment rises (μ_1 decreases), if there is increased differentiation out of the stem cells (b_2 increases), if there is a reduction in the proliferation rate in the stem cell compartment (b_1 decreases), or if there is increased apoptosis in the neutrophil line (a_n decreases). In some regions, an increase in b_1 may recover stability, but this recovery is not possible in the reduced model containing only the stem cells. The otherwise similar natures of the plots shown in this and the previous section lend credibility to the claim that destabilizations in the stem cell compartment are at the heart of oscillatory behavior in the hematological system.



(a) Low-amplitude branch (branch 1) with period 28 dimensionless time units



(b) High-amplitude branch (branch 2) with period 14 dimensionless time units

Figure 5. *Two locally stable periodic branches when $a_n = 58$ and all other parameters are set to their default values.*

3.3. Multistability. Not only does G-CSF not always suppress oscillations, it may even initiate them [16]. Furthermore, in a model containing only the stem cells and the neutrophils, Bernard, Bélair, and Mackey [1] found that there was a small range of a_n where there is bistability of two periodic solutions. Motivated by these results, we explore the existence of multistability when a_n is increased from its normal value (35.6) to 58. This is a simple way to mimic a patient undergoing G-CSF treatment. All the other parameters are left at the values given in Table 1; this point in parameter space will be referred to as point P.

At point P, the steady-state solution is locally stable. However, we also find two locally stable periodic orbits at P (see Figure 5). The low-amplitude orbit (Figure 5(a)) has a period of 28 units, and the orbit shown in Figure 5(b) has a period of 14 units with a higher-frequency oscillation at a period of 7 units, corresponding to the secondary bump in the stem cell profile. A wide range of simulations using different initial conditions failed to uncover any other locally stable periodic solutions.

The low-amplitude orbit is more reminiscent of treated neutropenia than the high-amplitude orbit, where neutrophil levels at maximum are more than 30 times their steady-state value (1 in these units). Also, the platelet levels in the high-amplitude branch are much higher than are observed clinically, and for more of the oscillation, than in the low-amplitude orbit.

4. Branch switching. The fact that multistability exists for this biologically reasonable choice of parameter values naturally leads to the question of how the oscillations might be stabilized; stabilization would correspond clinically to an end of disease symptoms. In this

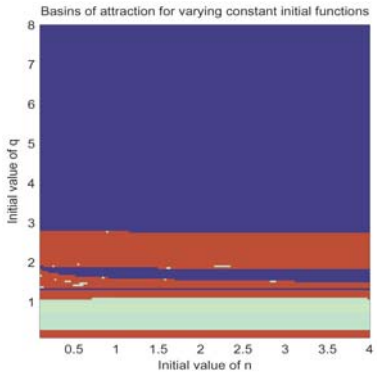


Figure 6. *Basins of attraction for varying initial q and n values. Blue corresponds to the steady-state branch, red to the low-amplitude branch, and green to the high-amplitude branch.*

section we investigate the possibility of changing the initial function to guide which behavior the system approaches, and then we examine the effects of temporary parameter changes on the system when it is oscillating on the low-amplitude branch in Figure 5. In the computations, we use a Runge–Kutta integration scheme with step sizes ranging from 0.005 to 0.05 (most commonly 0.01) dimensionless time units, in Bard Ermentrout's software *xppaut*.[1]

4.1. Effect of initial functions. Because the system in (2) has delays, it is necessary to specify initial functions for each of the four variables, during the period $[-\tau_{\max}, 0]$. We first set all variables constant on $[-\tau_{\max}, 0]$ and examine the effect of the choice of these constants. Figure 6 shows the results when only the initial values of q and n on $[-\tau_{\max}, 0]$ (the initial numbers of stem cells and neutrophils, respectively) are changed; each branch is assigned a color: blue for the stable steady-state solution, red for the low-amplitude orbit, and green for the high-amplitude orbit. The same color scheme applies to figures showing basins of attraction in this and subsequent sections. The lack of other colors (and hence of other periods) is evidence that these three solutions are the only locally stable solutions at this point in parameter space.

The most prominent feature of Figure 6 is that the initial value of n has almost no effect on the eventual behavior of the system, even when n is increased up to three times the normal value of 1. However, the initial population of stem cells has a significant effect between 0 and 4, though the value at the steady state is 3.2. This is further indication that oscillations in CN and PCML, and in the hematological system in general, are most strongly connected to the dynamics of the stem cell compartment.

This result leads to the question of what the effects of nonconstant initial functions for q, particularly periodic initial functions, would be. We next allow the initial function for q to change, leaving the initial functions for n, r, and p constant at their healthy, steady-state values. On $[-\tau_{\max}, 0]$, q is given by

$$q(t) = A_q \sin\left(\tfrac{2\pi}{T_q} t\right) + M_q, \tag{4}$$

[1] http://www.math.pitt.edu/~bard/xpp/xpp.html

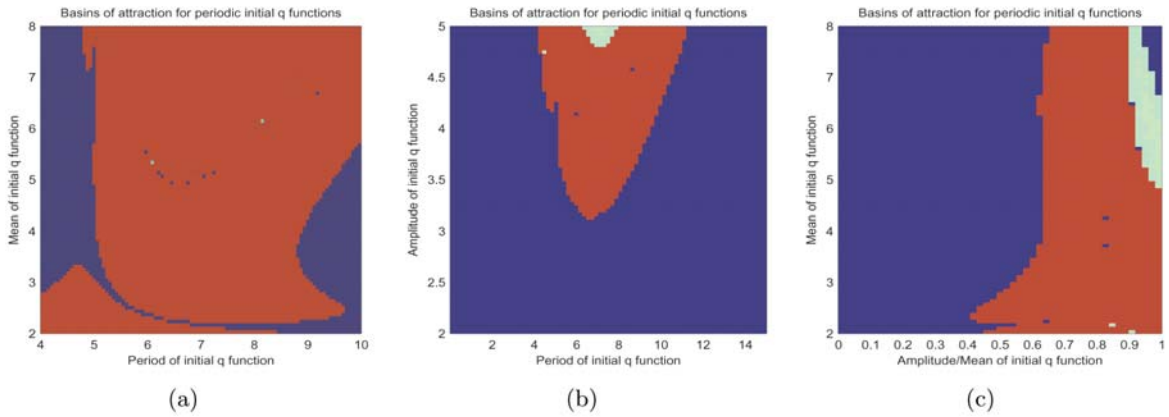


Figure 7. *Basins of attraction for various periodic initial functions. Only q is varying; the other variables are constant on $[-\tau_{\max}, 0]$. Blue is steady-state, red is the low-amplitude branch, and green is the high-amplitude branch.*

so that the three parameters defining this function are the amplitude of the oscillations, A_q, the period, T_q, and the mean, M_q. Figure 7 shows the resulting basins of attraction. In Figure 7(a), note that for most of the plotted range of periods and means the solution stabilizes to the low-amplitude branch, where in Figure 7(b) most of the solutions stabilize to the steady-state branch. Figure 7(c) shows the basins of attraction when the mean M_q and the fraction A_q/M_q are varied; note that a value of A_q/M_q greater than 1 is not possible because it would result in negative q values in the initial function for q on $[-\tau_{\max}, 0]$.

Figures 6 and 7 show the manner in which the initial function determines whether the system ultimately oscillates (mimicking CN) or stabilizes (mimicking a healthy individual). However, the stem cells, located in the bone marrow, are difficult to identify and manipulate, and any such manipulation would presumably also change several parameters in the system (and not only the values of q). It is therefore difficult to translate these results into realistic medical interventions. In the following section we investigate temporary interventions and their effect on the stability of the system.

4.2. Branch jumping by short-term parameter changes. We explore the possibility of branch jumping, i.e., perturbing the system so that it moves from one solution to another, using temporary pulses in the system parameters a_n, a_r, and a_p. These represent amplification in the neutrophil, erythrocyte, and platelet lines, respectively, and they are chosen because it is known that the administration of regulatory cytokines increases these amplification factors via the inhibition of apoptosis: G-CSF increases the neutrophil amplification, while erythropoietin and thrombopoietin have similar effects on the erythrocyte and platelet lines, respectively [3, 21, 23]. Thrombopoietin and erythropoietin are not used to treat neutropenia, because they are primilary associated with platelet and erythrocyte dynamics and neutropenia is characterized by decreased neutrophil levels. Furthermore, the administration of thrombopoietin causes an immune response [8], rendering it of limited value. However, there may be other clinical interventions which would raise the platelet amplification a_p in the manner we investigate here.

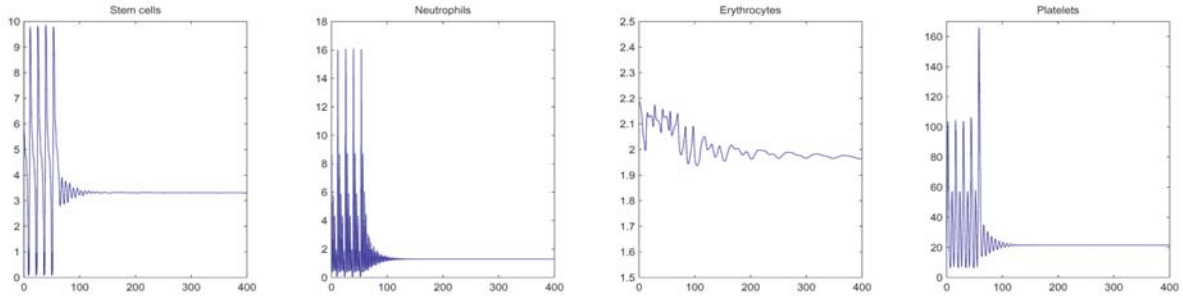


Figure 8. *Stabilization of the low-amplitude branch with a pulse in a_p of amplitude* 40 *and duration* 4.75*, mimicking thrombopoietin administration, delivered at* $t = 50$*.*

To simulate the effects of a brief administration of the relevant cytokines, we have computed the basins of attraction for parameter functions of the form

$$a_i = a_i^{\text{base}} + a_i^{\text{amp}} H(t - t_1) H(t_2 - t), \tag{5}$$

where $H(t)$ is the Heaviside function and $i \in \{n, r, p\}$. This function represents a change in the parameter a_i from a_i^{base}, a base value, to $a_i^{\text{base}} + a_i^{\text{amp}}$ during the time interval $[t_1, t_2]$. For each basin computation, we choose an initial function well within the basin of attraction of the periodic low-amplitude solution (Figure 5(a)). We define the phase to be 0 at time $t_1 = 200$ to consistently examine the effect of the phase time at which the pulse is delivered. Because each computation is performed from the same initial condition at the same parameter set, this is unambiguous. Figure 8 shows an example of a successful stabilization using a pulse of the parameter a_p, according to (5). Note that the erythrocytes are considerably slower in reaching equilibrium than the other variables, due to the long delay τ_{RS} in the erythrocyte compartment.

Because G-CSF is the most common treatment for CN, one would suspect that pulses of increased amplification a_n would have a significant effect on the qualitative behavior of the solutions. However, oscillations are rarely suppressed using pulses in a_n. Figure 9 shows the results of exploring various durations and amplitudes of the pulses. Note that the regions in which the stabilization attempt was successful (blue areas in the figure) are small, and that the amplitude of the pulse needs to be high. The normal value of a_n is 35.6, and so the values required for stabilization represent an increase by a factor of 6 or more. This would translate into a high dose of G-CSF, which would have to be precisely timed to stabilize the system.

Figure 10 shows similar basin of attraction plots for pulses in a_r while varying the amplitude, duration, and phase of the pulse. Again, while stabilization is possible using a pulse in a_r, regions where the solution stabilizes are small and the amplitude of the pulse is large compared to the healthy value of a_r (~ 1). However, the regions of stabilization are larger and the pulses do not need to be as precisely timed as those in a_n. This gives the somewhat counterintuitive result that stabilization of oscillations in CN and PCML may be more easily accomplished with erythropoietin administration than with G-CSF, though the dosage would have to be high to obtain the necessary large increases in a_r.

The most promising stabilization results we have obtained have been with pulses using

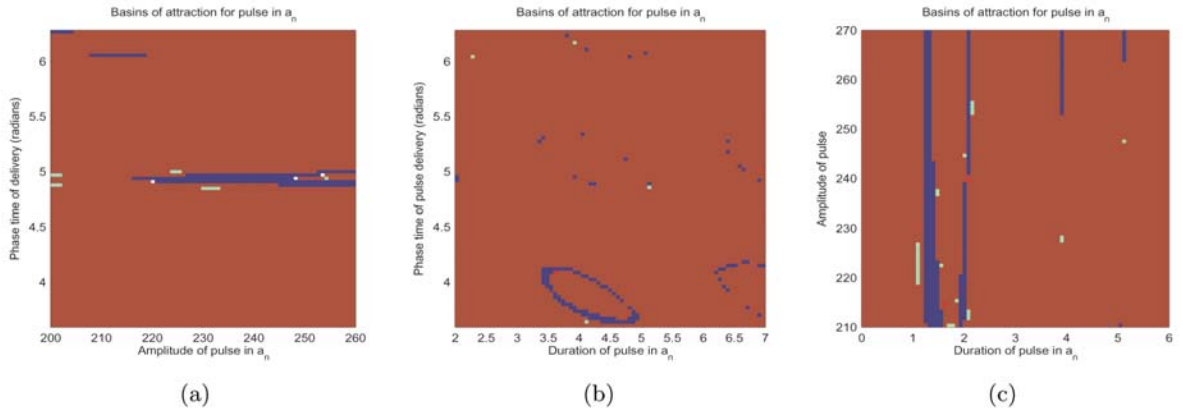


Figure 9. *Basins of attraction for an a_n pulse, simulating G-CSF administration.*

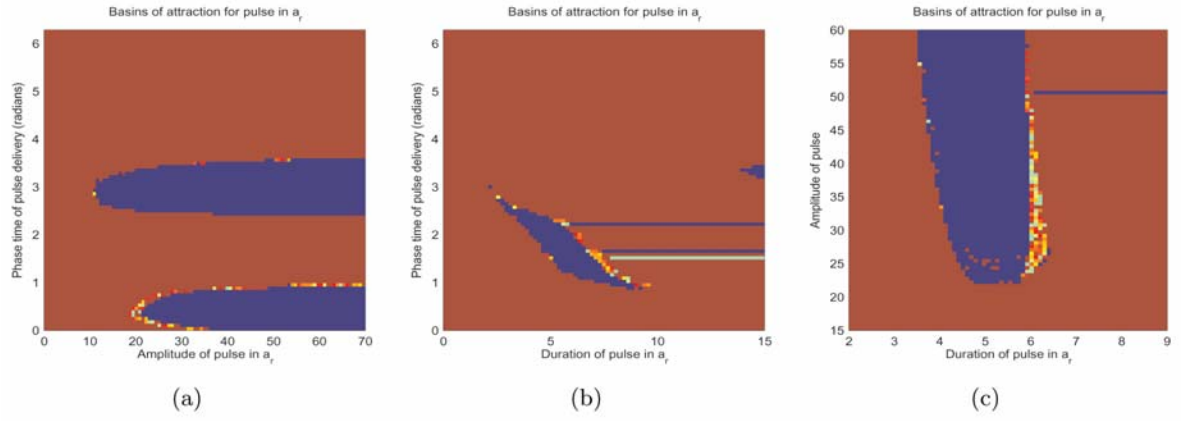


Figure 10. *Basins of attraction for pulses in a_r, simulating erythropoietin administration.*

the parameter a_p; Figure 11 shows the results. The blue regions, where the oscillations have been stabilized by the pulse, are comparatively large. This indicates that the stabilization is a robust phenomenon at this point (P) in parameter space.

The normal value of a_p is approximately 58 and the minimum additional pulse required is only about 20, so that a comparatively small and achievable pulse size is sufficient. Figure 11(a) shows clearly that some points in the oscillation are more advantageous times at which to deliver the pulse, namely, at phases 0, $\pi/2$, π, and $3\pi/2$. Figure 11(b) shows that changing the pulse duration shifts these bands. In Figure 11(c), it is interesting to note that increasing the duration of the pulse does not necessarily increase the likelihood that stabilization will occur. While it does appear that increasing the amplitude of the pulse makes stabilization more likely, it is not usually the best way to stabilize the system. Changing the duration or timing of the pulse is more effective.

With reference to (2), we can partially account for the somewhat surprising fact that perturbations to the platelets are best able to suppress oscillations. Any increase in a_n, a_p, or a_r will cause a decrease in δ, which we know from section 3.1 can stabilize the stem cell

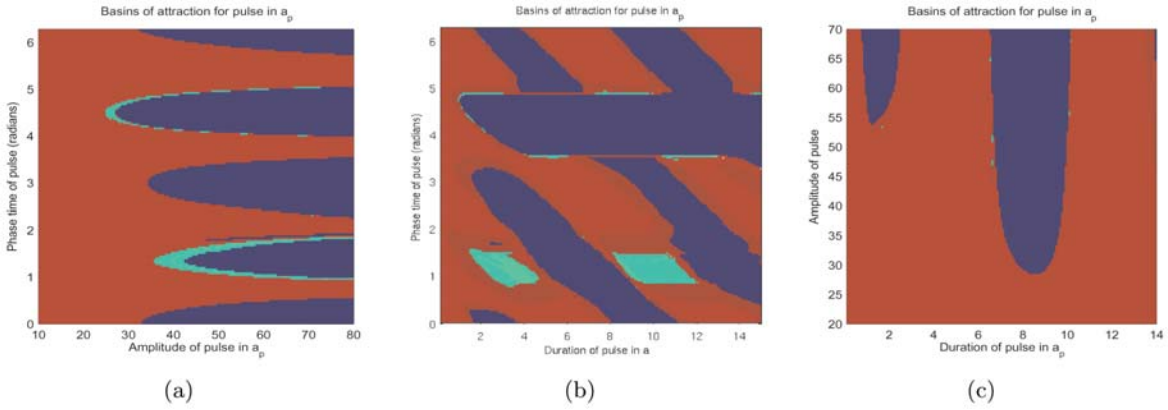


Figure 11. *Basins of attraction for pulses in a_p, simulating thrombopoietin administration.*

compartment (at least, when considered on its own). Suppose that when an amplification a_i increases, the corresponding compartment n, r, or p responds by trying to "track" the change and reach a new pseudosteady state. Then we can write

$$\begin{aligned}
\frac{\partial \delta}{\partial a_n} &= \frac{\partial \delta}{\partial n}\frac{\partial n}{\partial a_n} \approx \frac{-b_2^2\theta_1^2 q}{(\theta+n)^2(\gamma_n\theta+1)} = O(10^{-2}),\\
\frac{\partial \delta}{\partial a_p} &= \frac{\partial \delta}{\partial p}\frac{\partial p}{\partial a_p} \approx \frac{-b_3 s_p p^{s_p} q(1-\mu_3)}{(1+p^{s_p})^2(1_+ s_p)p^{s_p}} = O(10^{-3}),\\
\frac{\partial \delta}{\partial a_p} &= \frac{\partial \delta}{\partial p}\frac{\partial p}{\partial a_p} \approx \frac{-b_3 s_p p^{s_p} q(1-\mu_3)}{(1+p^{s_p})^2(1_+ s_p)p^{s_p}} = O(10^{-5}).
\end{aligned} \tag{6}$$

A change in a_p is far more effective than a change in a_r at changing δ. In addition, the time scales for the n and p equations ($1/\gamma_n$ and $1/\gamma_p$) are much faster than for the r equation, as $1/\gamma_r \sim O(10^3)$. The value of r thus does not respond quickly, and even if it did, (6) indicates that the response would not be as strong as the platelet response. While the neutrophil response time is adequate, the neutrophils are not in a pseudosteady state on the low-amplitude branch; rather, when q is high they undergo high-frequency transient oscillations (see Figure 5(a) and the discussion in [4]). They therefore do not have a consistent effect on δ when a_n is raised.

In addition, it has been found that q undergoes relaxation oscillations and therefore has slow and fast phases [12, 4]. This, together with the details of the response of p to an increase in a_p, probably accounts for the sensitivity of the results on the phase of oscillation; when q is in the lower half of the slow portion of the oscillation, decreasing δ may have more effect than when q is high.

5. Conclusions. Despite the potential complexity of stem cell/tissue models, the model analyzed here for hematopoietic production and regulation shows some interpretable dynamics. The location and behavior of the Hopf bifurcation believed to give rise to CN in the full model occurs in the stem cell model when it is decoupled from the mature tissues, suggesting a possible avenue for analysis of more complicated versions of the model given in (1). Inter-

estingly, the inclusion of the tissue types is a stabilizing influence: the full model of (2) is better able to recover from destabilizing changes than the decoupled model of (3).

The results presented here support the hypothesis that oscillations in dynamical hematological diseases are driven by oscillations in the stem cell compartment. We find that oscillations consistent with those observed for CN are initiated when an increase in demand for circulating blood cells causes the stem cell compartment to undergo a Hopf bifurcation. Our analysis accounts for the variable success of G-CSF in suppressing oscillations in CN. When G-CSF fails to suppress oscillations, this may be due to a lack of regenerative capacity in the hematopoeitic stem cells—a death rate that is too high, or too little re-entry into the proliferative compartment, or a combination. Our results also account for why stem cell factor may be a promising treatment for CN.

For G-CSF-treated CN, we find two locally stable periodic orbits and a locally stable steady state. The choice of solution depends most strongly on the initial values of the stem cells. We find that, using short-term pulses of increased amplification in any of the peripheral lines, it is possible to suppress oscillations with temporary measures that have clear medical interpretations. This is most easily accomplished with pulses of increased a_p, the amplification in the platelet line, mimicking temporary administration of thrombopoietin. While thrombopoietin may not be promising due to the initiation of an immune response that targets it, other interventions that temporarily raise platelet numbers could be considered. The timing of the pulse is important, but because the regions in parameter space where the stabilization is successful are quite large, finding an appropriate time is not difficult. These results suggest that combining G-CSF with short-term platelet-enhancing drugs may be a promising approach to abolishing oscillations. These somewhat surprising effects of tissue level perturbations on the full model may be understood in terms of their effects on the decoupled stem cell model: perturbations that would stabilize the decoupled stem cell model are more able to switch the full model from oscillatory to stable dynamics than those that would not.

Appendix.

Table 1

Definitions and typical values of the dimensionless parameters of the model. Dimensional parameters correspond to those given in [6].

Symbol	Description	Dimensional equivalent	Typical value
b_1	stem cell proliferation	$\tau_s k_0$	22.4
b_2	neutrophil differentiation factor	$\tau_s f_0$	1.1
b_3	platelet differentiation factor	$\tau_S \bar{\kappa}_P$	3.3
b_4	erythrocyte differentiation factor	$\tau_S \bar{\kappa}_R$	3.3
θ_1	Hill function parameter	θ/N^*	0.055
s_p	Hill exponent	m	1.29
s_r	Hill exponent	r	6.96
μ_1	proliferation with loss factor	$2e^{-\gamma_S \tau_S}$	1.6
μ_3	loss factor	$e^{-\gamma_P \tau_{PS}}$	0.24
μ_4	loss factor	$e^{-\gamma_R \tau_{RS}}$	0.9
γ_n	neutrophil death rate	$\tau_S \gamma_N$	6.7
γ_r	erythrocyte death rate	$\tau_S \gamma_R$	0.003
γ_p	platelet death rate	$\tau_S \gamma_P$	0.42
a_n	neutrophil amplification	$A_N \theta_2 / N_1^*$	35.63
a_p	platelet amplification	$A_P \theta_2 K_P^{1/s_p}$	58
a_r	erythrocyte amplification	$A_R \theta_2 K_R^{1/s_r}$	1.1
τ_s	stem cell proliferation time	τ_s/τ_S	1
τ_{nm}	neutrophil maturation time	τ_{NM}/τ_S	1.25
τ_{pm}	platelet maturation time	τ_{PM}/τ_S	2.5
τ_{ps}	platelet aging time to senescence	τ_{PS}/τ_S	5.9
τ_{psum}	$\tau_{pm} + \tau_{ps}$		8.4
τ_{rm}	erythrocyte maturation time	τ_{RM}/τ_S	2.1
τ_{rs}	erythrocyte aging time to senescence	τ_{RS}/τ_S	42.9
τ_{rsum}	$\tau_{rm} + \tau_{rs}$		45
q^*	stem cell steady-state value	Q^*/θ_2	3.6
n^*	neutrophil steady-state value	N^*/N^*	1
p^*	platelet steady-state value	$P^* K_p^{1/s_p}$	21
r^*	erythrocyte steady-state value	$R^* K_r^{1/s_r}$	2.2

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