Research article

A theoretical model for estimating the margination constant of leukocytes

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Abstract

Background: Blood leukocytes constitute two interchangeable sub-populations, the marginated and circulating pools. These two sub-compartments are found in normal conditions and are potentially affected by non-normal situations, either pathological or physiological. The dynamics between the compartments is governed by rate constants of margination (M) and return to circulation (R). Therefore, estimates of M and R may prove of great importance to a deeper understanding of many conditions. However, there has been a lack of formalism in order to approach such estimates. The few attempts to furnish an estimation of M and R neither rely on clearly stated models that precisely say which rate constant is under estimation nor recognize which factors may influence the estimation.

Results: The returning of the blood pools to a steady-state value after a perturbation (e.g., epinephrine injection) was modeled by a second-order differential equation. This equation has two eigenvalues, related to a fast- and to a slow-component of the dynamics. The model makes it possible to identify that these components are partitioned into three constants: R, M and S_B; where S_B is a time-invariant exit to tissues rate constant. Three examples of the computations are worked and a tentative estimation of R for mouse monocytes is presented.

Conclusions: This study establishes a firm theoretical basis for the estimation of the rate constants of the dynamics between the blood sub-compartments of white cells. It shows, for the first time, that the estimation must also take into account the exit to tissues rate constant, S_B.

Background

Blood leukocytes are found in two sub-populations constituting the circulating and the marginating pools. The elements of these two sub-populations are interchangeable, i.e., marginated leukocytes return to bloodstream and vice-versa [1–3]. Therefore, a dynamical equilibrium situation occurs, and blood cell counts should reflect the rate constants of margination and return to circulation of those cells.

Several studies addressed the relationships between the sub-populations of white cells within the blood pool.

Among other features, the ratio between the sub-populations under normal conditions [4], under altered states [4,5] and the stability of the equilibrium situation (i.e., to resume a previous value after perturbations, [e.g., [6–8]) were approached. It is presently accepted that the blood sub-pools play an important role as a white cell reservoir when increased demands supervene. Increased demands arise in both pathological and non-pathological situations, as during exercise [9,10], burns [11,12], infectious diseases [13], inflammatory processes [5,14], etc.. It is also currently recognized that some hormones acutely alter the dynamics of the blood sub-pools of white cells [6–8]. In this sense, epinephrine is known to possess a demarginating effect that lasts for less than 1 hour, and such an effect is thought to be the result of changes in the rate constants of margination and return [2,6]. Therefore, the interplay between marginating and circulating leukocytes is a relevant issue that should be taken into account in the interpretation of many results.

On the one hand, it is tacitly assumed that the rate constants of margination and return have higher values than other rate constants related to the dynamics of white blood cells (see below). On the other hand, there is no study addressing such an issue in a formal way in order to provide good estimation of these values. The aim of the present study is to provide the theoretical background to perform estimations of these rate constants, which may prove relevant for empirical studies on healthy normal situations as well as under altered states of the organisms.

Constructing the extensive model

In this sub-section, we present a model based mainly on data from monocytes and neutrophils. The model describes the dynamics of three compartments of these cells and also contains the dynamics of specific growth factors. In the next sub-section we show a reduced model from this one, which will then be employed to obtain an improved estimation for the rate constants values. The extensive model is presented to assure consistence of the analysis.

Bone marrow total rate production apparently has three components: a fixed production rate (P_1), a production rate dependent on self-regulatory factors (P_2) and a production rate dependent on inflammatory/infectious factors $(P(I))$ [3]. Therefore, the production rate is not a time independent function, and we denote it by the sum of the three terms above. Once the newly produced cell gains the bloodstream, it may marginate (rate constant M). A marginated cell may then return to circulation (rate constant R). Cells in the blood may leave to surrounding solid tissues, and they do that according to a time-independent rate constant S_B[1]. Exit to tissues is also a function of rate constants dependent on inflammatory/infectious factors

[3]. Thus, let us denote such a total exit rate constant from the circulation by $S_B + S(t,I)$. Tissue leukocytes may proliferate locally (rate constant D), produce self-regulatory factors and, eventually, die (rate constant Z) [15]. This set of empirical data suggests the scheme presented in Figure 1 and the following equation system for describing the dynamics of these cells:

$$\frac{d\phi_c}{dt} = P_1 + P(I) + P_2\alpha + R\phi_m - \left(M + S_B + S(t,I)\right)\phi_c \quad (1a)$$

$$\frac{d\phi_m}{dt} = M\phi_c - \left(R + S_B\right)\phi_m \quad (1b)$$
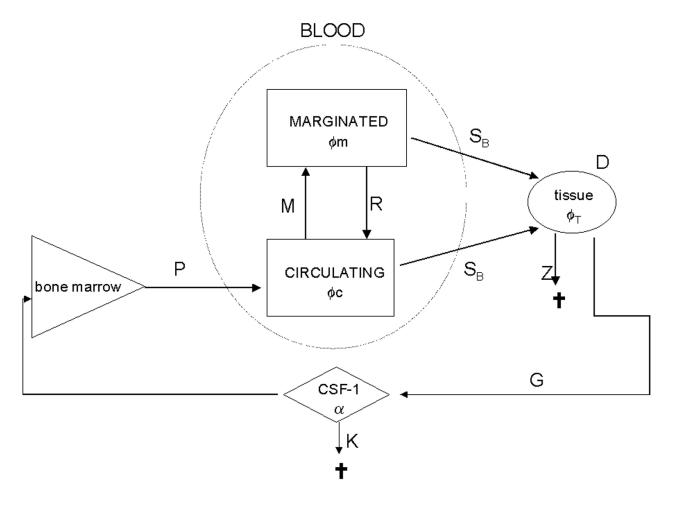
$$\frac{d\phi_T}{dt} = \left(S_c + S(t,I)\right)\phi_c + S_m\phi_m + \left(D - Z\right)\phi_T \quad (1c)$$

$$\frac{d\alpha}{dt} = G\phi_T - K\alpha \quad (1d)$$

Where α are self-regulatory factors (e.g., CSF-1) and ϕ_i refers to cells that take part in the circulating (c), marginal (m) and solid-tissues ($_T$) pools. G is a constant related to the production of self-regulatory factors, and K is the rate constant of their clearance. Notice that the exit to tissues comes both from the circulating pool, ϕ_c, and from the marginated pool, ϕ_m. Such an exit has the same rate constant, S_B, independently of the sub-pool. This partition of the total exit constant rate to tissues in two components is the basic reasoning that leads to the inference about the existence of two distinct sub-pools within the blood white cells [1]. Therefore, a cell that touches, attaches and passes through the vessel wall cannot return to the circulating pool, even though during a certain time interval this cell was marginated (semantically but not functionally). In this sense, these cells exit to the surrounding tissues coming directly from the circulating pool, and their dynamics is contemplated by the product $\phi_c S_B$ of the differential equation. Those marginated cells that can potentially return to circulation have their dynamics accounted for by the rate constant R. On the other hand, part of these marginated cells migrate to surrounding tissues and the product $\phi_m S_B$ contemplates this rate. This dynamics is also consistent to the empirical evidence that margination and diapedesis are distinct features arising from different signals and receptors [16].

Reducing to the compact model

The injection of some substances, epinephrine in particular, changes the rate constants of margination and return to circulation (M and R) for very short periods of time (e.g., see [2,6–8]). Under such a condition, the variable values change as the parameter values change. However, since the substance is rapidly metabolized, the parameters return to their previous value and then the variables

Figure 1
Schematic representation of the dynamics of white cells in the organism. The self-reproduction in tissues is represented by "D", without arrows. Crosses denote death or clearance. See text for details.

would also return to some value maintained previously to the perturbation (in the parameters). Thereby, these substances act on the system described by equations 1a-d analogously to an impulsive function [17]. The returning phase is the relaxing period of the system, and it behaves as the perturbation had occurred in the variables instead of in the parameters. During the short time interval of the relaxing period, the production rate can be considered constant, P. Therefore, equations 1a and 1b maintain no loop-connection to the other two equations (1c and 1d), and a compact system can be written as:

$$\frac{d\phi_c}{dt} = P + R\phi_m - \left(M + S_B \right)\phi_c \qquad (2a)$$

$$\frac{d\phi_m}{dt} = M\phi_c - \left(R + S_B \right)\phi_m \qquad (2b)$$

Results
Analysis of the steady-state condition
The first step in the analysis was to verify whether the extensive model allows the existence of a stable equilibrium point. An equilibrium point means a set of values of the variables (in this case, ϕ_c, ϕ_m, ϕ_T and α) that does not vary with time (if the system is left externally undisturbed). Stability is related to the behavior of the system in face of perturbations in the variables that displace them to some vicinity of an equilibrium point. The equilibrium point is said asymptotically stable if the system returns to the equilibrium point attained previously. Otherwise, the equilibrium point may be neutrally stable, if the system does not return to the equilibrium point but remains somewhere around it, or unstable, if the system leaves that vicinity away (e.g., [18]).

We verified whether the extensive model posses one asymptotically stable equilibrium point as a first step because the biological system modeled seems to behave like this. Once disturbed, it returns to a previous value. The analysis revealed the existence of one equilibrium point, and such a point is asymptotically stable (see Methods). Let * denote the value of the variable in the equilibrium point. Without loss of generality by considering the inflammatory/infectious factors absent and that ϕ_T and α are fixed (e.g., for a short time interval or changes in R or M, see Methods), the equilibrium values of the variables that we are interested in the present study are:

$$\phi_c{}^* = \frac{P}{S_B}\left(1 - \frac{M}{R + M + S_B}\right) \qquad (3a)$$

$$\phi_m{}^* = \frac{P}{S_B}\frac{M}{R + M + S_B} \qquad (3b)$$

As stated before, blood cell counts should reflect the rate constants of margination and return. Let f^* denote the ratio $\phi_m{}^*/\phi_c{}^*$ and thus:

$$f^* = \frac{\phi_m^*}{\phi_c^*} = \frac{M}{R + S_B} \qquad (4)$$

Notice that one is not able to know, directly from the ratio $\phi_m{}^*/\phi_c{}^*$, the values of R and M themselves.

The demarginated state and its return to the equilibrium condition

In a very short time interval succeeding a bolus injection of some substances (e.g., epinephrine) the parameters R and M change and return to their previous values. Therefore, the circulating and marginal pools attain different values from $\phi_c{}^*$ and $\phi_m{}^*$, respectively. Given the stability of the equilibrium point (see above) these variables return to $\phi_c{}^*$ and $\phi_m{}^*$. This returning is governed by a second-order differential equation (see Methods). Let $\tilde{\phi_c}(t)$ be the value attained above (or below) the equilibrium value $\phi_c{}^*$ by the circulating cells after the perturbation (see Methods). In short, $\tilde{\phi_c}(t) = \phi_c(t) - \phi_c^*$ (see equation 9). Therefore, taking the equilibrium value of the circulating pool as a reference value, $\tilde{\phi_c}(t)$ is the difference between this value and the value found at a time t after the beginning of the returning to the steady-state equilibrium condition. This leads to the following equation governing the fast-phase of resuming the steady-state condition:

$$\tilde{\phi_c}(t) = (\phi_{c0} - \phi_c{}^*)e^{\lambda \cdot t} \quad (5)$$

where ϕ_{c0} is the peak (or nadir) value of the circulating pool attained after the perturbation, i.e., the initial value of this variable, and λ is the fast component of the process (λ_2, see Methods equation 11b). Equation 5 can be linearized to simplify the estimation procedure. Finally, the λ of equation 5 is (see equation 11b):

$$\lambda = -(R + S_B)(f^* + 1)$$

This value of λ allows a good estimate of R during the fast phase of the returning to the equilibrium point (steady-state of the sub-pools). In the next section, we will work some examples of the application of the present model and its potential relevance.
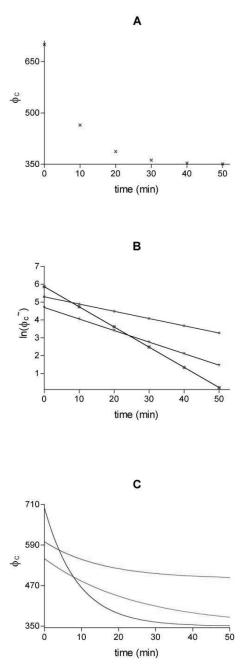
Worked examples

The present study offers the means to compute the individual values of R and M. As stated before, the ratio f^* is obtained by measuring the marginated and the circulating pools in steady-state conditions. This ratio is equal to $M/(R+S_B)$ (see equation 4). Therefore, its is important to note that the individual computation of R (and M) assumes that a series of other independent measurements were done: (a) $\phi_c{}^*$ (the steady-state value of the circulating pool); (b) $\phi_m{}^*$ (the steady-state value of the marginated pool); (c) S_B (the exit rate constant from the total blood pool to tissues). The knowledge of a general production rate value may also improve the picture, even though its is not a primary need.

The first example intends to compute the R value for mice monocytes under normal conditions. However, the paucity of adequate data prevents a true computation. Therefore, we will compute an approximate value. The other two examples are completely imaginary. They show the usefulness of the model to address changes in the rate constants that would otherwise pass unnoticed.

I. The normal condition

The computations are based on data from van Furth and Sluiter [1]. The report give $\phi_c{}^*$ = 350 monocytes/mm³, $\phi_m{}^*$ = 500 monocytes/mm³ and $S_B = 9.6 \times 10^{-4}$ min⁻¹. The f^* value is 500/350 = 1.43. An intravenous bolus injection of epinephrine caused a two-fold increase in ϕ_c after 10 minutes of the injection. Therefore, $\tilde{\phi_c}(0) = 350$ (or, $\phi_c(0) = 700$). After 60 minutes from the injection time (50 minutes after the monocytes peak), the variables resume their normal values. Notice the extremely limited temporal data. Let us suppose that we had a few more points, as illustrated in Figure 2A. From equation 5, the data could be linearized as:

$$\ln\left(\tilde{\phi_c}(T)\right) = \ln\left(\tilde{\phi_c}(0)\right) + \lambda T$$

A

B

C

Figure 2
Simulated perturbation in the blood sub-pools of a white cell type (e.g., monocytes) under three different conditions: normal (blue), tumor carrying animal (red), long-term aerobic exercising animal (green); see text for details. (A) "Raw experimental data", the "discrete" time course of ϕ_c after a bolus injection of epinephrine under the normal condition. (B) Log-transformed (linearized) data, under the three different conditions. Notice the linear relationship obtained. (C) "Raw "continuous" data" of the ϕ_c time course. Data generated in MatLab 5.3 (The MathWorks, Natick, MA) by built-in numerical integration routines of the set of differential equations of the model.

Where T is the discrete sampling time. Figure 2B shows the result of the linearization procedure. A linear regression would then result in $\lambda = -0.113$ min^{-1}. Finally, from equation 6, we know that this value must be decomposed as

$$-0.113 = -(R + S_B)(1 + f^*)$$

Thus, R = 0.0456 min^{-1}, a value 47-fold greater than S_B. In the data simulation, R = 0.045 min^{-1}, therefore the procedure seems quite adequate. This example highlights the use of the present model as well as the difference between the eigenvalue (λ, obtained directly from the data) and the constant R. Also, as far as we know, this is the first attempt to estimate the value of R (and M) under a complete and formal model.

II. Tumoral effects
Consider, as another example, that mice carrying a certain tumor type have the exit constant rate equal to the normal value ($S_B = 9.6 \times 10^{-4}$ min^{-1}). The f^* value is 1.3 and these mice have $\phi_c^* = 490$ monocytes/mm^3, $\phi_m^* = 637$ monocytes/mm^3. Obviously, production rate have increased. Notice the potential confusing situation arising from these data. Does the tumor growth alter the dynamics between the blood pools? The epinephrine experiment (or some analogue) was conducted, and $\tilde{\phi}_c(0) = 110$ (or, $\phi_c(0) = 600$). Figure 2C shows both the "normal" and the "tumoral" data. Linear regression of the log-transformed data computes $\lambda = -0.065$ min^{-1} (see Figure 2B) and thus R = 0.0273 min^{-1} since $f^* = 1.3$. In data simulation, R = 0.027 min^{-1}, again the results are in close agreement with the real parameter value. Therefore, in this constructed situation, one would be able to conclude that the rate constants of the transit between the blood pool were altered by the pathological condition. This could be an important result in tumor immunology, for example, allowing a better understanding of the pathology or devising new treatment strategies.

III. Long Distance Running Effects
Our final imaginary example is related to the effects of long sustained aerobic exercise. Consider that mice trained in this type of exercise are known to have increased production rate of monocytes. During the steady-state condition of the exercise, $\phi_c^* = 350$ monocytes/mm^3, $\phi_m^* = 700$ monocytes/mm^3, thus $f^* = 2$. S_B increased to 8.5×10^{-3} min^{-1}. An epinephrine experiment is performed during the exercise section and samples are taken during the decaying phase after the injection (the exercise section proceeds during the sampling period). The "continuos" count profile is illustrated in Figure 2C. Linear regression of the log-transformed data (see Figure 2B) computes $\lambda = -0.041$ min^{-1} and thus R = 0.0052 min^{-1} (in data simulation, R = 0.005 min^{-1}). This is a 9-fold decrease in the R value in relationship to the normal condition. On the oth-

er hand, the rate constant M decreased less than 2.5 times in relationship to its value in normal conditions (see above). This result might be important in the understanding of many immune suppression/enhancement phenomena related to certain exercise protocols. Notice that the f* value by itself do not tell anything about the change in the individual value of each rate constant.

Discussion

The compartmentalization of blood leukocytes in two sub-pools is an important feature of these cells. For example, the marginal pool may acutely function as an extra source of cells at increased demand conditions. In this sense, knowledge about the rate constants governing the transit between the sub-pools may prove relevant in studies that approach both physiological and pathological situations. In the present manuscript we provide the theoretical background to support empirical studies related to the calculation of these rate constants.

From the general perspective of an extensive model, we first showed that this model corresponds to what is experimentally found in terms of existence and stability of an equilibrium point in the variables (the sub-pools). After that, we reduced the model to a compact one, concerning only the two blood sub-pools of interest here. Within the context of the compact model, it was shown how an impulsive function (e.g., an intravenous epinephrine bolus injection) perturbing the parameters (the rate constants of margination and return to circulation) can be translated to a perturbation in the variables. Then, the returning of the variables to their equilibrium values allows the estimation of the rate constants. This was done by the use of a second-order differential equation (see Methods). From that equation, the fast-decay component was identified and the corresponding eigenvalue contains the rate constants to be calculated.

The present study demonstrates, for the first time, how to adequately and completely estimate the rate constants of margination and return to circulation of white blood cells. Notice that first-order differential equations (as equation 5) were employed before in gross estimates of these rate constants (e.g., [19]). However, the constant then computed comprised the whole eigenvalue, without discriminating its parts. In other words, its was not recognized that the computed value should be partitioned into three components: R, M and S_B.

The model also allows to establish the relationship between R and M. This relationship includes the S_B constant and the ratio between the marginal and the circulating sub-pools, f* (see equation 4). The need to take into account the f* value in the estimation of the constants is another important aspect for the first time presented.

Therefore, the model leads to a more precise evaluation on how imposed disturbances interfere with the dynamics of the blood sub-compartments and thus increase our understanding of the physiology/pathophysiology of many conditions. Two imaginary situations of this kind were constructed and analyzed as examples of the use of the present model (see "Worked Examples" II and III).

Conclusions

This study provides a complete model to approach the estimation of rate constants of margination and return to circulation of with blood cells. It shows, for the first time, how the value empirically found should be partitioned in order to adequately obtain the desired rate constants. In this sense, we were able to recognize that both an exit rate constant to tissues and the ratio between marginated and circulating cells should be taken into account in the computation procedure.

Methods

1. Equilibrium point and its stability

The equilibrium point of the system described by equations 1a-d is found by setting all the derivatives to zero and computing the corresponding values of the variables that lead to such a condition. Without loss of generality, the inflammatory/infectious factors can be taken as zero. Therefore, the equilibrium point (denoted by *) corresponds to the following values:

$$\phi_c^* = \frac{\left(P_1 + P_2\alpha^*\right)\left(S_B + R\right)}{S_B\left(R + M + S_B\right)} \qquad (7a)$$

$$\phi_m^* = \frac{M}{R + S_B}\phi_c^* \qquad (7b)$$

$$\phi_T^* = \frac{P_1 + P_2\alpha^*}{Z\text{-}D} \qquad (7c)$$

$$\alpha^* = \frac{P_1 G}{K\left(Z\text{-}D\right) - P_2 G} \qquad (7d)$$

Conditions of existence: (1) Z > D; and (2) K(Z-D) > P_2G. The first condition reflects that tissue leukocytes must die at a rate higher than their own local replication otherwise their population would increase forever. The second condition is similar to the first in the sense that it reflects that the self-stimulatory loop must be lower than the loss loop of the system.

The stability of the equilibrium point is verified by the construction of the determinant of the matrix of the coefficients of the variables of the equations of the system [17,18,20]. In order to be asymptotically stable, all the eigenvalues (roots) of the characteristic polynomial of the

matrix must have real parts lower than zero. Considering that the system has four equations, the characteristic polynomial is of the 4th-order with the following general formula:

$$\lambda^4 + a_1\lambda^3 + a_2\lambda^2 + a_3\lambda + a_4 = 0$$

where λ is an eigenvalue of the equation (note that the equation has 4 roots). The coefficients (a_i) come from the parameters of the system. To have all the eigenvalues with real part lower than zero, the Routh-Hurwitz conditions must verify. In short:

1A. All a_i must be greater than zero. This condition is true for the system.

1B. $a_1 a_2 > a_3$. This condition is true for the system.

1C. $a_3(a_1 a_2 - a_3) > a_4 a_1{}^2$. This condition is not easily verified analytically. Therefore, we performed a numerical analysis. Random values were assigned to the parameters (conditions of existence of the equilibrium point verified, see above) and the condition 1C checked (pseudo-random number generator normally distributed built-in function of MatLab 5.3, The MathWorks, Natick MA). This procedure was taken 150,000 times by a routine specifically written for it and the condition always verified true. Therefore, the equilibrium point seems asymptotically stable.

The main point in this sub-section is to realize that both $\phi_T{}^*$ and α^* are not affected by R and/or M. This is what assures the results concerning the compact model below.

2. The compact model and its second-order differential equation form

Considering that production rate and inflammatory/infectious factors would not change in a short time interval, the extensive model allows the blood pool to be treated as having no loop connection to the rest of the system (see equations 2a and 2b). The determinant of the Jacobian matrix of the compact model (i.e., the sub-system represented by equations 2a and 2b) is to set to zero:

$$\ddot{J} = \begin{vmatrix} -M - S_B - \lambda & R \\ M & -R - S_B - \lambda \end{vmatrix} = 0 \qquad (8)$$

Consider now a "push" in $\phi_c{}^*$ (and $\phi_m{}^*$) in such a way that:

$$\tilde{\phi_c}(t) = \phi_c(t) - \phi_c{}^* \qquad (9)$$

$\tilde{\phi_c}(t)$ is, therefore, the time course of the perturbation around the equilibrium point of ϕ_c. Equation 8 is homogeneous for $\tilde{\phi_c}(t)$:

$$\frac{d^2\tilde{\phi_c}}{dt^2} + (R + M + 2S_B)\frac{d\tilde{\phi_c}}{dt} + S_B(R + M + S_B)\tilde{\phi_c} = 0 \qquad (10)$$

Taking into account that $M = (R + S_B)f^*$ (see equation 4), the eigenvalues of equation 10 are:

$$\lambda_1 = \frac{-[R(1+f^*) + S_B(1+f^*) + S_B] + \sqrt{[R(1+f^*) + S_B(1+f^*) + S_B]^2 - 4S_B(R+S_B)(1+f^*)}}{2} \qquad (11a)$$

$$\lambda_2 = \frac{-[R(1+f^*) + S_B(1+f^*) + S_B] - \sqrt{[R(1+f^*) + S_B(1+f^*) + S_B]^2 - 4S_B(R+S_B)(1+f^*)}}{2} \qquad (11b)$$

Notice that: (a) both eigenvalues are pure real (this means that the equilibrium point is attained without oscillations in the variables); and (b) both eigenvalues are negative (the equilibrium point is asymptotically stable). Given that $\lambda_2 < \lambda_1$, λ_2 is the fast component of the process of resuming the equilibrium value $\phi_c{}^*$ (see sub-section "the demarginated state and its return to the equilibrium condition"). The value of λ_2 is $-(R + S_B)(f^* + 1)$. This value is presented in equation 6.

List of abbreviations

ϕ_c: the number of cells (or concentration) in the circulating pool;

ϕ_m: the number of cells (or concentration) in the marginal pool;

ϕ_T: the number of cells (or concentration) in solid tissues;

α: quantity (or concentration) of self-regulatory factors;

R: rate constant of return to circulation;

M: rate constant of margination;

S_B: rate constant of exit to tissues from the blood pool;

P: bone marrow production rate of the cells;

f^*: the ratio between the marginal and the circulating pools at steady-state condition.

Acknowledgments

References

1. van Furth R, Sluiter W: **Distribution of blood monocytes between a marginating and a circulating pool.** *J Exp Med* 1986, **163:**474-479
2. Berkow RL, Dodson RW: **Functional analysis of the marginating pool of human polymorphonuclear leukocytes.** *Am J Hematol* 1987, **24:**47-54
3. van Furth R: **Development and Distribution of Mononuclear Phagocytes.** *In: Inflammation: Basic Principles and Clinical Correlates.* 1992325-339
4. Meuret G, Hoffmann G: **Monocyte kinetc studies in normal and disease states.** *Br J Haematol* 1973, **24:**275-285
5. van Furth R, Dulk M, Mattie H: **Quantitative study on the production and kinetics of mononuclear phagocytes during an acute inflammatory reaction.** *J Exp Med* 1973, **138:**474-479
6. Fehr J, Grossmann HC: **Disparity between circulating and marginated neutrophils: evidence from studies on the granulocyte alkaline phosphatase, a marker of cell maturity.** *Am J Hematol* 1979, **7:**369-379
7. Okada Y, Kawagishi M, Kusaka M: **Neutrophil kinetics of recombinant human granulocyte colony-stimulating factor-induced neutropenia in rats.** *Life Sci* 1990, **47:**PL/65-70
8. Ulich TR, Castillo J, McNiece I, Watson L, Yin S, Andresen J: **Hematologic effects of recombinat murine granulocyte-macrophage colony-stimulating factor on the peripheral blood and bone marrow.** *Am J Phatol* 1990, **137:**369-376
9. Mackinnon LT: **Immunity in athletes.** *Int J Sports Med* 1997, **suppl 1:**s62-8
10. Pedersen BK, Hoffman-Goetz L: **Exercise and the immune system: regulation, integration and adaptation.** *Physiological Reviews* 2000, **80:**1055-1081
11. Dong YL, Abdulllah K, Yan TZ, Rutan T, Broemeling L, Robson M, Herndon DN, Waymack JP: **Effects of thermal injury and sepsis on neutrophil function.** *J Trauma* 1993, **34:**417-421
12. Santangelo S, Gamelli RL, Shankar R: **Myeloid commitment shifts toward monocytopoiesis after thermal injury and sepsis.** *Ann Surg* 2001, **233:**97-106
13. Wing EJ, Waheed A, Shadduck RK: **Changes in serum Colony-Stimulating Factor and monocytic progenitor cells during *Listeria monocytogenes* infection in mice.** *Infect Immun* 1984, **45:**180-184
14. Sluiter W, Elzenga-Claasen I, Andel AV, van Furth R: **Differences in the response of inbred mouse strain to the factor increasing monocytopoiesis.** *J Exp Med* 1984, **159:**524-536
15. Bartocci A, Mastrogiannis DS, Migliorati G, Stockert RJ, Wolkoff AW, Stanley ER: **Macrophages specifically regulate the concentration of their own growth factor in the circulation.** *Proc Natl Acad Sci USA* 1987, **84:**6179-6183
16. Colditz IG: **Margination and emigration of leukocytes.** *Surv Synth Pathol Res* 1985, **4:**44-68
17. Boyce WE, DiPrima RC: **Elementary Differential Equations and Boundary Value Problems.** *John Wiley & Sons, Inc* 1997
18. Strogatz SH: **Nonlinear Dynamics and Chaos. Reading,** *Addison-Wesley* 1994
19. Martin BA, Wright JT, Thommasen H, Hogg JC: **Effects of pulmonary blood flow on the exchange between the circulating and marginating pool of polymorphonuclear leukocytes in dog lungs.** *J Clin Invest* 1982, **69:**1277-1285
20. Murray JD: **Mathematical Biology.** *Berlin* 1989