A model of lymphocyte recirculation

Dov J. Stekel, Claire E. Parker and Martin A. Nowak

Dov Stekel, Claire Parker and Martin Nowak present a mathematical model of T-cell recirculation between blood, spleen and the lymphatic system. Comparison with classical experiments on lymphocyte recirculation and localization demonstrates that simple kinetics are sufficient to explain lymphocyte homeostasis between these organs, and that T-cell counts in blood are highly sensitive to changes in the recirculation parameters.

There is a central problem in studying the dynamics of human immunodeficiency virus (HIV) infection: the number of $CD4^+$ T cells in the blood of HIV-infected patients is commonly used as a marker for disease progression, and yet only about 2% of T cells are located in the blood of a healthy adult human, the remaining cells being principally located in the spleen and lymphatic system. It is unknown to what extent T-cell measurements in the blood of HIV-infected patients reflect the overall status of the immune system. Recent work on simian immunodeficiency virus (SIV)-infected macaque monkeys[1] and HIV-infected humans[2] has shown that the decline in the number of $CD4^+$ T cells in the blood may not be reflected in the lymphoid tissue. This raises the possibility that the decline represents some degree of redistribution of T cells from blood to lymphoid tissue, rather than purely a depletion in the total $CD4^+$ T-cell pool. It has also been suggested that, in experiments estimating the *in vivo* daily turnovers of HIV virus and $CD4^+$ T cells[3,4], the increase in total $CD4^+$ T cells observed in the blood may also be due to changes in migration that result from viral clearance[5–7].

While there has been a great deal of work published on theoretical models of the immune system, the majority of existing work has focused on the dynamic interactions between immune cells and molecules, and between the immune system and pathogens, without taking into account the complex spatial heterogeneity of the immune system. The aims of this work are to construct a minimal model of T-cell

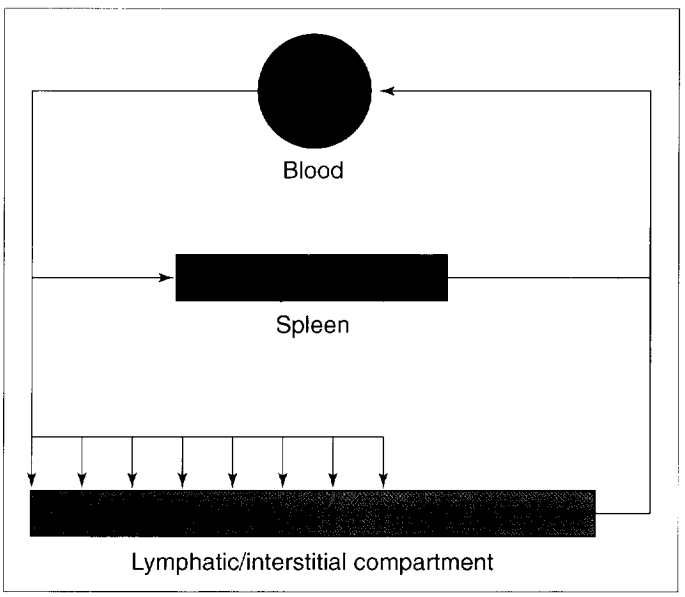

Fig. 1. *Schematic diagram for the transport of lymphocytes between the blood, spleen and lymphatic/interstitial compartments. T cells enter the spleen from the blood at the left-hand edge and traverse in six hours. T cells enter the lymphatic/interstitial compartment from the blood uniformly along the stretch of the compartment shown. Traversal time of the whole compartment is 20 h. The equations for the model are as follows:*

$$\frac{dT(t)}{dt} = -\alpha_s T(t) - \alpha_l T(t) + v_s u_s(1, t) + v_l u_l(1, t) \qquad (1)$$

$$\frac{\partial u_s(x, t)}{\partial t} = -v_s \frac{\partial u_s(x, t)}{\partial x} + D_s \frac{\partial^2 u_s(x, t)}{\partial x^2} + \alpha_s T(t)\delta(x) \qquad (2)$$

$$\frac{\partial u_l(x, t)}{\partial t} = -v_l \frac{\partial u_l(x, t)}{\partial x} + D_l \frac{\partial^2 u_l(x, t)}{\partial x^2} + \alpha_l T(t)\frac{\theta(y - x)}{y} \qquad (3)$$

where T *represents the number of T cells in the blood;* u_s *and* u_l *represent the number of T cells per unit length of spleen or lymphatic system, respectively. T cells enter the spleen at rate* α_s *and travel along the spleen at rate* v_s*. In addition, there is a small diffusion term of strength* D_s*. The length of the spleen is normalized so that it is of length = 1. Cells enter the spleen at the* $x = 0$ *end of the spleen (*δ *is a delta function that controls this entry; for more information on delta functions see Ref. 32), and cells re-enter the circulatory system from the* $x = 1$ *end of the spleen. The equation for the transport of cells through the lymphatic/interstitial compartment is similar to that for the spleen, but has different parameter values,* α_l*,* v_l *and* D_l*. However, cells do not enter the lymphatic/interstitial compartments at the* $x = 0$ *end, as in the spleen, but over the interval* $[0 \leq x \leq y]$ *(*y *can take a value between 0 and 1; in Eqn 3,* θ *is a step function that takes the value 1 for* $x \leq y$ *and 0 for* $x > y$*). The length of the lymphatic/interstitial compartment is also normalized to 1. This is a mathematical trick, and the effective lengths of the two lymphoid compartments are controlled by the parameters* v_s *and* v_l*, which determine the speed at which lymphocytes move through these compartments. The parameter values used throughout the paper, with units in hours, are* $\alpha_s = 1.0$*;* $\alpha_l = 1.5$*;* $v_s = 1/6$*;* $v_l = 1/20$*;* $D_s = 0.005$*;* $D_l = 0.005$*;* $y = 0.65$*.*

PII: S0167 5699(97)01036-0

Box 1. Simulations of classical experiments

The model is simulated for several different classical experiments on lymphocyte localization and recirculation. The same parameter values are used in all the simulations (which have been chosen to fit the results of the experiments on rats[12]), in order to compare the recirculation experiments with the drainage experiments. Very similar parameter values can fit the experiments on mice[11].

Localization experiments

Radiolabelled lymphocytes were injected into animals, who were then slaughtered at different time points[11,26]: rapid mobilization of cells to the spleen (40%) was observed, followed by a decline to steady-state levels; mobilization of cells to lymph nodes was slower. According to the model simulation (Fig. 2a), the percentage of lymphocytes in the spleen reaches a peak of about 40% after 2 h, declining after 4 h to reach a steady state of about 20% by 24 h. The cells in the lymphatic/interstitial compartment accumulate in two phases: there is a rapid mobilization of about 60% of the cells, which occurs in just 2 h, followed by a slower increase over 24 h to a steady state of about 77%. The remaining 3% of the cells at steady state are found in the blood compartment.

Recirculating lymphocyte distribution

Analysis of the distribution of recirculating lymphocytes indicates approximately 20% in the spleen, 3% in the blood, and 60% in lymph nodes and Peyer's patches[27]. These data compare favourably with the steady-state distribution of the model (Fig. 2a). Thus, the simple mechanisms of entry, flow and exit of lymphocytes, along with the parameters chosen to fit the kinetics of the dynamic recirculation and localization experiments, are able to provide correct steady-state distribution of cells in the blood, spleen and lymphatic system. This demonstrates that these processes are sufficient to account for the homeostatic control of the proportions of lymphocytes in these compartments.

Recirculation experiments

Radiolabelled lymphocytes were injected into animals and their appearance in a thoracic duct cannula was measured[11,12,16,26]: peak output occurred in 12–20 h. Figure 2b shows the results of the model simulation, alongside the results of one of these experiments[12]. There is very close agreement between the experimental results and the mathematical model with the parameters as given. A small change to the equations that are used is necessary: the term for the re-entry of lymphocytes into the blood must now have subtracted from it that proportion of lymphocytes that leave the system via the thoracic duct cannula; it is assumed that this is 85% of lymphatic flow.

Drainage experiments

The thoracic duct of animals was cannulated and the numbers of lymphocytes emerging counted[13,26,28,29]: there was a decline in the rate of emergence of lymphocytes, to a species-dependent low steady state. The simulation results are shown in Fig. 2c for a period of four days. There is a decline with an average half-time of 10 h to a zero steady state. According to the highest resolution data[13], the half-time is 16 h; thus, the rate of flow of lymphocytes through the lymphatic system probably slows down in a lymphocyte-depleted animal (see Box 2). In addition, in the laboratory experiments, a non-zero steady state is reached because new lymphocytes are continually being produced in these animals. Terms for the production, proliferation and death of lymphocytes are not included in the model. Furthermore, drainage experiments are also complicated by the possibility that long-term non-recirculating cells are starting to recirculate once the rapidly mobilizable pool of lymphocytes is depleted.

recirculation between blood, spleen and the lymphatic system, setting up a framework within which to answer more complex questions on lymphocyte migration and recirculation, and to relate this work to the problems in HIV dynamics described above. The models in this paper focus only on the recirculation of T cells. Other immune processes, such as the production, activation, proliferation and death of lymphocytes are important, but provide additional layers of complexity that are not necessary for the types of questions that will be asked here. A similar approach has been taken in existing models of lymphocyte recirculation[8,9].

The model

Lymphatic circulation is divided into three compartments: the blood, the spleen (i.e. the white pulp of the spleen), and the lymphatic/interstitial compartment (including the lymph nodes, lymphatic vessels, thoracic duct, Peyer's patches, and also the interstitium, from which some lymphocytes will enter the lymphatic system). The blood is considered here as a homogeneous, mixed fluid with respect to lymphocyte distribution. This is reasonable as the blood circulates approximately once every minute, while it takes six hours for T cells to traverse the spleen, and even longer to traverse the lymphatic system. On the other hand, the spleen and the lymphatic/interstitial compartment are considered as one-dimensional tubes, through which the lymphocytes can travel. Figure 1 demonstrates the flow between and within these compartments. A partial differential equation model has been used to describe this flow, and the equations are given in the legend to Fig. 1.

Cells enter the spleen from the blood at one end of the compartment and traverse through, re-entering the blood from the other end. Cells enter the lymphatic/interstitial compartment uniformly along a length of the compartment (which can be varied in the

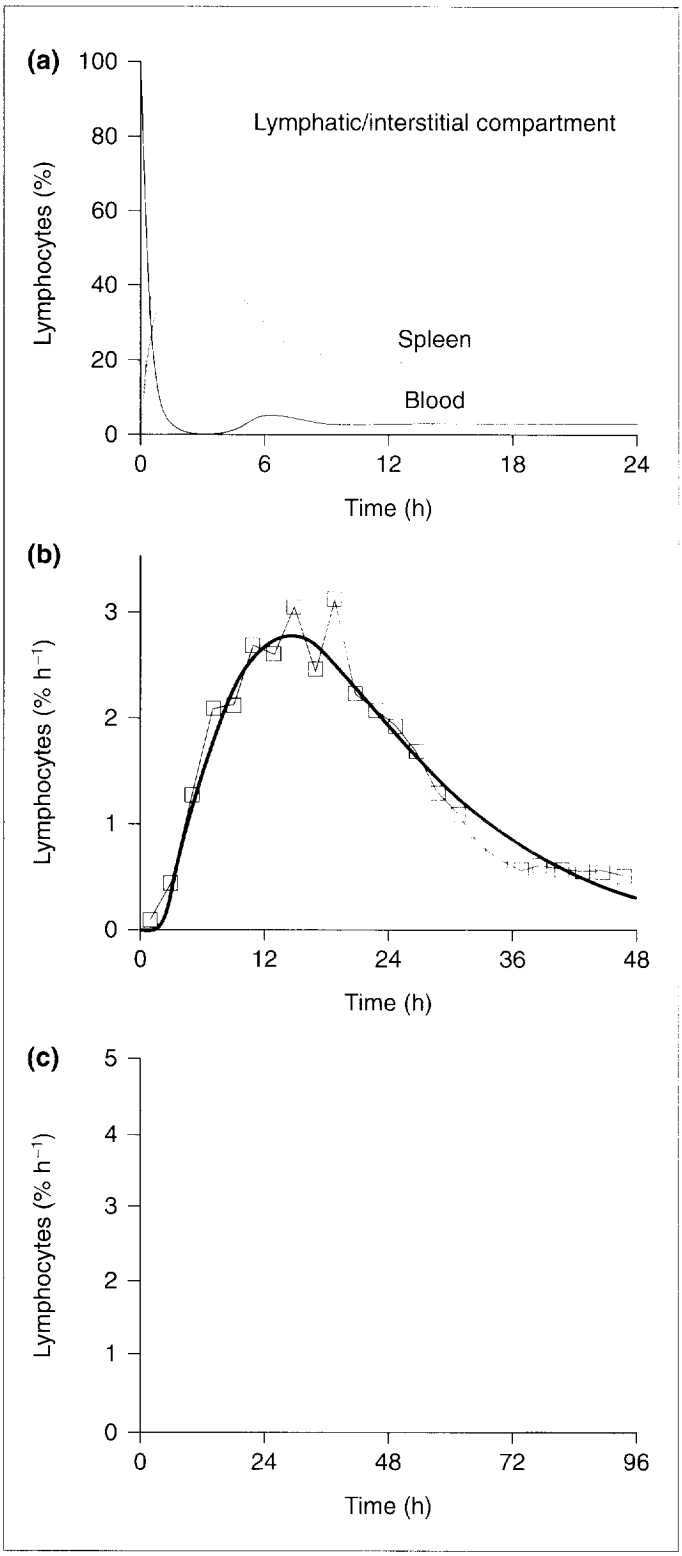

Fig. 2. *(a) Simulation of the percentages of lymphocytes in the blood, spleen and lymphatic/interstitial compartments after an inoculum of cells has been injected into the blood at time* t = 0. *This figure also shows the steady-state levels predicted by the model (77% in the lymphatic/interstitial compartment, 20% in the spleen and 3% in the blood). (b) The rate of appearance in a thoracic duct cannula of radiolabelled lymphocytes injected into the blood at time* t = 0 *for the model (thick line) and experiment*[12] *(squares). It is assumed that only 75% of the injected cells will recirculate, and that 85% of lymphocyte flow occurs through the thoracic duct cannula. (c) Simulation of the drainage of lymphocytes following thoracic duct cannulation. The output goes to zero because there is no term for production of lymphocytes in the model.*

model), as shown in Fig. 1. This mode of entry is intended to mimic two biological phenomena. The first is that cells enter the lymphatic system in two different ways, either from the interstitium via the afferent lymph, or directly from the blood via the high endothelial venules (HEVs). The second is that lymphocytes enter into lymph nodes at different points along the system, so that some cells have less distance to traverse before reaching the thoracic duct than others.

In both the spleen and the lymphatic/interstitial compartment, the model includes a mixing term that is an amalgamation of three different processes: (1) the random motion and mixing of cells, which is true diffusion; (2) the different rates of fluid flow and hence differential advection within the tissue compartments; and (3) the differential adhesion of T cells to other cells of the lymphoid tissue (principally dendritic cells and other antigen-presenting cells), which will result in different cells effectively travelling at different speeds. While a more advanced model may take these factors into account separately, they have been grouped together here in a single term for the sake of simplicity.

Model simulations

The model has been simulated numerically on a computer, using standard routines for solving partial differential equations[10]. The first aim was to establish that the model is capable of simulating the results of the classical experiments on migration and recirculation of lymphocytes (Box 1; Fig. 2a,b), particularly the high-resolution localization and recirculation experiments[11,12]. It was found that the simple mechanisms proposed are sufficient to explain homeostasis of lymphocytes between the lymphoid organs and the blood.

Using the same parameters, simulations of thoracic duct drainage produce a decline of lymphocyte output with a half-time of 10 h (Box 1; Fig. 2c), which is significantly faster than the 16 h half-time observed experimentally[13]. In Box 2, it is postulated that the rate of flow of lymphocytes through lymphoid organs is dependent on the density of lymphocytes in these organs, so that recirculation slows down in lymphocyte-depleted animals.

Next investigated was the effect of short-term changes in the values of the lymphocyte migration parameters of the model (i.e. entry rates into organs and flow rates within these organs) on the steady-state level of lymphocytes in the blood. Figure 3 demonstrates the exquisite sensitivity of T-cell counts in the blood to such changes, this being a consequence of the small proportion of lymphocytes resident in the blood. The proportions of lymphocytes in the spleen and lymphatic system do not change greatly during these simulations.

Finally, the principles of the model were applied to the analysis of T-cell subsets. The initial model focuses only on total T cells

(B cells have very different recirculation kinetics[12]), and ignores distinctions between $CD4^+/CD8^+$ or $CD45R0^+/RA^+$ T-cell subsets. $CD4^-/CD8^+$ and $CD45R0^+/RA^+$ T cells have identical recirculation kinetics[12,14], and yet it has been suggested that $CD45R0^+/RA^+$ may migrate via different routes[15]. Box 3 incorporates the distinction between $CD45R0^+$ and RA^+ cells, and establishes that these experimental results are not contradictory[14], but give important information about the relative efficiencies in the uptake of naive and memory T cells.

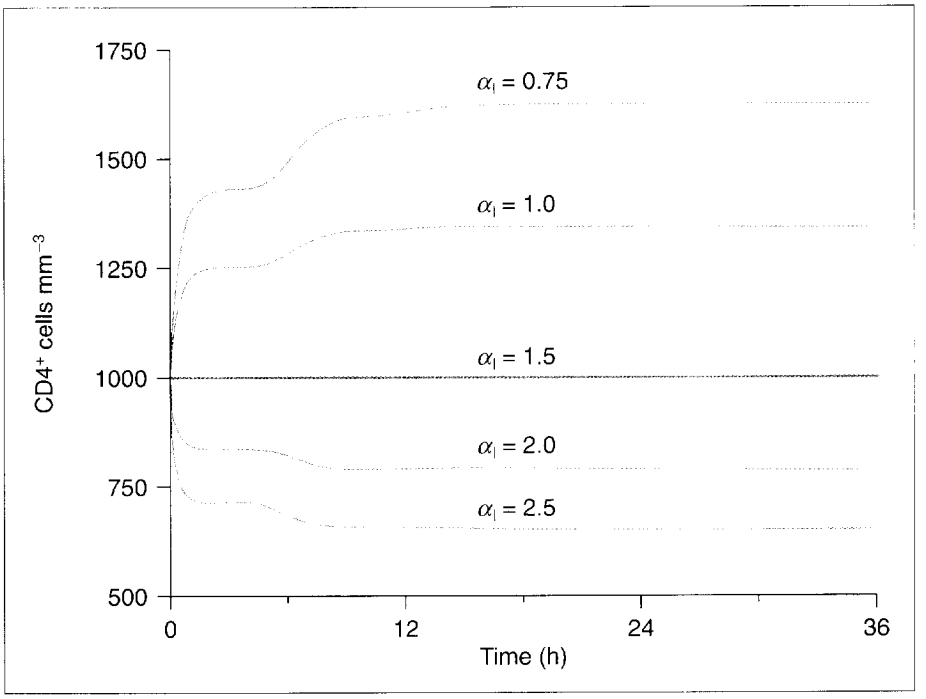

Fig. 3. *The numbers of $CD4^+$ T cells mm^{-3} of blood plotted against time when the rate of entry of lymphocytes from the blood into the lymphatic/interstitial compartment (α_l) is changed at time t = 0 from its default value of 1.5 h^{-1}. All other parameters are held constant. The steady state of the system is normalized so that this count is 1000 cells mm^{-3}. For all alterations in α_l, there is a rapid change in the T-cell count over 2 h, followed by a slower settling to the new steady state over 24 h. Similar results are observed when changes are made to the other entry and flow rate parameters in the model. The proportions of lymphocytes in the spleen and lymphatic system do not change greatly during these simulations. It is clear that simultaneous changes to multiple parameters will result in even greater variability of the T-cell counts in blood.*

Concluding remarks

This article has set out to develop a mathematical model of lymphocyte recirculation, based on the simple processes of lymphocyte entry, transport, mixing and exit. In doing so, the model successfully matches the results of several classical experiments. The parameter values that fit the high-resolution recirculation and localization experiments predict correct proportions of lymphocytes in the different compartments, and indicate that these simple processes are sufficient to explain this homeostasis.

The inability of the simplest model to reproduce the different time-scales of the recirculation and drainage experiments is a clear example of the value of simple mathematical models: it demonstrates that there must be a process other than entry, flow and exit at work in explaining even the simplest-seeming experiments on lymphocyte recirculation. The hypothesis of density-dependent transport implicates cell–cell adhesion as playing an important role in lymphocyte transport. The slower recirculation times observed in nude rats compared with euthymic rats[16], together with the slower recirculation times in irradiated recipients[17,18], provide strong evidence in favour of this hypothesis. There is, however, experimental evidence that suggests the contrary. First, the recirculation kinetics are the same in lambs thymectomized immediately before birth as in normal lambs[19]. Second, the isolated perfused spleens from normal and irradiated rats show similar recirculation kinetics[20]. In the first case, the ovine thymus produces large numbers of T cells before birth[21], and so these animals will not be lymphocyte depleted to the same extent as the nude and irradiated rats. In the second case, the extent to which the isolated perfused spleen reflects the *in vivo* situation of a whole rat is unclear (discussed in Ref. 17). Clearly, this is an area for further experimental testing.

Density-dependent transport may have implications during the asymptomatic phase of HIV infection: if, in a lymphocyte-depleted individual, the rate of traffic of lymphocytes through lymphoid tissue is slower, then the relative proportion of lymphocytes in the lymphoid tissue as compared with the blood is increased. Thus, T-cell counts in the blood may give a disproportionately low estimate of the true numbers of T cells in the patient. Conversely, if as a result of drug treatment there is an increase in the number of T cells, then the rate of traffic will increase, and so there may be a redistribution of lymphocytes back to the blood. Thus, the increases in T-cell counts observed in the blood may give a disproportionately high estimate of the rate of replication of T cells.

The potentially important role of cell–cell adhesion is further exemplified in the cell-mixing term that is included in the model. The value of the mixing parameter that allowed successful simulation of the precise distribution of transit times seen in the recirculation experiments was found to be very much higher than the true rate of diffusion for cells[22]. This implicates adhesion processes as being a major contributor to the mixing of lymphocytes in the lymphoid tissues. Recent work addressing adhesive interactions between T cells and dendritic cells can generate the distribution of transit times seen in the recirculation experiments[23], and can also simulate both the results of the drainage experiments and the slower recirculation times observed in nude and irradiated rats (D.J. Stekel, unpublished).

Box 2. Density-dependent traffic of lymphocytes through lymphoid organs

- In drainage experiments, the number of lymphocytes emerging declines with a half-time of 16 h; in our simulations, this half-time is only 10 h. These different time-scales are irreconcilable if one considers only entry, flow and exit of lymphocytes.

- We postulate that, in a lymphocyte-depleted animal, the flow of lymphocytes through lymphoid organs slows down. While traversing through the lymphoid organs, T cells are coming into contact with other cell types (e.g. dendritic cells) to which they bind; once bound, their motion temporarily stops. If an animal is lymphocyte depleted, fewer T cells are competing for the same number of binding sites, and so the proportion of time each lymphocyte spends bound to other cell types is increased, thereby decreasing their rate of traversal.

Experimental evidence in favour of this hypothesis

- When radiolabelled lymphocytes from euthymic donor rats were injected into nude (congenitally athymic) and euthymic (normal) recipients, the rate of recirculation was considerably slower in the nude rats[16]. When nude rats were reconstituted with thymus grafts, their recirculation pattern was the same as euthymic rats. Nude rats have a near-complete absence of lymphocytes in the T-cell regions of their lymphoid tissue, but large numbers of interdigitating dendritic cells are present[30].

- When radiolabelled lymphocytes were injected into irradiated recipients, they recirculated more slowly than in non-irradiated recipients[17,18], with peak emergence in 20–30 h (Ref. 18).

Box 3. CD45R0$^+$ and RA$^+$ T cells: incorporation into the model

For any T-cell subset at steady state, the rate of migration of cells from the blood into a lymphoid organ must be equal to the rate of return of these cells to the blood. Thus, we can write:

$$\alpha_+ T_+ = v_+ u_+ \tag{4}$$

$$\alpha_- T_- = v_- u_- \tag{5}$$

where the + subscripts are for R0$^+$ cells and the − subscripts for RA$^+$ cells, T denotes the cell concentrations in the blood, α the rate of entry of these cells into the lymphatic system, v the average rate of traffic through the lymphatic system and u the concentration of these cells in efferent (thoracic duct) lymph. We know that $v_+ = v_-$ (Ref. 14), and so dividing Eqn 4 by Eqn 5 gives:

$$\frac{\alpha_+}{\alpha_-} = \frac{u_+}{u_-} \div \frac{T_+}{T_-} \tag{6}$$

or, the ratio of the rate of uptake from blood of R0$^+$ cells to RA$^+$ cells is equal to the ratio of R0$^+$ to RA$^+$ cells in efferent lymph divided by the ratio of R0$^+$ to RA$^+$ cells in blood. Most T cells in efferent lymph display a naive phenotype, while there is a higher proportion of memory cells in the blood[15]. This indicates a greater rate of uptake from the blood of naive cells compared with memory cells. It has been suggested that memory cells migrate to lymph nodes preferentially via afferent lymph, and naive cells via high endothelial venules (HEVs)[15]. In that case, one might conclude from this result that the uptake of lymphocytes from the blood via HEVs is more efficient than uptake via afferent lymph. Alternatively, naive cells have also been found to migrate via afferent lymph[31], and so this result may reflect the possibility that naive cells have available to them more routes of entry from blood into lymph nodes than do memory cells.

The model contained only three compartments: blood, spleen and a lymphatic/interstitial compartment. A potential refinement for this type of model would be to contain many lymphatic compartments[9]. Such a refinement would enable the analysis of the different recirculatory behaviours of gut-homing and lymph-node-homing lymphocytes[19,24].

The steady-state levels of T cells in the blood are highly sensitive to changes in the model parameters over short time-scales. The effects of a large variety of factors on the composition of lymphocyte subsets in the blood, including everyday activities, drugs, hormones, medical procedures and diseases, are well documented[25]. The variations seen in the model probably account for some of these phenomena. In addition, the potential for large variation in the levels of T cells in the blood as a result of changes in the migrational properties of these cells provides support for the idea that redistribution of T cells from blood to lymph may, to some extent, underlie the decline in CD4$^+$ T cells seen in HIV and SIV infection[1,2]. This variation also calls into question the significance of T-cell data in HIV-infected patients, particularly those whose levels of CD4$^+$ cells are only being measured once every few years. Clearly, this is an important area for further work, as it is essential to understand the precise implications of measurements made in the blood on the overall state of the immune system.

Thanks to E. Bell, M. Binks, S. Frost, S. Hunt, D. Katz, A. Lloyd, A. McLean, G. McPherson and R. Payne for helpful discussion, and the referees for their constructive comments. D.J.S., C.E.P. and M.A.N. are all funded by the Wellcome Trust (grant numbers 040947, 038318 and 035528 respectively).

Dov Stekel *(dov.stekel@zoo.ox.ac.uk),* ***Claire Parker*** *and* ***Martin Nowak*** *are at the Dept of Zoology, University of Oxford, South Parks Road, Oxford, UK OX1 3PS.*

References

1 Rosenberg, Y.J., Zack, P.M., White, B.D. *et al.* (1993) *AIDS Res. Hum. Retroviruses* 9, 639–646
2 Røsok, B.I., Bostad, L., Voltersvik, P. *et al.* (1996) *AIDS* 10, F35–F38
3 Wei, X., Ghosh, S.K., Taylor, M.E. *et al.* (1995) *Nature* 373, 117–122
4 Ho, D.D., Neumann, A.U., Perelson, A.S. *et al.* (1995) *Nature* 373, 123–126
5 Mosier, D.E. (1995) *Nature* 375, 193–194
6 Sprent, J. and Tough, D. (1995) *Nature* 375, 194
7 Dimitrov, D.S and Martin, M.A. (1995) *Nature* 375, 194–195
8 Ottoway, C.A. and Parrott, D.M.V. (1981) *Immunol. Lett.* 2, 283–290
9 Farooqi, Z.H. and Mohler, R.R. (1989) *IEEE Trans. Biomed. Eng.* 36, 355–362
10 Press, W.H., Flannery, B.P., Teukolsky, S.A. and Vetterling, W.T. (1990) *Numerical Recipes in C: The Art of Scientific Computing*, pp. 636–656, Cambridge University Press
11 Smith, M.E. and Ford, W.L. (1983) *Immunology* 49, 83–94
12 Westermann, J., Puskas, Z. and Pabst, R. (1988) *Scand. J. Immunol.* 28, 203–210
13 Westermann, J., Matyas, J., Persin, S. *et al.* (1994) *Scand. J. Immunol.* 39, 395–402
14 Westermann, J., Persin, S., Matyas, J. *et al.* (1994) *J. Immunol.* 152, 1744–1750
15 Mackay, C.R., Marston, W.L. and Dudler, L. (1990) *J. Exp. Med.* 171, 801–817
16 Fossum, S., Smith, M.E. and Ford, W.L. (1983) *Scand. J. Immunol.* 17, 551–557
17 Bell, E.B. and Shand, F.L. (1975) *Eur. J. Immunol.* 5, 1–7
18 Dorsch, S. and Roser, B. (1975) *Nature* 258, 233–235
19 Cahill, R.N.P., Kimpton, W.G., Washington, E.A. *et al.* (1996) *Immunology* 88, 130–133
20 Ford, W.L. (1969) *Cell Tissue Kinet.* 2, 171–191
21 Sprent, J. (1993) in *Fundamental Immunology* (3rd edn) (Paul, W.E., ed.), pp. 74–109, Raven Press
22 Sackmann, E. (1983) in *Biophysics* (Hoppe, W., Lohmann, W., Markl, H. and Ziegler, H., eds), pp. 425–457, Springer-Verlag
23 Stekel, D.J. *J. Theor. Biol.* (in press)
24 Cahill, R.N.P., Poskitt, D.C., Frost, H. and Trnka, Z. (1977) *J. Exp. Med.* 145, 420–428
25 Westermann, J. and Pabst, R. (1990) *Immunol. Today* 11, 406–410
26 Sprent, J. (1973) *Cell. Immunol.* 7, 10–39
27 Trepel, F. (1974) *Klin. Wschr.* 52, 511–515
28 Mann, J.D. and Higgins, G.M. (1950) *Blood* 5, 177–190
29 Gowans, J.L. (1957) *Br. J. Exp. Pathol.* 38, 67–78
30 Fossum, S., Smith, M.E., Bell, E. and Ford, W.L. (1980) *Scand. J. Immunol.* 12, 421–432
31 Kimpton, W.G., Washington, E.A. and Cahill, R.N.P. (1995) *Int. Immunol.* 7, 1567–1577
32 Jordan, D.W. and Smith, P. (1994) *Mathematical Techniques*, p. 392, Oxford University Press

Views on Vav

Tassie L. Collins, Marcel Deckert and Amnon Altman

Vav is a 95 kDa protooncogene product whose expression is largely limited to cells of hematopoietic origin. Although reports on the role of Vav in Ras activation have generated conflicting results, it has become evident that Vav plays a crucial positive role in lymphocyte development and stimulation. This article summarizes current knowledge on Vav and speculates on its role in hematopoietic cell signaling.

Vav, named after the sixth letter in the Hebrew alphabet, was originally identified as a transforming gene product in fibroblasts that results from a deletion of the N-terminal 67 residues from the proto-oncogene product[1]. Since its identification, Vav has been the subject of intense scrutiny owing to its structural similarity to several families of signaling molecules. Vav displays several distinct domains (Fig. 1)[2], including: a cysteine-rich zinc-binding domain similar to that found in members of the protein kinase C family; and an arrangement of signaling motifs that are highly reminiscent of 'adaptor' molecules, particularly the ubiquitously expressed growth factor receptor-bound protein 2 (Grb2) [i.e. a phosphotyrosine-binding *src*-homology 2 (SH2) domain flanked by two proline-binding SH3 domains]. Vav also appears to be involved in the activation of members of the Ras superfamily of small G proteins, since it contains a region that is highly homologous to the guanine nucleotide exchange factor (GEF) domain of the Rho exchange protein Dbl (this has been termed the Dbl-homology, or DH, domain)[3]. In addition, Vav has a pleckstrin-homology (PH) domain. Although initial reports described the presence of domains characteristic of transcription factors, such as nuclear localization signals, an amphipathic helix–loop–helix (HLH), and a leucine zipper (LZ), clear evidence for these has failed to materialize.

Several key findings have demonstrated the importance of Vav to hematopoietic cell function. First, Vav is phosphorylated on

PII: S0 167-5699(97)01037-7
 0167-5699/97/$17.00