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MODELING IMMUNOTHERAPY FOR ALLERGY

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Type I hypersensitivity, which functions to protect the organism from parasites, is caused by binding of antigen to IgE antibodies pre-attached to the cell surface of tissue mast cells and their circulating counterparts, the basophils. In “allergy,” type I hypersensitivity is inappropriately induced by protein-based foreign substances (such as pollen) or protein components of insect stings, which in the normal course of events would be cleared from the organism without causing any damage. Paradoxically, a successful clinical treatment of allergy involves repeated immunization of allergic persons with low doses of the allergen—*immunotherapy*. Investigation of the available experimental evidence leads to the conclusion that the phenomena of immunotherapy are best addressed in terms of the interplay among the mechanism(s) of immune memory—Th1/Th2 cross-regulation—and the physical compartmentalization of the immune system. These conclusions are illustrated with a numerical simulation. © 1996 Society for Mathematical Biology

1. Introduction. Type I hypersensitivity is caused by binding of antigen to IgE antibodies pre-attached to the cell surface of tissue mast cells and their circulating counterparts, the basophils. Multivalent antigen binding to the pre-attached IgE promotes their cross-linking. Extensive cross-linking (Dembo *et al.*, 1978) results in a release of a variety of mediators that collectively cause increased vascular permeability, vasodilation, smooth muscle contraction and local inflammation. Because these reactions occur rapidly, within minutes of repeat exposure to antigen, type I hypersensitivity is also called *immediate* hypersensitivity. In their benign form these reactions are thought to protect the organism from parasites (Abbas *et al.*, 1991).

Type I hypersensitivity is inappropriately induced by protein-based foreign substances (such as pollen) or protein components of insect stings that in the normal course of events would be cleared from the organism without causing any damage. In its most extreme systemic form, *anaphylaxis*, mast cell/basophil-derived mediators can restrict airways to the point of asphyxiation and produce lethal cardiovascular collapse (Abbas *et al.*, 1991). Such outcomes are fortunately rare. However, more moderate allergic responses

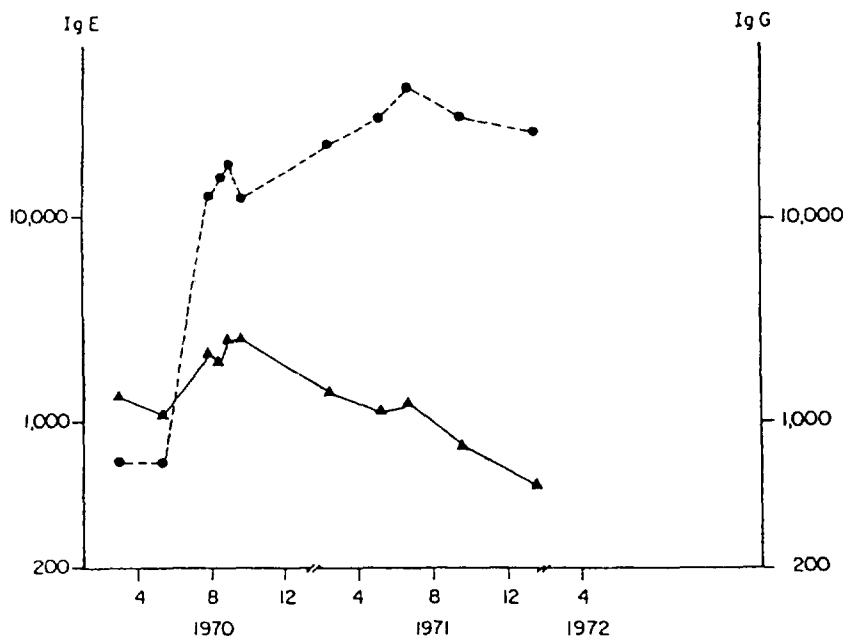


Figure 1. IgE (triangles) and IgG (circles) measurements in sera following immunotherapy from patients sensitive to ragweed. Reproduced with permission from Ishizaka (1976).

are widespread, e.g. an estimated 20% of the U.S. population suffers from an allergy to some common substance (Lichtenstein, 1993). The list of allergy-inducing substances includes foods, plant pollen (hay fever), antibiotics and protein components of insect bites.

An often successful clinical treatment of allergy involves repeated immunization of allergic persons with low doses of allergen—*immunotherapy* (Ishizaka, 1976). The treatment, if successful, continues for years. Observable outcomes of this treatment are (a) an increase in the concentration of circulating IgG antibodies, (b) slow (half-life = years) decrease in the concentration of circulating IgE antibodies and (c) suppression of the secondary IgE responses after long-term treatment (Ishizaka, 1976; Graft *et al.*, 1987) (Fig. 1). The primary purpose of this paper is to formulate a hypothesis for understanding immunotherapy for pollen antigens and to explore some of its aspects via mathematical modeling.

2. Formulation of the Hypothesis. Continuous production of allergen-specific antibody in allergic individuals (Ishizaka, 1976; Graft *et al.*, 1987) indicates that type I hypersensitivity is an immune memory phenomenon. This fact plays an important role in our model.

It is known (Coffman and Carty, 1986) that Th2 cells promote IgE production, whereas Th1 cells promote IgG production. Thus the observed outcome of immunotherapy may represent a switch from a Th2 dominated memory state to a Th1 dominated one. This hypothesis is substantiated by the results of Secrist *et al.* (1993), who have observed that during immunotherapy the relative fraction of (allergen-specific) Th1 cells increases. In this paper we address immunotherapeutic phenomena in terms of the underlying causes of Th1/Th2 cross-regulation.

Activated Th1 and Th2 cells are cross-suppressive (Fernandez-Botran *et al.*, 1988; Fiorentino *et al.*, 1989, 1991). Both empirical observation (Mosmann and Moore, 1991) and analytical extrapolation of the cross-suppression dynamics (Fishman and Perelson, 1994) indicate that an antigenic challenge results in either Th1 domination or Th2 domination. The results of Fishman and Perelson (1994) indicate that once Th2 dominance is established, its stability is increased by the addition of antigen. Hence, an established Th2 dominance can not be broken by adding antigen. Therefore, the first question about immunotherapy that we ask is the following:

- Why does “natural” exposure to the allergen induces Th2 dominance (and, consequently, IgE production), while subsequent immunization with the allergen (immunotherapy) induces Th1 and, hence, IgG production?

For pollen antigen a possible answer to this question was suggested by recent findings on *compartmentalization* of the immune system. Kroemer *et al.* (1993) reviewed evidence indicating that, for certain antigens and for experimental subjects bearing susceptible human leukocyte antigens (HLA) haplotypes (Marsh *et al.*, 1982), an antigen introduced to lymph nodes that drain mucosal surfaces—sites of the “natural” introduction of pollen—induces response of predominantly Th2 type. When the same antigen is introduced into non-mucosal lymph nodes or into the spleen, the response is dominated by Th1 cells. Similarly, Nabors *et al.* (1996) have shown that cross offspring of *Leishmania* susceptible and *Leishmania* resistant mouse strains develop a Th1 response if inoculated with *Leishmania* in the footpad, but develop a Th2 response if inoculated with *Leishmania* in the dorsal skin.

Thus, unlike the earlier studies of Fishman and Perelson (1993, 1994, 1995), in immunotherapy we cannot view the immune system as a single compartment, but rather we must consider, at minimum, two compartments: One is centered on the mucosal lymph nodes—compartment μ —and one is centered on non-mucosal lymphatic tissue(s) that drain the sites of immunotherapeutic injections—compartment η .

In these terms the difference in immunoglobulin isotypes observed between the "natural" exposure to an allergen and immunotherapy is interpreted as follows. Pollen enters the patient through mucosal surfaces, which initiates an immune response in lymph nodes draining these surfaces. In susceptible persons, i.e. persons that possess certain HLA haplotypes, this response becomes dominated by a Th2 subset. Th2 dominance leads to the establishment of IgE production. Immunotherapy presumably introduces the allergen into non-mucosal lymph nodes and (see above) may result in a Th1 dominated response leading to IgG production.

This, however, raises an additional problem.

- Even though immunotherapy expands the allergen-specific Th1 subset in compartment η , an explanation needs to be given for the fact that the observed (Secrist *et al.*, 1993) replacement of the allergen-specific Th2 cells is systemic.

The solution to this problem is suggested by the fact that recirculating lymphocytes traffic continuously between blood and lymph. Their route takes them through various lymphoid organs in the body (reviewed by Woodroof *et al.*, 1987). In particular, T cells, initially recruited into non-mucosal lymphatic tissue (compartment η) will pass through mucosal lymphatic tissues and vice versa. Thus, in principle, migration of allergen-specific Th1 cells, induced by immunotherapy, into mucosal compartment can overwhelm the local Th2 response, which yields systemic Th1 dominance. Examination of this proposition constitutes the principal aim of our paper.

3. Construction of the Model. It is known that both Th cell maturation (reviewed by Seder and Paul, 1994) and the eventual outcome of the Th1/Th2 cross-regulation (Fishman and Perelson, 1994) are influenced by the *microenvironment*. However, it appears that the proximate reason for the difference, with respect to Th1/Th2 responses, between mucosal and non-mucosal lymph nodes, has not been documented at present.

In constructing our model we reasoned as follows. While it is known that both maturation of Th1/Th2 cells and the cross-regulation between them are influenced by the microenvironment, available evidence (Seder and Paul, 1994) indicates that the microenvironment plays a crucial role in recruitment (maturation) of Th cells into Th1 and Th2 subsets. Thus, we assume that the difference between mucosal and non-mucosal lymphatic tissue can be accounted for by the differences in recruitment. In the current model we represent these differences by assuming that the recruit-

ment rates for Th1 versus Th2 cells in the two compartments are different (Fig. 2a).

Our mathematical description of intracompartamental memory maintenance and Th1/Th2 cross-regulation is based on Fishman and Perelson (1995) and (1994), respectively. We summarize the salient features in Fig. 2b. We note that for reasons of mathematical simplicity and in the absence of evidence to the contrary, migration of Th cells between the two compartments is assumed to be symmetric. Supplementary modeling, not shown, indicates that while a small degree of asymmetry affects the numerical results, it will not change the basic properties of the model.

In terms of ordinary differential equations, our model for the mucosal compartment, indexed with subscript μ , has the following form:

$$\begin{aligned} \frac{dR_{\mu 0}}{dt} = & G_{\mu}(t) - k_{b_1} R_{\mu 0} T_{\mu 1} + (k_a + k_{d_1}) R_{\mu 1} - k_{b_2} R_{\mu 0} T_{\mu 2} \\ & + (k_a + k_{d_2}) R_{\mu 2} - d_R R_{\mu 0}, \end{aligned} \quad (1a)$$

$$\frac{dR_{\mu 1}}{dt} = k_{b_1} R_{\mu 0} T_{\mu 1} - (k_a + k_{d_1}) R_{\mu 1} - d_R R_{\mu 1}, \quad (1b)$$

$$\frac{dR_{\mu 2}}{dt} = k_{b_2} R_{\mu 0} T_{\mu 2} - (k_a + k_{d_2}) R_{\mu 2} - d_R R_{\mu 2}, \quad (1c)$$

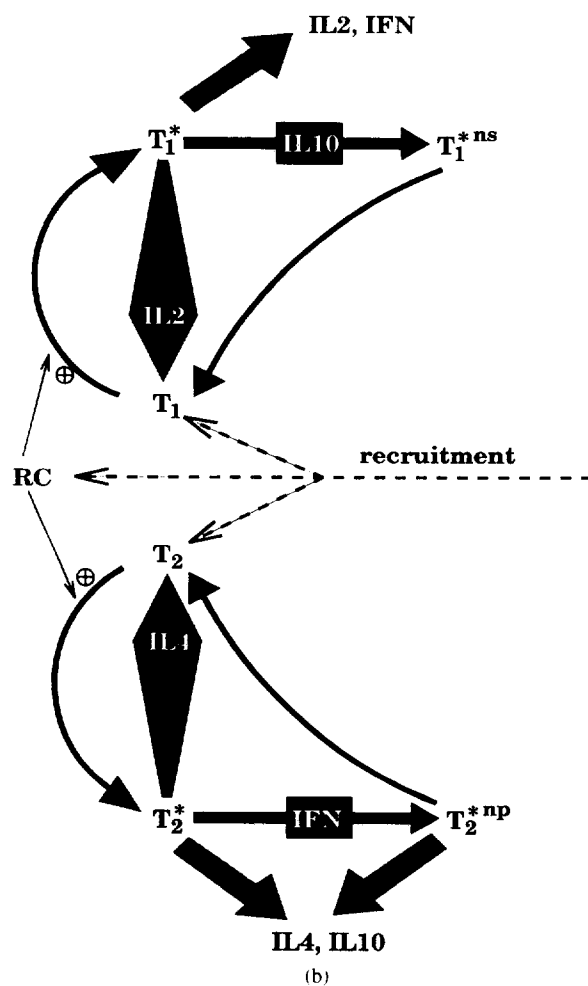
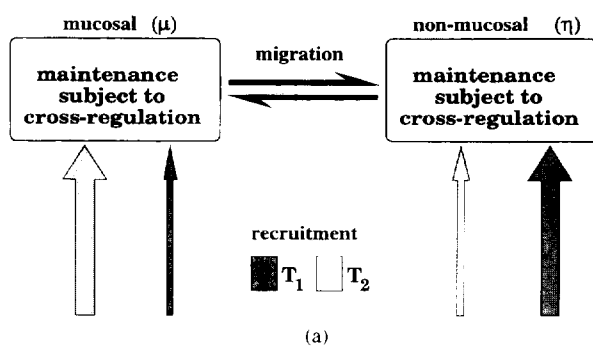
$$\begin{aligned} \frac{dT_{\mu 1}}{dt} = & H_{\mu 1}(t) + k_m(T_{\eta 1} - T_{\mu 1}) - d_T T_{\mu 1} - k_{b_1} R_{\mu 0} T_{\mu 1} \\ & + k_d R_{\mu 1} + (2k_p + k_r) T_{\mu 1}^* + k_r T_{\mu 1}^{*ns}, \end{aligned} \quad (1d)$$

$$\begin{aligned} \frac{dT_{\mu 2}}{dt} = & H_{\mu 2}(t) + k_m(T_{\eta 2} - T_{\mu 2}) - d_T T_{\mu 2} - k_{b_2} R_{\mu 0} T_{\mu 2} \\ & + k_d R_{\mu 2} + (2k_p + k_r) T_{\mu 2}^* + k_r T_{\mu 2}^{*np}, \end{aligned} \quad (1e)$$

$$\frac{dT_{\mu 1}^*}{dt} = k_a R_{\mu 1} - \left(k_p + k_r + k_{ns} \frac{\text{IL-10}_{\mu}}{K_{\text{IL-10}} + \text{IL-10}_{\mu}} \right) T_{\mu 1}^*, \quad (1f)$$

$$\frac{dT_{\mu 2}^*}{dt} = k_a R_{\mu 2} - \left(k_p + k_r + k_{np} \frac{\text{IFN}_{\mu}}{K_{\text{IFN}} + \text{IFN}_{\mu}} \right) T_{\mu 2}^*, \quad (1g)$$

$$\frac{dT_{\mu 1}^{*ns}}{dt} = k_{ns} \frac{\text{IL-10}_{\mu}}{K_{\text{IL-10}} + \text{IL-10}_{\mu}} T_{\mu 1}^* - k_r T_{\mu 1}^{*ns}, \quad (1h)$$



$$\frac{dT_{\mu 2}^{*np}}{dt} = k_{np} \frac{\text{IFN}_{\mu}}{K_{\text{IFN}} + \text{IFN}_{\mu}} T_{\mu 2}^{*} - k_r T_{\mu 2}^{*np}, \quad (1i)$$

$$\frac{d\text{IFN}_{\mu}}{dt} = p_{\text{IFN}} T_{\mu 1}^{*} - d_{\text{IFN}} \text{IFN}_{\mu}, \quad (1j)$$

$$\frac{d\text{IL-10}_{\mu}}{dt} = p_{\text{IL-10}} (T_{\mu 2}^{*} + T_{\mu 2}^{*np}) - d_{\text{IL-10}} \text{IL-10}_{\mu}. \quad (1k)$$

Here R_{μ} represents the population of restimulator sites (on restimulator cells) situated in the mucosal compartment. Restimulator cells are long-lived antigen-retaining cells; see Fishman and Perelson (1995) for details. We take their lifetime to be 100 times that of the T cells ($\rho = 0.01$; see below), but any large multiple will do. $G_{\mu}(t)$ represents recruitment of restimulator cells/sites (due to repeated exposure to allergen), while their decay is linear with coefficient d_R . $R_{\mu 0}$, $R_{\mu 1}$ and $R_{\mu 2}$ represent the subpopulation of sites that are, respectively, unoccupied, occupied by Th1 cells (T_1) or occupied by Th2 cells (T_2). Recruitment rates for these, *resting*, Th1 and Th2 cell populations are given by $H_{\mu 1}(t)$ and $H_{\mu 2}(t)$, respectively. A T_i cell binds to a restimulator site with a coefficient k_{b_i} , as discussed by Fishman and Perelson (1994); Th1 and Th2 cells responding to the same antigen are invariably directed to different determinants and, thus, have different

Figure 2. Schematic description of the model. (a) As discussed, there are two compartments, one centered on the mucosal lymph nodes (compartment μ) and one centered on non-mucosal lymphatic tissue(s) that are targeted by immunotherapeutic injections (compartment η). Antigenic challenges directed to compartment μ preferentially yield Th2 cell recruitment. The situation is reversed for compartment η . (b) Single compartment maintenance of (T-cell) memory, subject to Th1/Th2 cross-regulation. Introduction of antigen leads to the recruitment of antigen-specific Th1 and Th2 cells (T_1 and T_2). Exposure to antigen also promotes recruitment of *restimulator cells* (RC). Restimulator cells are long-lived antigen-retaining cells that maintain immune memory by presenting sequestered antigen to memory lymphocytes (reviewed in Fishman and Perelson, 1995). Interaction between an RC and a T cell induces T-cell activation (sometimes called sensitization). Activated Th1 cells (T_1^*) secrete IL-2 and IFN- γ . Activated Th2 cells (T_2^*) secrete IL-4 and IL-10. IL-10 inhibits production of cytokines by activated Th1 cells. The resulting, inhibited, non-secreting cells (T_1^{*ns}) cannot produce cytokines and thus cannot proliferate in the absence of externally added growth factors. The inhibition may not be permanent and the cells are assumed to return their uninhibited resting state at some rate. IFN- γ inhibits cytokine utilization by T_2^* cells, inducing cells that produce cytokines but cannot utilize them. Thus, these cells are non-proliferative (T_2^{*np}) and ultimately relax to the resting state without proliferation. Activated cells, T_1^* and T_2^* , proliferate, symbolized by broadening arrows, by utilizing endogenously produced growth factors (IL-2 or IL-4—not explicitly considered in model) and ultimately relax to the resting state.

interaction coefficients. A bound T_i cell can disassociate non-productively, with a coefficient k_{d_i} , or it can become activated, yielding T_i^* , with coefficient k_a .

T_1^* cells secrete interleukin-2 (IL-2) and interferon- γ (IFN). T_2^* cells secrete interleukin-4 (IL-4) and interleukin-10 (IL-10). T_1^* (T_2^*) utilizes IL-2 (IL-4) for proliferation in autocrine fashion. Fishman and Perelson (1993) showed that this autocrine utilization allows a particularly simple formulation of proliferation dynamics. Because the cells that utilize IL-2 (IL-4) growth factors also produce them, the concentration of the growth factor is highest in the immediate neighborhood of the secreting cell. Thus, it turns out that for an individual cell the probability of proliferation is only weakly dependent on the bulk growth factor concentration. Hence, the per capita proliferation rate k_p can be taken to be constant and IL-2 and IL-4 need not be explicitly considered in our model. Interferon- γ affects T_2^* cells, yielding inhibited T_2^{*np} cells that function normally, but cannot utilize IL-4 and thus do not proliferate. IL-10 acts on T_1^* , yielding inhibited T_1^{*ns} cells that do not secrete lymphokines and hence cannot proliferate. Th1/Th2 cross-regulation processes are very complex and give rise to many special cases. Here we utilize the results of Fishman and Perelson (1994), in which an extensive—though by no means exhaustive—analysis of Th1/Th2 cross-regulation was performed, in order to present a (relatively) simple formulation of the Th1/Th2 cross-regulation dynamics. Finally, resting cells migrate from compartment μ to compartment η and vice versa with coefficient k_m .

The equations for Th1 and Th2 dynamics in compartment η are essentially the same as those for compartment μ , with obvious notational changes. For example the counterpart of Equation (1a) is

$$\begin{aligned} \frac{dR_{\eta 0}}{dt} = & G_{\eta}(t) - k_{b_1} R_{\eta 0} T_{\eta 1} + (k_a + k_{d_1}) R_{\eta 1} - k_{b_2} R_{\eta 0} T_{\eta 2} \\ & + (k_a + k_{d_2}) R_{\eta 2} - d_R R_{\eta 0}. \end{aligned}$$

As discussed by Fishman and Perelson (1994), Th1/Th2 cross-regulation involves two time scales. There is a *fast* (several hours to one day) time scale of restimulator cell-T-cell interactions and activated T-cell relaxation to a resting state and a *slow* (several weeks) time scale prescribed by the T-cell's life-span. Thus, analysis of the governing equations can be facilitated by non-dimensionalization followed by separation of time scales. We select the scales for non-dimensionalization of Equations (1) as described

by Fishman and Perelson (1994, 1995):

$$\tau = d_T t, \quad K_1 = \frac{k_{b_1}}{k_a + k_{d_1}}, \quad K_2 = \frac{k_{b_2}}{k_a + k_{d_2}}, \quad (2a)$$

$$\begin{aligned} & (r_{\mu 0}, r_{\mu x}, r_{\mu y}, r_{\eta 0}, r_{\eta x}, r_{\eta y}) \\ &= \frac{k_a k_p K_1}{d_T (k_p + k_r)} (R_{\mu 0}, R_{\mu x}, R_{\mu y}, R_{\eta 0}, R_{\eta x}, R_{\eta y}), \end{aligned} \quad (2b)$$

$$(x_\mu, x_\eta) = K_1 (T_{\mu x}, T_{\eta x}), \quad (y_\mu, y_\eta) = K_2 (T_{\mu y}, T_{\eta y}), \quad (2c)$$

$$\begin{aligned} & (x_\mu^*, x_\eta^*, x_\mu^{*ns}, x_\eta^{*ns}, l_{\mu x}, l_{\eta x}) \\ &= \frac{k_p K_1}{d_T} (T_{\mu x}^*, T_{\eta x}^*, T_{\mu x}^{*ns}, T_{\eta x}^{*ns}, \frac{d_{\text{IFN}}}{p_{\text{IFN}}} \text{IFN}_\mu, \frac{d_{\text{IFN}}}{p_{\text{IFN}}} \text{IFN}_\eta), \end{aligned} \quad (2d)$$

$$\begin{aligned} & (y_\mu^*, y_\eta^*, y_\mu^{*np}, y_\eta^{*np}, l_{\mu y}, l_{\eta y}) \\ &= \frac{k_p K_2}{d_T} (T_{\mu y}^*, T_{\eta y}^*, T_{\mu y}^{*np}, T_{\eta y}^{*np}, \frac{d_{\text{IL-10}}}{p_{\text{IL-10}}} \text{IL-10}_\mu, \frac{d_{\text{IL-10}}}{p_{\text{IL-10}}} \text{IL-10}_\eta), \end{aligned} \quad (2e)$$

$$(g_\mu(\tau), g_\eta(\tau)) = \frac{k_a k_p K_1}{d_T^2 (k_p + k_r)} (G_1(d_T^{-1} \tau), G_2(d_T^{-1} \tau)), \quad (2f)$$

$$\begin{aligned} & (h_{\mu x}(\tau), h_{\mu y}(\tau), h_{\eta x}(\tau), h_{\eta y}(\tau)) \\ &= \frac{K_1}{d_T} (H_{\mu x}(d_T^{-1} \tau), H_{\mu y}(d_T^{-1} \tau), H_{\eta x}(d_T^{-1} \tau), H_{\eta y}(d_T^{-1} \tau)), \end{aligned} \quad (2g)$$

$$\begin{aligned} \beta &= \frac{k_a + k_{d_1}}{k_a}, \quad \gamma = \frac{k_p + k_r}{k_a + k_{d_1}}, \quad \delta_{\text{IFN}} = \frac{d_{\text{IFN}}}{k_a + k_{d_1}}, \\ \delta_{\text{IL-10}} &= \frac{d_{\text{IL-10}}}{k_a + k_{d_1}}, \quad \varepsilon = \frac{d_T}{k_a + k_{d_1}}, \quad \kappa = \frac{K_2}{K_1}, \quad \Lambda = \frac{k_{b_2}}{k_{b_1}}, \\ \chi &= \frac{k_p + k_r}{k_p}, \quad \xi = \frac{k_{ns}}{k_p + k_r}, \quad \zeta = \frac{k_{np}}{k_p + k_r}, \quad \rho = \frac{d_R}{d_T}, \\ \theta &= \frac{k_m}{d_T}, \quad \Omega = \frac{k_p K_1}{d_T} \frac{d_{\text{IFN}} K_{\text{IFN}}}{p_{\text{IFN}}}, \\ \phi &= \frac{d_{\text{IL-10}}}{d_{\text{IFN}}} \frac{K_{\text{IL-10}}}{K_{\text{IFN}}} \frac{p_{\text{IFN}}}{p_{\text{IL-10}}}, \quad \nu = \frac{k_{np}}{k_r}. \end{aligned} \quad (2h)$$

The relationship between the time scales is represented by the magnitude of the dimensionless parameter ε . As discussed, $d_T \ll k_a + k_{d_1}$; hence

$\varepsilon \ll 1$. By taking $\varepsilon \equiv 0$, a separation of time scales, we obtain, where $' \equiv d/d\tau$,

$$r'_\mu = g_\mu - \rho r_\mu \quad : \quad rho'_\eta = g_\eta - \rho r_\eta, \quad (3a, b)$$

$$x'_\mu = h_{\mu x} + x_\mu^* - (1 + \theta)x_\mu + \theta x_\eta, \quad (3c)$$

$$y'_\mu = \kappa h_{\mu y} + y_\mu^* - (1 + \theta)y_\mu + \theta y_\eta, \quad (3d)$$

$$x'_\eta = h_{\eta x} + x_\eta^* - (1 + \theta)x_\eta + \theta x_\mu, \quad (3e)$$

$$y'_\eta = \kappa h_{\eta y} + y_\eta^* - (1 + \theta)y_\eta + \theta y_\mu. \quad (3f)$$

Table 1 recapitulates the symbols used in equations (3) and in subsequent text.

In the differential equations (3), as a consequence of the separation of time scales, x_μ^* and y_μ^* can be described as algebraic functions of r_μ , x_μ and y_μ ,

$$y_\mu^* = \frac{\Omega + x_\mu^*}{\Omega + (1 + \zeta)x_\mu^*} \kappa r_{\mu y}, \quad (4a)$$

where x_μ^* is the real positive root of

$$q_{\mu 0} x_\mu^{*2} + q_{\mu x} x_\mu^* - q_{\mu y} = 0. \quad (4b)$$

Here

$$q_{\mu 0} = (1 + \zeta)\Omega\phi + (1 + \xi)\nu r_{\mu y},$$

$$q_{\mu 1} = \Omega[\Omega\phi + (1 + \xi)r_{\mu y}] - [(1 + \zeta)\Omega\phi + \nu r_{\mu y}]r_{\mu x},$$

$$q_{\mu 2} = \Omega[\Omega\phi + r_{\mu y}]r_{\mu x}$$

Table 1. Variables in final model of (6) and (4)

Variables (dimensionless)
x : Th1
y : Th2
r : Restimulator sites
Sub- and superscripts
μ : Mucosal compartment
η : Non-mucosal compartment
$*$: Activated

and

$$(r_{\mu 0}, r_{\mu x}, r_{\mu y}) = \frac{r_{\mu}}{1 + x_{\mu} + y_{\mu}} (1, x_{\mu}, y_{\mu}).$$

Finally, g_{μ} , g_{η} , $h_{\mu x}$, $h_{\mu y}$, $h_{\eta x}$ and $h_{\eta y}$ are the scaled representations of the recruitment functions. The foregoing expressions are duplicated in compartment η . Note that θ represents the degree of connectivity between the two compartments. In the following text we present an investigation of system (3) by numerical simulation.

Final formulation. In order to simulate system (3) we must introduce explicit formulations for the time functions $g_{\mu}, \dots, h_{\eta y}$. At present the functional form of the recruitment dynamics is unknown. Consequently, we shall introduce some approximations. Typical pollen antigens are seasonal. Since exposure to such allergen is occasional, rather than persistent, we shall assume that $g_{\mu}, \dots, h_{\mu y}$ are approximately zero during the immunotherapy phase. We shall represent further exposures to allergen as perturbations.

During immunotherapy the exposure to allergen, by approximately weekly injections, is effectively persistent. We can moreover make the reasonable approximation that the recruitment of allergen-specific T cells, of both types, is proportional to the recruitment of restimulator cells. Denoting the constant of proportionality by f , we have

$$h_{\eta x}(\tau) + h_{\eta y}(\tau) = f \cdot g_{\eta}(\tau). \quad (5a)$$

Next, let us define $p(\tau)$ by

$$p(\tau) = \frac{h_{\eta x}(\tau)}{h_{\eta x}(\tau) + h_{\eta y}(\tau)}, \quad (5b)$$

i.e. $p(\tau)$ represents Th1 cell recruitment as a fraction of the total T-cell recruitment in compartment η . Since successive immunotherapeutic injections introduce fixed doses of antigen, we can approximate $p(\tau)$ by a constant, $0 \leq p \leq 1$.

In approximating the recruitment of restimulator cells (into the non-mucosal compartment) by immunotherapy, we are guided by the results of Golden *et al.* (1980), who compared several regimens (schedules of injections and doses per injection) of immunotherapy, while keeping the total dose of injected antigen constant. In all cases the efficacy of the treatments

was similar. Thus, we conclude that, within a broad range of regimens, it is the total of the antigen injected over time that is important. In other words, we expect that all reasonable functional representations of g_η that result in the same total amount of recruitment over time will have equivalent influences on the dynamics of the model. For reasons of mathematical simplicity we shall use the simplest representation possible, i.e. we shall take the recruitment rate to be constant:

$$g_\eta \approx a_\eta. \quad (5c)$$

Note. Since the constant a_η stands for the time average of the recruitment rate, which is an increasing function of antigen dose, increasing the value of a_η permits us to examine effects of higher antigen doses.

We suppose that, during immunotherapy, exposure to the allergen is relatively negligible, so that for our purposes restimulator site recruitment can be neglected in the mucosal compartment. Thus we set $g_\mu = 0$ in (3a). Our model thus takes the following, final, form:

$$r'_\mu = -\rho r_\mu, \quad (6a)$$

$$r'_\eta = a_\eta - \rho r_\eta, \quad (6b)$$

$$x'_\mu = x_\mu^* - (1 + \theta)x_\mu + \theta x_\eta, \quad (6c)$$

$$y'_\mu = y_\mu^* - (1 + \theta)y_\mu + \theta y_\eta, \quad (6d)$$

$$x'_\eta = f a_\eta \cdot p + x_\eta^* - (1 + \theta)x_\eta + \theta x_\mu, \quad (6e)$$

$$y'_\eta = f \kappa a_\eta \cdot (1 - p) + y_\eta^* - (1 + \theta)y_\eta + \theta y_\mu. \quad (6f)$$

As a consequence of the separation of time scales, x_μ^* , y_μ^* , x_η^* and y_η^* can be expressed in terms of the remaining variables; see equations (4). The cross suppression of Th1 (x^*) and Th2 (y^*) is implicit in these algebraic equations.

The variables are summarized in Table 1. Information concerning the various dimensionless parameters of the model is collected in Table 2. In interpreting a parameter (Table 2, column 3) we choose its salient feature, but implicit is another feature of the same dimension. For example, in a fuller description ρ is the "restimulator cell decay rate" d_R , as stated, divided by the T-cell death rate d_T .

Numerical investigation. Some of the parameter values—those that pertain to the intracompartamental Th1/Th2 cross-regulation and those that

Table 2. Parameters in final model of (6) and (4)

Parameter	Definition ^a	Interpretation	Control value	Reference ^b
ρ	(2h)	Restimulator cell decay rate	0.01	Text, under (3)
θ	(2h)	Intercompartment migration rate	0.01	Fig. 7
f	(5a)	Ratio restimulator cell to T memory recruitment	0.1	Fig. 6
a_η	(5c)	Restimulator recruitment rate in immunotherapy	1	Fig. 5
p	(5b)	Th1 recruitment	1	Fig. 4
ζ	(2h)	Efficiency, Th2* inhibition by IFN- γ	1	FP'94
ξ	(2h)	Efficiency, Th1* inhibition by IL-10	9	FP'94
Ω	(2h)	Half saturation, Th2* inhibition by IFN- γ	1	FP'94
κ	(2h)	Relative strength of Th2 vs. Th1 activation	0.3	FP'94
ϕ	(2h)	$\Omega\phi\kappa$: half saturation, Th1* inhibition by IL-10	1	FP'94
ν	(2h)	Relates IL-10 concentration to Th2*	3	FP'94

^aEquation where parameter is defined.

^bFP'94 = Fishman and Perelson (1994). When a figure is given, the behavior shown in that figure allows us to pick a control value.

pertain to the restimulation dynamics—are “carried over” from Fishman and Perelson (1994, 1995). At present we shall examine the effects that the magnitudes of the *phenomenological* parameters θ , a_η , p and f have on the dynamics of the system.

The parameter $\theta = k_m/d_T$, which represents the connectivity between the two compartments, is the central component of the current model. We chose to focus on this aspect of the immunological mechanisms involved in immunotherapy because, in our view, the extent of intercompartmental lymphocyte traffic determines the observed properties of the system. The observations of Kroemer *et al.* (1993) indicate that traffic cannot be extensive. Otherwise, introduction of antigen into mucosal surfaces would lead to systemic immunity, i.e. systemic Th2 dominance. On the other hand, if there is no lymphocyte traffic between compartments, then the systemic decrease in IgE production (an estimator of the allergen-specific Th2 activity) observed in immunotherapy would not be expected to occur. In the present model we attempt to give these qualitative considerations a quantitative representation. However, before we proceed to simulate the system's

behavior for different values of θ , we must examine the effects of the other parameters and thereby determine their allowable ranges.

In Fig. 3a a simulation of an immunotherapeutic treatment, represented by switching the value of a_η from $a_\eta = 0$ to $a_\eta = 1.0$, is presented. We see that the antigen-specific Th1 populations (x_η and x_μ) increase with time, whereas the antigen-specific Th2 populations (y_η and y_μ) decrease. In the presentation of the experimental observations (Fig. 1) only systemic concentrations are reported. Thus, in Fig. 3b, we present the same results in terms of the total Th1 and Th2 concentrations, $x = x_\mu + x_\eta$, and $y = y_\mu + y_\eta$. Finally, in order to be able to compare the dynamics of our model under varying simulation conditions, we need a one-dimensional estimator. We choose the Th1 fraction $x/(x + y)$ (Fig. 3c) to be this estimator.

In Fig. 3 we took p , the probability that a T cell recruited into compartment η will mature into a Th1 cell, to be unity. In Fig. 4 we examine the robustness of this assumption by varying p , while keeping the rest of the parameters constant. We see that there is a threshold in p . That is, if the fine antigen-specificity differences between (allergen-specific) Th1 and Th2 cells are such that less than 80% of the T cells recruited into compartment η mature into Th1 cells, then immunotherapy will not be successful. However, since for $p \geq 0.8$ the simulation "succeeds," we shall use $p = 1.0$ for subsequent simulations.

In Fig. 5 we examine the effects of varying the magnitude of a_η , which represents the dosage rate (allergen per unit time) of the immunotherapeutic treatments, on the outcomes of the simulated treatments. We see that the eventual outcome, although not the treatment time, is quite independent of the dosage rate. Th1 dominance is attained for all dosage rates, but more rapidly at higher doses. For each dose there is a critical time t_c such that if treatment is discontinued after t_c the Th1 dominance is maintained (cure, according to our model) but relapse to Th2 dominance occurs if the treatment is discontinued before t_c . For example, if $a_\eta = 0.5$, $f = 1$, $\rho = 0.001$ and the other parameters have control values then t_c is of magnitude 10 (definitely between 2.5 and 25).

A possibly non-trivial result is that the dependence of the desired result on the dosage rate is saturable. That is, after the dosage rate exceeds a certain value there is a negligible improvement in the time it takes to achieve effective replacement of Th2 by Th1. Thus, it appears to be possible, as has indeed been found experimentally (Lichtenstein, 1993), to find a roughly optimal dosage rate, such that one can achieve the desired result with a minimal allergen dose over time.

In Figs. 3–5 we have taken the value of f , which represents the proportionality between restimulator cell recruitment and T-cell recruitment, as $f = 0.1$. We examine the validity of this estimate by varying f through 2

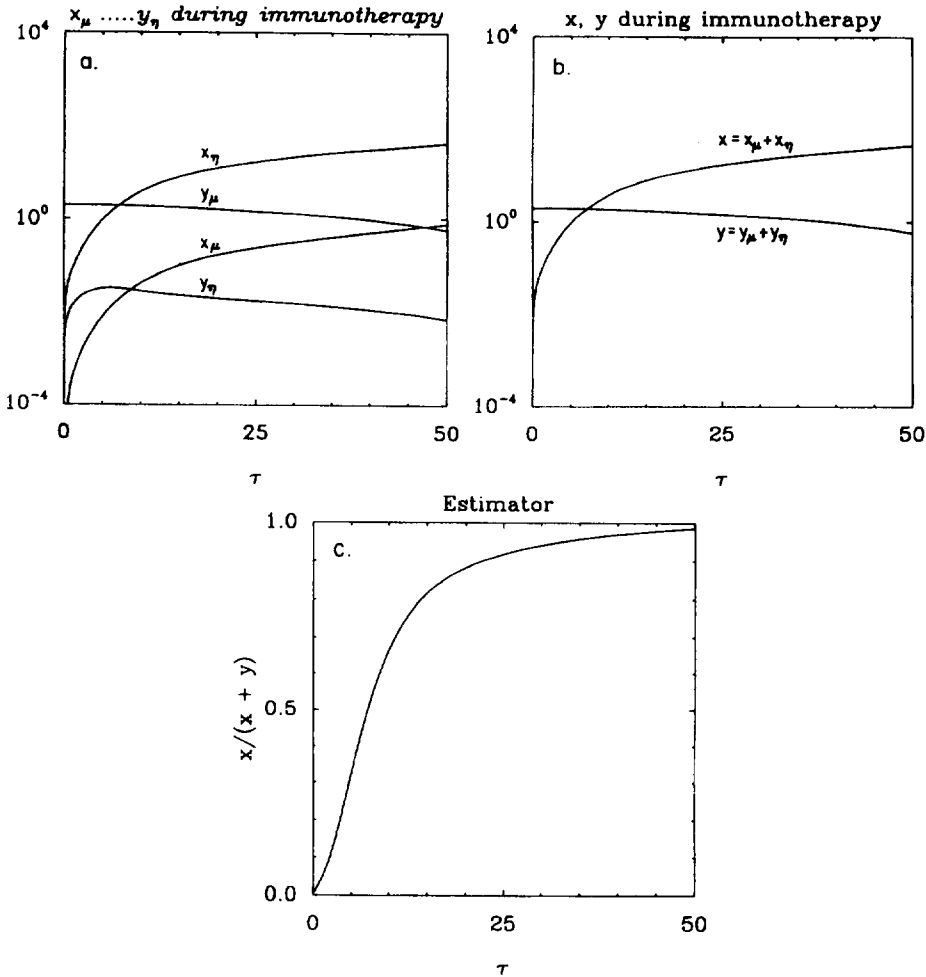


Figure 3. Numerical simulation of immunotherapy. The simulation starts at the steady state representing allergy, i.e. $r_\mu(0) = 10$, $r_\eta(0) = 0.0$, $x_\mu(0) = 0.0$, $x_\eta(0) = 0.0$, $y_\mu(0) = 1.975$, $y_\eta(0) = 1.955 \times 10^{-3} = y_\mu(0) \times \theta/(1 + \theta)$. See Table 2 for parameter values. (a) Dynamics of $x_\mu(\tau), \dots, y_\eta(\tau)$ as immunotherapy is initiated (by switching the value of a_η , the restimulator cell recruitment rate, from 0.0 to 1.0). Note the delay between the increase in the non-mucosal Th1 concentration $x_\eta(\tau)$ and the subsequent increase in mucosal Th1 concentration $x_\mu(\tau)$. This delay is prescribed by the rate of the intercompartmental lymphocyte traffic. (b) Dynamics of the systemic scaled concentrations ($x(\tau)$ and $y(\tau)$) of the antigen-specific Th1/Th2 cells. Since a τ unit corresponds to a T-cell life-span (approximately 50 days), the results are roughly in agreement with Fig. 1. (c) The dynamics of the one-dimensional estimator, $x(\tau)/(x(\tau) + y(\tau))$, of the systemic Th1/Th2 dynamics.

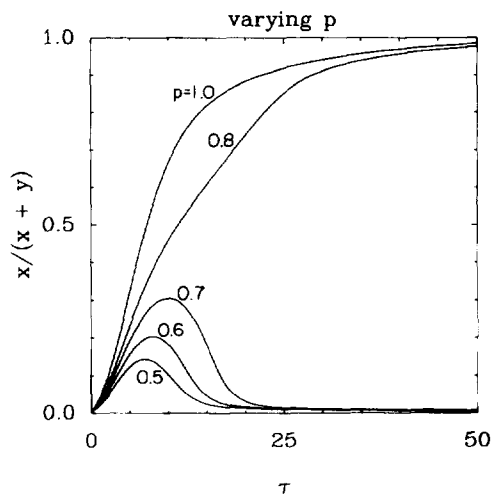


Figure 4. The effects of the magnitude of the parameter p that represents the relative fraction of the T-cell recruitment that is dedicated to recruitment of Th1 cells. Other parameters as in Fig. 3.

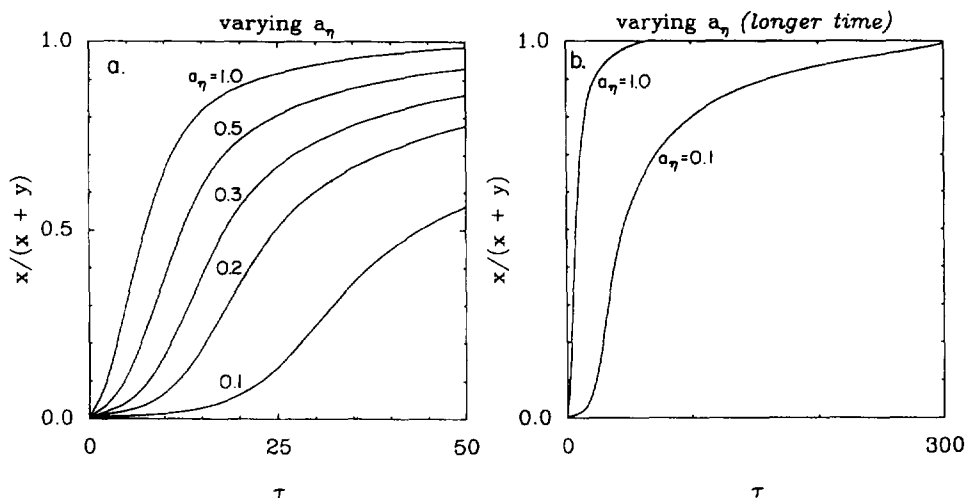


Figure 5. The effects of the magnitude of restimulator cell recruitment rate, a_η , on model dynamics. Other parameters as in Fig. 3. (a) Values of a_η , varying over an order of magnitude, lead to systemic Th1 dominance. (b) Long-term dynamics for two extreme values. As can be seen, both produce the same result, albeit at different rates.

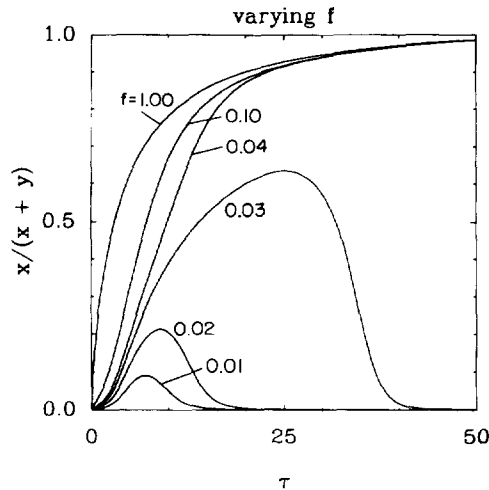


Figure 6. The effects of the magnitude of the proportionality constant f that relates restimulator cell recruitment rate to the Th1/Th2 recruitment rates. Other parameters as in Fig. 3.

orders of magnitude. As can be seen in Fig. 6, the value $f = 0.1$ is within the range of f values for which a simulation successfully predicts immunotherapy. The reason that a sufficiently large value of f is required for successful immunotherapy, according to our model, is because sufficient restimulator recruitment is required to give effect to the higher probability that a recruited cell will be Th1 (sufficiently large p).

In Fig. 7 we present an investigation of the effect of the values of θ . The parameter $\theta \equiv k_m/d_T$ determines the probability of intercompartmental migration. If θ is (relatively) small, i.e. k_m , the probability of migration, is small relative to the probability of cell death d_T , then the two compartments will be essentially independent of each other (Fig. 7a). That is, compartment μ will remain Th2 dominated, whereas compartment η will be Th1 dominated. In terms of the systemic concentrations of the antigen-specific IgE and IgG antibodies, we would expect to see an increase in IgG without a decrease in IgE. If θ is relatively large, i.e. the probability of migration approaches the probability of cell death, then the system will behave as essentially one-compartmental, i.e. it will be impossible to switch from Th2 dominance to Th1 dominance (Fig. 7b).

Thus, the magnitude of θ reflects two opposing tendencies. When θ is small, a Th1-dominated response in compartment η is easy to establish. However, the mucosal compartment remains Th2 dominated. If θ is large, then, due to migration, compartment η contains a high concentration of Th2 cells. Thus, Th1 response cannot be established. A quantitative exami-

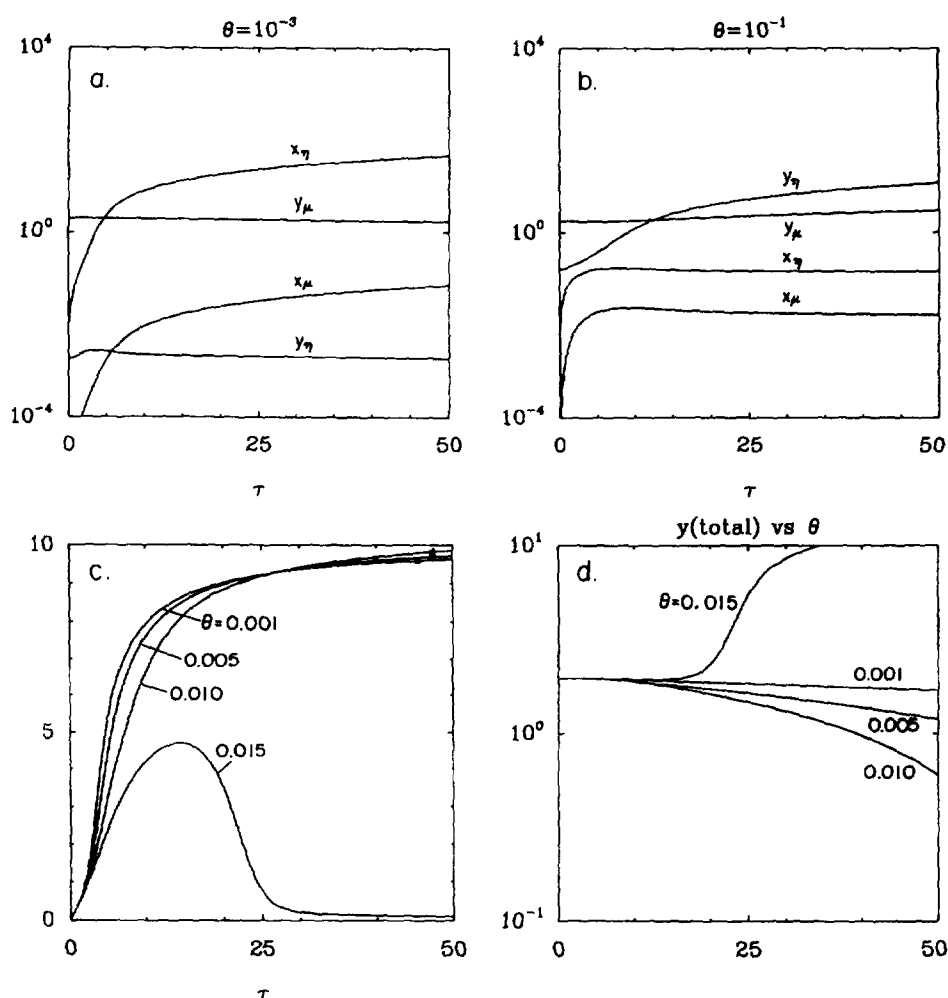


Figure 7. The effects that the magnitude of θ , a parameter that represents the per capita rates of intercompartmental lymphocyte traffic, on the outcomes of immunotherapy. Other parameters as in Fig. 3. (a) Dynamics of $x_\mu(\tau), \dots, y_\eta(\tau)$ when $\theta = 0.001$. (b) Dynamics of $x_\mu(\tau), \dots, y_\eta(\tau)$ when $\theta = 0.1$. (c) Dynamics of $x(\tau)/(x(\tau) + y(\tau))$ when θ is varied. (d) Dynamics of $y(\tau)$ corresponding to $x(\tau)/(x(\tau) + y(\tau))$ dynamics depicted in (c).

nation of the foregoing considerations is presented in Fig. 7c and d, where the dynamics of (i) the ratio $x/(x + y)$ (Fig. 7c) and (ii) y (Fig. 7d) for various values of θ are presented. Figure 7d is made necessary by the fact that accumulation of Th1 cells, which is due to immunotherapy, is higher by 2 orders of magnitude than the pre-immunotherapy level of Th2 cells (Fig. 1). In particular, examination of these figures indicates that the value

$\theta = 10^{-2}$, used in Figs. 3–6, is reasonable. Note also that Fig. 7d indicates that significant changes in Th2 levels take on the order of θ^{-1} time units, i.e. θ^{-1} T-cell life-spans.

4. Discussion. Allergies are of considerable medical interest since they constitute a nuisance to many people and are life-threatening to some. Although detailed study of allergies has (so far) been somewhat limited, allergies are also interesting from a basic science point of view. The reason is that allergies seem to represent a failure of the immune system. Unlike auto-immune disease, the problem is not a failure to distinguish between self and non-self, but rather an inappropriate response to a foreign antigen. Probing the reasons for inappropriate responses, and for their cure, is of course one way to clarify immune mechanisms.

Here we have presented a model that combines Th1/Th2 cross-regulation and immune memory maintenance—as modeled by Fishman and Perelson (1994, 1995)—with a simple two-compartment representation of the key effects of spatial inhomogeneity. A study such as this is more convincing if it is based on a carefully thought out representation of Th1/Th2 cross-regulation, but we expect that our major conclusions will not change if experiment reveals that there must be changes in our assumptions concerning how the regulation operates. This expectation flows from the fact that the 22 differential equations of our original model have been boiled down to just 6 in the final model [equations (6a)–(6f)]. It is qualitative features of this model that determine its overall behavior; other assumptions would lead to similar qualitative features.

To recapitulate, available evidence, reviewed in section 2, indicates that immunotherapeutic phenomena involve (i) Th1/Th2 cross-regulation and (ii) compartmentalization of the immune system. The key point of evidence is the fact that for certain antigens, presumably including the common allergens, immune responses to an antigen introduced in different sites differ with respect to the eventual Th1/Th2 dominance (Kroemer *et al.*, 1993).

In these terms the difference in immunoglobulin isotypes observed between “natural” exposure to an allergen and immunotherapy is interpreted as follows. Allergen enters the patient through mucosal surfaces, which initiates an immune response in the lymph nodes draining these surfaces. In susceptible persons, i.e. persons that possess certain HLA haplotypes (Marsh *et al.*, 1982), this response becomes dominated by a Th2 subset. Th2 dominance leads to the establishment of IgE production. Immunotherapy, presumably, introduces the allergen into non-mucosal lymph nodes and may result in a Th1 dominated response there. Subse-

quent migration of allergen-specific Th1 cells, induced by immunotherapy, into the mucosal compartment can overwhelm the local Th2 response, which yields systemic Th1 dominance and IgG production.

In our model we have focused on the extent of the traffic between the compartments. The observations of Kroemer *et al.* (1993) indicate that traffic cannot be extensive. Otherwise, introduction of an antigen into mucosal surfaces would lead to a systemic immunity, i.e. systemic Th2 dominance. On the other hand, if there is no lymphocyte traffic between the two compartments, then the systemic decrease in IgE production (an estimator of the allergen-specific Th2 activity) observed in immunotherapy would not be expected to occur. In the present model we have given these qualitative considerations a quantitative representation.

One result from our qualitative analysis is the demonstration that, for certain parameter values, such as $p < 0.8$ (Fig. 4), immunotherapy will not be successful, and in fact will lead to the exacerbation of allergy, i.e. higher Th2 levels in the presence of therapy (not shown), as indeed has been observed for some patients (Graft *et al.*, 1987). This is due to the fact that the magnitude of the phenomenological parameter p stands for the diversity of factors that determine the relative recruitment of Th1 and Th2 cells by immunotherapeutic injections. Now, as discussed in Fishman and Perelson (1994), Th2 cells are by order of magnitude more effective in suppressing Th1 response than vice versa. Thus, if the value of p is less than a certain threshold value (0.8 for these particular parameters), the ratio of recruited Th2 cells to recruited Th1 cells is such that Th2 cells eventually dominate. Possibly a more detailed model of the process will be able to point out specific factors contributing to this failure of immunotherapy.

A remarkable feature of immunotherapy is that the treatments are lengthy—persisting for periods of years. This is connected with the fact that expansion of IgG production is not sufficient for the abrogation of the allergy. The time for patient cure is correlated with decrease in IgE production (Ishizaka, 1976), i.e. with the time when Th2 dominance in the mucosal compartment is switched to Th1 dominance. This process, which follows the establishment of Th1 dominance in the non-mucosal compartment, is governed by the extent of lymphocyte traffic between compartments. However, as shown by numerical investigation this traffic must be low, otherwise Th2 cells would dominate both compartments. For our control parameter value $\theta = 0.01$, during its lifetime a Th1 cell in compartment η has only a 1% chance of migrating into compartment μ . Consequently, replacement of Th2 cells by Th1 cells takes several tens of T-cell life-spans of (with an approximately 50 day life-span) several years.

Understanding immunotherapy for allergy is a difficult matter; at best our contribution can be but a small step in the right direction; a great many

issues remain to be explored. We will comment on just two of these issues; immunotherapy for insect stings and the role of Th1 and Th2.

Our explanation for immunotherapy has relied on the fact that pollen allergens enter via the mucosal compartment while immunotherapy involves injections into a spatially separate non-mucosal compartment. However, what of insect stings, which are also injected? Immunotherapy for bee stings has the same gross effects as immunotherapy for pollen allergies (Graft *et al.*, 1987). We believe that our basic idea that the essence of the matter is weakly interacting heterogeneous T-helper populations can explain matters here too. To show this would require at least an additional paper, but we can briefly sketch certain principles.

Let us, for definiteness, consider immunotherapy for a bee sting. It is known that the retained antigen is predominantly located in lymph nodes draining the site of antigen introduction (Tew *et al.*, 1980). Thus, unless the immunotherapy patient is injected at the site of the bee sting, the sting itself and immunotherapy will create two spatially distinct populations of antigen-presenting cells. A recent experimental report (Hosken *et al.*, 1995) indicates that recruitment of virgin T cells into either Th1 or Th2 classes is determined by the density of peptide-MHC complexes on an antigen-presenting cell. It is reasonable to assume that the density of peptide-MHC complexes varies with the circumstances of antigen introduction. Moreover, a bee sting introduces antigen amidst a variety of ancillary factors and thus in a fashion differing from the introduction of an antigen by immunotherapy. Consequently, it is likely that the two populations of antigen presenting cells (due to the sting versus due to immunotherapy) will differ in Th1 versus Th2 recruitment, as well as in their spatial location. A mathematical model of such a situation could be produced by straightforward modification of our present model.

In the older allergy literature stress is placed on IgG and IgE levels. We regard these levels first and foremost as indicators of the relative dominance of Th1 and Th2. Which of the multiple effects of Th1 and Th2 cells are the key to immunotherapy remains unclear, with new ideas frequently emerging. (For example Daëron *et al.* (1995) find evidence that the proper class of IgG may be very important; of course class switching is regulated by Th cells.)

According to our model, failure of immunotherapy can be transformed into success by suitable manipulation of the Th1 recruitment parameter p or the proportionality factor f between restimulator cell and Th1/Th2 recruitment rates. More robust is the model's demonstration that suitable quantitative changes in the balance between Th1 and Th2 can result in a switch to Th1 and hence successful immunotherapy.

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