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Immune response dynamics

N.J. Burroughs [a], B.M.P.M. Oliveira [b], A.A. Pinto [c,d], M. Ferreira [e,d,*]

[a] *Mathematics Institute, University of Warwick, UK*

[b] *Faculdade de Ciências da Nutrição e Alimentação da Universidade do Porto, Portugal*

[c] *Fundação para a Ciência e a Tecnologia, Portugal*

[d] *Departamento de Matemática da Escola de Ciências da Universidade do Minho, Portugal*

[e] *Escola Superior de Estudos Industriais e de Gestão, Instituto Politécnico do Porto, Portugal*

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ABSTRACT

The consequences of regulatory *T* cell (Treg) inhibition of interleukine 2 secretion are examined by mathematical modelling. We determine the analytic formula that describes the fine balance between Regulatory *T* cells and *T* cells at controlled and immune response equilibrium states. We demonstrate that cytokine dependent growth exhibits a quorum *T* cell population threshold that determines if immune responses develop on activation. We determine the analytic formulas of *T* cell proliferation thresholds that allow us to study the sensibility of the quorum growth thresholds controlling immune responses.

1. Introduction

Regulatory *T* cells, or Tregs, have emerged over the last decade as a fundamental component of the *T* cell repertoire, being generated in the thymus under positive selection by self-peptides [1].

In [2], it is proposed a dynamical model to explain how the delicate balance between appropriate immune activation and immune response suppression is achieved. In [3] we present analytic formulas that are used to study the biological effect of each parameter in the model. In this paper, we prove the above-mentioned results. The analytic formula describing the fine balance between Regulatory *T* cells and *T* cells at controlled and immune response equilibrium states allows us to study geometrically the sensitivity of the equilibria with the parameters. When the antigenic stimulation b of *T* cells rises above the threshold b_H control is lost and autoimmunity arises. After an autoimmune response, the control state is recovered when the stimulation falls below a lower threshold b_L. This phenomenon is due to the equilibria manifold being a hysteresis. The transition between one state and the other results from a sudden change of the dynamics. The antigenic thresholds b_L and b_H of *T* cells bound the biphasic behavior of our immune response model. This means that we have two stable regions. Connecting the two stability regions there is an unstable manifold. A cross section of the equilibria manifold is an *s* shaped curve (see Fig. 3) usually designated hysteresis. Some parameters unfold the hysteresis of the antigenic stimulation of *T* cells at a cusp. Here, we present an explicit condition for the appearance of a cusp bifurcation. This is a biologically relevant property of the model because the number of stable and unstable equilibria changes when the hysteresis unfolds with those parameters.

* Corresponding author at: Escola Superior de Estudos Industriais e de Gestão, Instituto Politécnico do Porto, Portugal.
E-mail addresses: bmpmo@fcna.up.pt (B.M.P.M. Oliveira), migferreira2@gmail.com (M. Ferreira).

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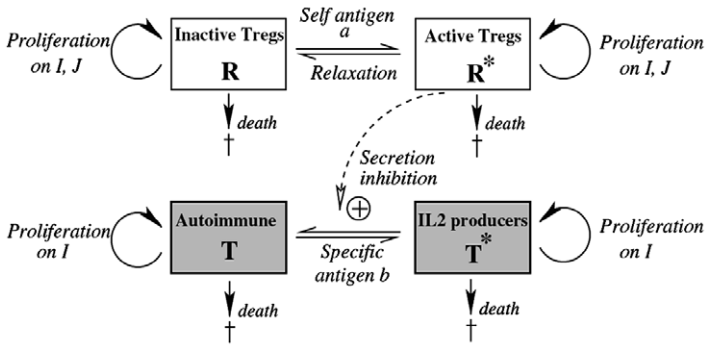

Fig. 1. Model schematic showing growth, death and phenotype transitions of the Treg populations R, R^*, and autoimmune T cell T, T^* populations. Cytokine dynamics are not shown: IL-2 (denoted I) is secreted by activated T cells T^*, adsorbed by all the T cell populations equally, cytokine J is produced from a tissue source and adsorbed by Tregs only.
Source: Reproduced from [2].

2. Theory

The model in [2] uses a population of Tregs and conventional T cells with processes shown schematically in Fig. 1. Both populations require antigenic stimulation for activation. Levels of antigenic stimulation are denoted a and b for Tregs and conventional T cells respectively. Tregs are activated by self-antigens from an inactive state, denoted R, to an active state R^*. The IL-2 secreting T cells are denoted T^* and the non-secreting T cells are denoted T. On activation conventional T cells secrete IL-2 and acquire proliferative capacity in the presence of IL-2. Tregs also proliferate in the presence of IL-2 although less efficiently than normal T cells [4], and they do not secrete IL-2. The model uses a generic mechanism that utilizes a cytokine (denoted J), analogous to interleukine 7 which is known to homeostatically regulate memory T cells [5]. Finally, we include an influx of (auto) immune T cells into the tissue (T_{input}), which can represent T cell circulation or naive T cell input from the thymus.

The model consists of a set of ordinary differential equations that is employed to study the dynamics, with a compartment for each T cell population (inactive Tregs R, active Tregs R^*, non-secreting T cells T, secreting activated T cells T^*), interleukine 2 density I and the homeostatic Treg cytokine J:

$$\begin{aligned}
\frac{dR}{dt} &= (\epsilon\rho(I+J) - \beta(R+R^*+T+T^*) - \hat{d})R + \hat{k}(R^* - aR),\\
\frac{dR^*}{dt} &= (\epsilon\rho(I+J) - \beta(R+R^*+T+T^*) - \hat{d})R^* - \hat{k}(R^* - aR),\\
\frac{dJ}{dt} &= \hat{\sigma}(S - (\hat{\alpha}(R+R^*) + \hat{\delta})J),\\
\frac{dT}{dt} &= (\rho I - \beta(R+R^*+T+T^*) - d)T + k(T^* - bT + \gamma R^*T^*) + T_{\text{input}},\\
\frac{dT^*}{dt} &= (\rho I - \beta(R+R^*+T+T^*) - d)T^* - k(T^* - bT + \gamma R^*T^*),\\
\frac{dI}{dt} &= \sigma(T^* - (\alpha(R+R^*+T+T^*) + \delta)I).
\end{aligned} \tag{1}$$

Parameters are defined in Table 1. Our model has components that have been used in previous models, for instance cytokine dependent growth [11,12], cytokine kinetics [13], Fas–FasL mediated death [14], and positive feedback of T cells on Tregs [15,16], in model [2] this is explicitly through IL-2. The important aspects of this model are a mechanism to sustain a population of Tregs, secretion inhibition of T cells with a rate that correlates with Treg population size, and growth and competition for IL-2 with a higher growth rate of T cells relative to Tregs.

3. The fine balance between Tregs and T cells

We study the equilibria of the immune system in a neighbourhood of the default values for the parameters and variables. The concentration of T cells varies between a minimum and a maximum value. The minimum value corresponds to the *homeostasis* concentration of T cells T_{hom}, i.e. when there is no antigenic stimulation of T cells ($b = 0$). The maximum value is called the *capacity* of T cells T_{cap}, and is obtained for high levels of antigenic stimulation of T cells ($b = +\infty$). Using Theorem 2, the values T_{hom} and T_{cap} are implicitly determined as zeros of a polynomial. In particular, for the default values of the parameters, these values are given by $T_{\text{hom}} = 9.6 \times 10^2$ cells/ml and $T_{\text{cap}} = 9.7 \times 10^6$ cells/ml. When the system

Table 1
Model parameters.
Source: Reproduced from [2].

Parameter	Symbol	Range	Value
T cell T, T^*			
T cell maximum growth rate[a]	ρ/α	<6 day^{-1}	4 day^{-1}
Death rate of T cells	$d = \hat{d}$	0.1–0.01 day^{-1} [6]	0.1 day^{-1}
Capacity of T cells[b]	$\rho/(\alpha\beta)$	10^6–10^7 cells/ml [7]	10^7 cells/ml
Input rate	T_{input}	0–10^4 cells/ml/day	0, 100 cells/ml/day
Secretion reversion (constant)[c]	k	hours–days	0.1 h^{-1}
Antigen stimulation level	bk	0.001–$200 \times a\hat{k}$	Bifurcation parameter
Tregs R, R^*			
Growth rate ratio $T_{\text{reg}} : T$	ϵ	<1	0.6
Homeostatic capacity R_{hom}	$(\epsilon\rho S/\hat{d} - \hat{\delta})/\hat{\alpha}$	10–10^5 cells/ml	10^4 cells/ml
Relaxation rate	$\hat{k}$	hours–days	0.1 h^{-1}
Death rate ratio $T_{\text{reg}} : T$	$\hat{d}/d$		1
T_{reg} antigen stimulation level	$a\hat{k}$	0–10 per day	1 per day
Secretion inhibition[d]	γ	0.1–$100 \times R_{\text{hom}}^{-1}$	$10\, R_{\text{hom}}^{-1}$
Cytokines			
Max. cytokine concentration[e]	$1/\alpha$	100–500 pM	200 pM
IL-2 secretion rate	σ	[f]0.07, 2 fgrms h^{-1} [8]	10^6 molecules s^{-1} cell^{-1}
Relative adsorbance J to IL-2	$\hat{\sigma}\hat{\alpha}/\sigma\alpha$	<1	0.1
Relative secretion rate of J	$\hat{\sigma}/\sigma$	<1	0.01
Cytokine decay rate	$\sigma\delta = \hat{\sigma}\hat{\delta}$	hours–days	1.5 h^{-1} [9]

[a] Minimum duration of SG_2M phase $\alpha\rho^{-1} \approx 3$ h.
[b] Maximum T cell density for severe infections (based on LCMV).
[c] This is in absence of Tregs.
[d] This is in terms of the homeostatic Treg level R_{hom} which we set to 10^4 cells/ml..
[e] This is taken as 20 times the receptor affinity (10 pM [10]).
[f] Naive and memory cells respectively. This corresponds to 3000–10^5 molecules per h, IL-2 mass 15–18 kDa.

is at equilibrium, we present, in Theorem 1, the relation between the concentration of T cells and the concentration of the Tregs for values of the concentration of T cells between T_{hom} and T_{cap} (see Fig. 2).

Let Y_1, Y_2, Y_3 be the following polynomials

$$Y_1(x) = -\hat{\alpha}C(x) - \beta\hat{\delta}B(x)$$
$$Y_2(x) = 2\hat{\alpha}\beta B(x)$$
$$Y_3(x) = Y_1^2(x) - 2(\delta C(x) - \epsilon\rho Sx)Y_2(x),$$

where $B(x) = (1-\epsilon)x$ and $C(x) = \epsilon T_{\text{input}} + B(x)(\beta x + d)$.

Let X_1, X_2, X_3 be the following polynomials

$$X_1(y) = \hat{B}\hat{C}(y)\hat{D}(y) - \epsilon\rho S$$
$$X_2(y) = -2\beta\hat{B}\hat{C}(y)$$
$$X_3(y) = X_1^2(y) - 2\epsilon T_{\text{input}}\hat{C}(y)X_2(y),$$

where $\hat{B} = 1 - \epsilon$ and $\hat{C}(y) = \hat{\alpha}y + \hat{\delta}$, and $\hat{D}(y) = \beta y + d$.

Let $x = T + T^*$ be the total concentration of T cells and $y = R + R^*$ be the total concentration of Tregs.

Theorem 1. *When the system is at equilibrium, the concentration of Tregs $y = R + R^*$ is given by the* Treg curve

$$y = Y(x) = \frac{Y_1(x) + \sqrt{Y_3(x)}}{Y_2(x)}, \tag{2}$$

where $x = T + T^*$ is the total concentration of T cells. Conversely, the concentration of T cells $x(y)$ is either determined by

$$x = X_-(y) = \frac{X_1(y) - \sqrt{X_3(y)}}{X_2(y)} \quad \textit{or} \tag{3}$$

$$x = X_+(y) = \frac{X_1(y) + \sqrt{X_3(y)}}{X_2(y)}. \tag{4}$$

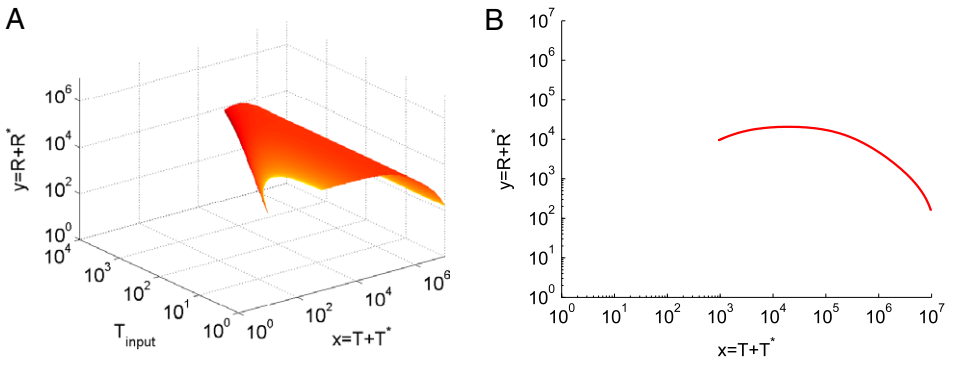

Fig. 2. A: The equilibria manifold for Thymic inputs $T_{\text{input}} \in [1, 10\,000]$. Low values of b are darker and higher values are lighter. B: Cross section of the equilibria manifold for $T_{\text{input}} = 100$. It illustrates Theorem 1, showing the total concentration of Tregs $y(x) = R + R^*$ as a function of the total concentration of T cells $x = T + T^*$. The parameters are at their default values.

See proof of Theorem 1 in Appendix. For simplicity of notation we write $y(x)$ instead of $y = Y(x)$. We also write $x(y)$ when either $x = X_-(y)$ or $x = X_+(y)$ should be used.

The maximum concentration $R_{\max}$ of Tregs is a zero of a fourth order polynomial $X_3(y)$ and, so, $R_{\max}$ has an explicit solution. In particular, for the default values of the parameters, the maximum concentration $R_{\max}$ of Tregs is given by $R_{\max} = 2.1 \times 10^4$ cells per ml, and the corresponding concentration of T cells is given by $x(R_{\max}) = 1.9 \times 10^4$ cells per ml. The minimum concentration $R_{\min}$ of Tregs is given by $R_{\min} = 156$ cells per ml, and the corresponding concentration of T cells is given by $T_{\text{cap}} = 9.7 \times 10^6$ cells per ml.

4. The equilibria manifold

When the system is at equilibrium, we obtain the level of the antigenic stimulation $b(x, y(x))$ of T cells from the concentration x of the T cells, using the auxiliary Treg curve $y(x)$ (see Theorem 1 and Fig. 3). Let the *antigen function* $b(x, y)$ be given by

$$b(x, y) = \frac{\varphi(x, y)(kx(1 + \gamma A y) + T_{\text{input}})}{k(1 - \epsilon)\rho x^3(\hat{\alpha} y + \hat{\delta}) - kx\varphi(x, y)}, \tag{5}$$

where $A = a/(1 + a)$ and $\varphi(x, y) = (\epsilon\rho S x - T_{\text{input}}(\hat{\alpha} y + \hat{\delta}))(\alpha(x + y) + \delta)$.

Theorem 2. *Let $b(x, y)$ be the antigen function, and let $x(y)$ and $y(x)$ be as in Theorem 1. The level of the antigenic stimulation of T cells is given by $b(x, y(x))$, or, equivalently, by $b(x(y), y)$, when the system is at equilibrium (stable or unstable). Conversely, given an antigenic stimulation level b of T cells, the concentration x of T cells and the concentration y of Tregs are zeros of the twelfth order polynomials that can be explicit constructed.*

See proof of Theorem 2 in Appendix.

In Theorem 3, we obtain the level of the antigenic stimulation of T cells from the concentration x of the T cells, for the simplified model without Tregs (see Fig. 3D). Let $\tilde{b}(x)$ be the antigen function in the absence of Tregs given by

$$\tilde{b}(x) = \frac{(\alpha x + \delta)(kx + T_{\text{input}})(\beta x^2 + \mathrm{d}x - T_{\text{input}})}{kx(\rho x^2 + (T_{\text{input}} - \beta x^2 - \mathrm{d}x)(\alpha x + \delta))}. \tag{6}$$

Theorem 3. *Let us consider the simplified model with the concentration of Tregs equal to zero (i.e. $y = 0$). The level of the antigenic stimulation of T cells is given by $\tilde{b}(x)$, when the system is at equilibrium (stable or unstable). Conversely, given an antigenic stimulation level b, the concentration x of T cells is a zero of the fourth order polynomial that can be explicit constructed.*

See proof of Theorem 3 in Appendix.

5. Sensibility of the antigenic thresholds

When the system is at equilibrium, the threshold values of antigen stimulation b_L and b_H of T cells are determined using zeros of a polynomial. The *antigen threshold function* $V(x, y, z)$ is equal to $V_6(y, z)x^6 + \cdots + V_0(y, z)$, where the functions

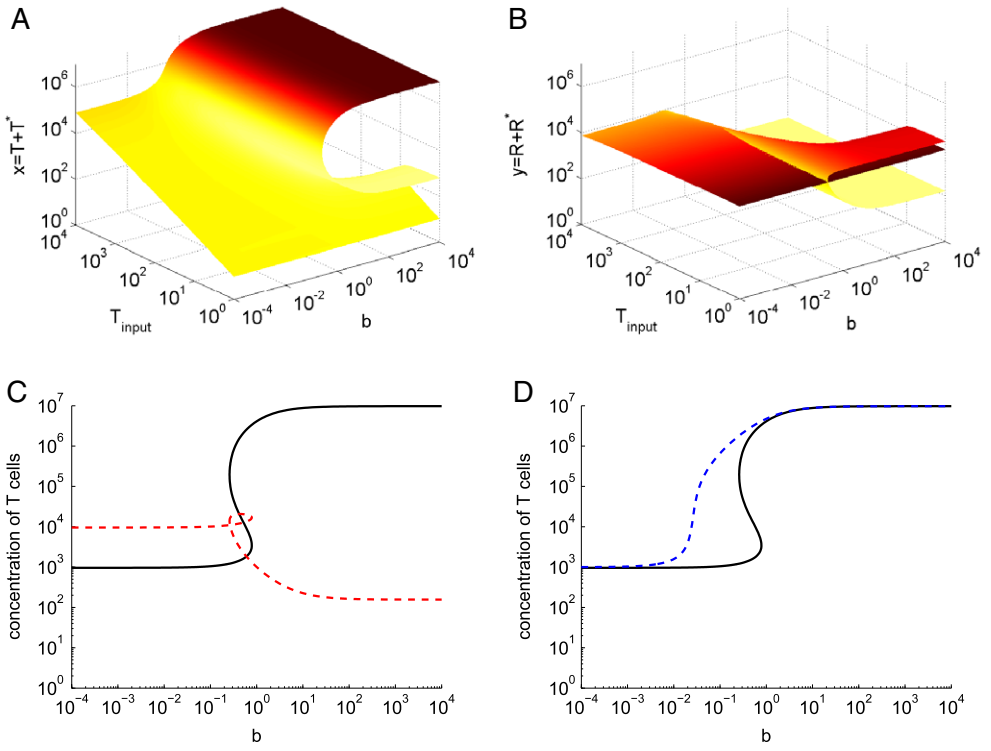

Fig. 3. The hysteresis of the equilibria manifold for Thymic inputs $T_{\text{input}} \in [1, 10\,000]$, with the other parameters at their default values. These figures show the relation between the antigenic stimulation level b, the concentration of T cells $x = T + T^*$, and the concentration of Tregs $y = R + R^*$. The hysteresis unfolds for high values of the parameter T_{input}. A: Low values of $y = R + R^*$ are darker and higher values are lighter. B: Low values of $x = T + T^*$ are darker and higher values are lighter. C: Cross section of the equilibria manifold for $T_{\text{input}} = 100$, illustrating Theorem 2, with the concentration of T cells x (black solid line) and the concentration of Tregs y (red dashes). D: Cross section of the equilibria manifold for $T_{\text{input}} = 100$, illustrating Theorem 3, with the concentration of T cells x (blue dashes) for the simplified model without Tregs. We also show the concentration of T cells x (black solid line) from Theorem 2.

$V_0(y, z), \ldots, V_6(y, z)$ are given by the polynomials

$$V_0(y,z) = kC^2F^2T_{\text{input}}^3$$
$$V_1(y,z) = 2kCFT_{\text{input}}^2H$$
$$V_2(y,z) = kT_{\text{input}}(H^2 + CFT_{\text{input}}(3\rho CE - \gamma ACFkz - 2\alpha G))$$
$$V_3(y,z) = kT_{\text{input}}(2FG(\alpha G - \rho CE + \gamma BCFkz) + 2\rho CEFk(1 + \gamma Ay) - \alpha CT_{\text{input}}(2\gamma AFkz + \rho Ez - 2\rho E + 2\alpha G))$$
$$V_4(y,z) = k(C(-G(\rho E(\alpha T_{\text{input}}(1 - z) + kF(1 + \gamma Ay)) - 4kz\alpha\gamma AFT_{\text{input}}) - kCT_{\text{input}}(\alpha\rho E(z - 1)(1 + \gamma Ay) + z\gamma A(\rho EF + \alpha^2 T_{\text{input}}))) + G(-kz\gamma BF^2G + \alpha^2 GT_{\text{input}} - \rho z\hat{\alpha}EFT_{\text{input}}))$$
$$V_5(y,z) = \rho Gk(\gamma BFk(\hat{\delta}\rho E - 2\alpha G) + \rho E(k\alpha C(1 + \gamma Ay) - k\hat{\alpha}F - \alpha\hat{\alpha}T_{\text{input}}))z$$
$$V_6(y,z) = \alpha Gk^2(-\alpha\gamma AG + E\rho(\gamma\hat{\delta}A - \hat{\alpha}))z,$$

where $A = a/(1 + a), B = \beta\delta - \alpha d, C(y) = \hat{\alpha}y + \hat{\delta}, D(y) = \beta y + d, E = 1 - \epsilon, F(y) = \alpha y + \delta, G = \epsilon\rho S$ and $H(y) = \alpha C(y)T_{\text{input}} - F(y)G$.

Theorem 4. *When the system is at equilibrium, a threshold of the antigenic stimulation b_M of T cells exists, if and only if, there is a zero $x_M \in [T_{\text{hom}}, T_{\text{cap}}]$ of the antigen threshold function $V(x, y(x), y'(x))$. This zero is such that $b_M = b(x_M, y(x_M))$, where $M \in \{L, H\}$. The equality $V(x, y(x), y'(x)) = 0$ is equivalent to $\tilde{V}(x) = 0$, where $\tilde{V}(x)$ is a polynomial that can be explicit constructed.*

See proof of Theorem 4 in Appendix.

For the parameters that unfold the hysteresis, the antigenic thresholds b_L and b_H form a cusp. The *cusp bifurcation* at the antigenic stimulation b_C of T cells is an origin of the unfold of the hysteresis, with respect to a parameter, and, so, biologically relevant. The concentration x_C of the T cells corresponding to levels of the antigenic stimulation $b_C = b(x_C, y(x_C))$ satisfies the following equalities

$$V(x, y(x), y'(x)) = 0 \quad \text{and}$$
$$W(x, y(x), y'(x), y''(x)) = 0, \tag{7}$$

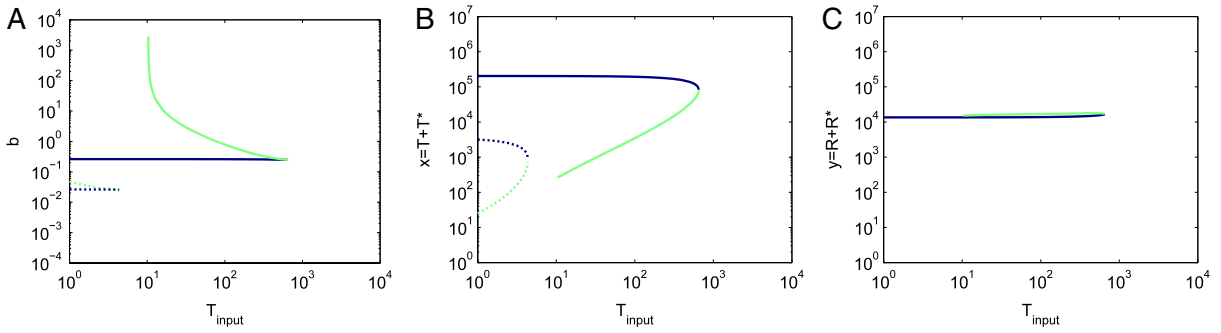

Fig. 4. Dependence of the thresholds with the Thymic input parameter $T_{\text{input}} \in [1, 650]$ with the other parameters at their default values. The model with Tregs is with bold lines and the simplified model without Tregs is with dotted lines. A: The thresholds of the antigenic stimulation b_L (dark blue) and b_H (light green). B: The concentration $x(b_L)$ (dark blue) of T cells and the concentration $x(b_H)$ (light green) of T cells. C: The concentration $y(b_L)$ (dark blue) of Tregs and the concentration $y(b_H)$ (light green) of Tregs. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

where

$$W(x, y, z, v) = \sum_{i=1}^{n} V_i(y, z)\frac{x^{i-1}}{i} + \sum_{i=0}^{n} \frac{\partial V_i(y, z)}{\partial y} x^i z + \frac{\partial V_i(y, z)}{\partial z} x^i v.$$

In Fig. 4, the antigenic thresholds b_H and b_L and the ratio b_H/b_L decrease with T_{input}. The cusp b_C occurs at $T_{\text{input}} \approx 650$ cells per ml, unfolding the hysteresis. When T_{input} gets close to the value 10.34 cells per ml the threshold b_H tends to infinity. The concentration $x(b_L)$ of T cells decreases and the concentration $x(b_H)$ of T cells increases with T_{input}. The concentration $y(b_L)$ of Tregs increases with T_{input} and the concentration $y(b_H)$ of Tregs has a maxima for $T_{\text{input}} \approx 500$ cells per ml.

6. Discussion

In this paper, we examined a mechanism proposed in [2] of Treg control of immune responses through regulation of cytokine dependent T cell proliferation. In Theorem 1, we determine the analytic formula that describes the fine balance between Regulatory T cells and T cells at controlled and immune response equilibrium states. In Theorem 4, we determine the explicit formula of these threshold mechanism.

If antigenic stimulation rises above the threshold b_H, control is lost and autoimmunity arises. Note that even if the antigenic stimulation level b falls to the original value, at which control was originally achieved, control may not be reacquired. Control is only attained if stimulation falls below the second threshold b_L. This phenomena, termed hysteresis, is common in many physical and biological systems. In Theorems 2 and 3, we exhibit the unfolding of the equilibria manifold using the explicit formula for the antigenic stimulation $b(x, y(x))$ presented here.

In [3], we observed that the hysteresis is unfold for low values of the secretion rate of cytokine J (parameter S), for low values of the growth rate ratio between Tregs and T cells (parameter ϵ), for low growth rates (parameter ρ), and for high thymic inputs (parameter T_{input}). These results are proven using Theorem 4 and Eq. (7) of this paper. As we change the parameters towards the point where a cusp bifurcation occurs and the hysteresis is unfold, we observe that the thresholds b_L and b_H decrease. Hence, there is a drastic change in the dynamical behavior, resulting in an immune response occurring at lower antigenic stimulation of T cells. Therefore, the likelihood of autoimmunity is increased.

We observe two distinct causes for the unfold of the hysteresis: low homeostatic concentrations of Tregs or high homeostatic concentration of T cells. A consequence of low values of the secretion rate of cytokine J (parameter S) or low values of the growth rate ratio between Tregs and T cells (parameter ϵ) is the presence of Tregs in low numbers (or even absent). This leads to different characteristics of the equilibria manifold (see Fig. 3D). For low growth rates (parameter ρ) we observe that the Tregs are more affected since they proliferate at a lower rate than T cells. Hence, the T cells dominate the dynamic behavior and, for the values of the parameters we are using, the hysteresis is unfold. High values of the thymic input (parameter T_{input}) result in higher homeostatic values of the concentration of T cells, blurring the distinction between controlled state and immune response state.

The presence of a cusp bifurcation allows another way of switching between a controlled state and an immune response state. If we consider that the parameter values slowly change in time (for example during puberty), a parameter may achieve a value that unfolds the hysteresis and be changed back to a value near its initial value. As a result, we would observe a controlled state being driven to (or from) an (auto)immune state.

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Appendix

Proof of Theorem 1. At equilibrium we have that:

$$\hat{\sigma}(S - (\hat{\alpha}y + \hat{\delta})J) = 0, \tag{8}$$

$$\sigma(T^* - (\alpha(x+y) + \delta)I) = 0 \tag{9}$$

$$(\epsilon\rho(I+J) - \beta(x+y) - d)R + \hat{k}(R^* - aR) = 0, \tag{10}$$

$$(\epsilon\rho(I+J) - \beta(x+y) - d)R^* - \hat{k}(R^* - aR) = 0, \tag{11}$$

$$(\rho I - \beta(x+y) - d)T + k(T^* - bT + \gamma R^* T^*) + T_{\text{input}} = 0 \tag{12}$$

$$(\rho I - \beta(x+y) - d)T^* - k(T^* - bT + \gamma R^* T^*) = 0. \tag{13}$$

From (8), we get

$$J = \frac{S}{\hat{\alpha}y + \hat{\delta}}. \tag{14}$$

From (9), we have

$$T^* = I(\alpha(x+y) + \delta). \tag{15}$$

Adding (10) and (11), we obtain

$$\epsilon\rho(I+J) - \beta(x+y) - d = 0 \tag{16}$$

(or $y = 0$).

Subtracting (11) from (10), we get

$$(\epsilon\rho(I+J) - \beta(x+y) - d)(R - R^*) + 2\hat{k}(R^* - aR) = 0. \tag{17}$$

From (16) and (17), we have

$$R^* = aR. \tag{18}$$

Let $A = a/(a+1)$. From (18), we get

$$R^* = Ay. \tag{19}$$

Adding (12) and (13), we obtain

$$\rho I - \beta(x+y) - d + \frac{T_{\text{input}}}{x} = 0 \tag{20}$$

(or $x = 0$).

Subtracting (20) from (16), we get

$$\epsilon\rho J - (1-\epsilon)\rho I - \frac{T_{\text{input}}}{x} = 0. \tag{21}$$

Subtracting (13) from (12), we have

$$(\rho I - \beta(x+y) - d)(T - T^*) + 2k(T^* - bT + \gamma R^* T^*) + T_{\text{input}} = 0. \tag{22}$$

From (20) and (22), we have

$$T^*(kx(1 + \gamma R^*) + T_{\text{input}}) = kxbT. \tag{23}$$

From (23), we get

$$T^* = \frac{kbx^2}{kx(1+b+\gamma R^*)+T_{\text{input}}}. \tag{24}$$

From (19) and (24), we obtain

$$T^* = \frac{kbx^2}{kx(1+b+\gamma Ay)+T_{\text{input}}}. \tag{25}$$

From (15) and (25), we get

$$I(\alpha(x+y)+\delta) = \frac{kbx^2}{kx(1+b+\gamma Ay)+T_{\text{input}}}. \tag{26}$$

Replacing (14) and (26) in (21), we have

$$\frac{\epsilon\rho S}{\hat{\alpha}y+\hat{\delta}} - \frac{(1-\epsilon)\rho kbx^2}{(\alpha(x+y)+\delta)(kx(1+b+\gamma Ay)+T_{\text{input}})} - \frac{T_{\text{input}}}{x} = 0. \tag{27}$$

Replacing (26) in (20), we obtain

$$\frac{\rho kbx^2}{(\alpha(x+y)+\delta)(kx(1+b+\gamma Ay)+T_{\text{input}})} - \beta(x+y) - d + \frac{T_{\text{input}}}{x} = 0. \tag{28}$$

From (28), we get

$$\frac{\rho kb}{(\alpha(x+y)+\delta)(kx(1+b+\gamma Ay)+T_{\text{input}})} = \beta(x+y) + d - \frac{T_{\text{input}}}{x}. \tag{29}$$

Replacing (29) in (27), we have

$$\frac{\epsilon\rho S}{\hat{\alpha}y+\hat{\delta}} - (1-\epsilon)\left(\beta(x+y)+d-\frac{T_{\text{input}}}{x}\right) - \frac{T_{\text{input}}}{x} = 0. \tag{30}$$

From (30), we obtain

$$x\epsilon\rho S - (1-\epsilon)(\beta(x+y)x + \mathrm{d}x - T_{\text{input}})(\hat{\alpha}y+\hat{\delta}) - T_{\text{input}}(\hat{\alpha}y+\hat{\delta}) = 0, \tag{31}$$

which, solving (31) for y and considering only the positive root, proves equality (2). □

Proof of Theorem 2. From (21), we have

$$I(1-\epsilon)\rho x = J\epsilon\rho x - T_{\text{input}}. \tag{32}$$

Replacing (14) in (32), we get

$$I = \frac{\epsilon\rho Sx - T_{\text{input}}(\hat{\alpha}y+\hat{\delta})}{(1-\epsilon)\rho x(\hat{\alpha}y+\hat{\delta})}. \tag{33}$$

Replacing (33) in (26), we obtain

$$(\epsilon\rho Sx - T_{\text{input}}(\hat{\alpha}y+\hat{\delta}))(\alpha(x+y)+\delta) = \frac{(1-\epsilon)\rho kbx^3(\hat{\alpha}y+\hat{\delta})}{kx(1+b+\gamma Ay)+T_{\text{input}}}. \tag{34}$$

Hence, solving (34) for b, we prove equality (5). □

Proof of Theorem 3. At equilibrium we have that:

$$\sigma(T^* - (\alpha x+\delta)I) = 0, \tag{35}$$

$$(\rho I - \beta x - d)T + k(T^* - bT) + T_{\text{input}} = 0, \tag{36}$$

$$(\rho I - \beta x - d)T^* - k(T^* - bT) = 0. \tag{37}$$

From (35), we have

$$T^* = I(\alpha x+\delta). \tag{38}$$

Adding (36) and (37), we obtain

$$\rho I - \beta x - d + \frac{T_{\text{input}}}{x} = 0 \tag{39}$$

(or $x = 0$).

Subtracting (37) from (36), we have

$$(\rho I - \beta x - d)(T - T^*) + 2k(T^* - bT) + T_{\text{input}} = 0. \tag{40}$$

From (39) and (40), we have

$$T^*(kx + T_{\text{input}}) = kxbT. \tag{41}$$

From (41), we obtain

$$T^* = \frac{kbx^2}{kx(1+b) + T_{\text{input}}}. \tag{42}$$

From (38) and (42), we get

$$I(\alpha x + \delta) = \frac{kbx^2}{kx(1+b) + T_{\text{input}}}. \tag{43}$$

From (20), we have

$$I = \frac{\beta x^2 + \mathrm{d}x - T_{\text{input}}}{\rho x}. \tag{44}$$

Finally, from (43) and (44), we get

$$\tilde{b}(x) = \frac{(\alpha x + \delta)(kx + T_{\text{input}})(\beta x^2 + \mathrm{d}x - T_{\text{input}})}{kx(\rho x^2 + (T_{\text{input}} - \beta x^2 - \mathrm{d}x)(\alpha x + \delta))}. \quad \square \tag{45}$$

Proof of Theorem 4. By equality (5), we have that $b(x, y) = N(x, y)/D(x, y)$, where N and D are the following cubic polynomials in x and y

$$N(x, y) = (\epsilon\rho Sx - T_{\text{input}}(\hat{\alpha}y + \hat{\delta}))(\alpha(x + y) + \delta)(kx(1 + \gamma Ay) + T_{\text{input}})$$
$$D(x, y) = k(1 - \epsilon)\rho x^3(\hat{\alpha}y + \hat{\delta}) - kx(\epsilon\rho Sx - T_{\text{input}}(\hat{\alpha}y + \hat{\delta}))(\alpha(x + y) + \delta).$$

If $T_{\text{input}} > 0$, the points x_L and x_H exist if and only if, the function $\mathrm{d}b(x, y(x))/\mathrm{d}x$ have two distinct positive zeros. The values x_L and x_H are these zeros. If $T_{\text{input}} = 0$, the point x_L exists if and only if, the function $\mathrm{d}b(x, y(x))/\mathrm{d}x$ has one positive zero. Furthermore, x_L is such zero. From Eq. (5), we have that $b(x, y) = N(x, y)/D(x, y)$, with $N(x, y)$ and $D(x, y)$ cubic polynomials in x and y. Hence, $\mathrm{d}b(x, y(x))/\mathrm{d}x$ is equal to $V(x, y(x), y'(x))$ where

$$V(x, y, z) = \frac{\partial N(x, y)}{\partial x}D(x, y) - N(x, y)\frac{\partial D(x, y)}{\partial x} + \left(\frac{\partial N(x, y)}{\partial y}D(x, y) - N(x, y)\frac{\partial D(x, y)}{\partial y}\right)z.$$

Since

$$\frac{\partial N(x, y(x))}{\partial x} = \epsilon\rho SVW + \alpha UV + kUW(1 + \gamma Ay)$$
$$\frac{\partial N(x, y(x))}{\partial y} = -\hat{\alpha}VWT_{\text{input}} + \alpha UV + kx\gamma AUW$$
$$\frac{\partial D(x, y(x))}{\partial x} = -kUW - kx\epsilon\rho SW - kx\alpha U + 3k\rho(1 - \epsilon)(\hat{\alpha}y + \hat{\delta})x^2$$
$$\frac{\partial D(x, y(x))}{\partial y} = kx\hat{\alpha}WT_{\text{input}} - kx\alpha U + k\hat{\alpha}\rho(1 - \epsilon)x^3$$

where $U(x, y) = \epsilon\rho Sx - T_{\text{input}}(\hat{\alpha}y + \hat{\delta})$, $V(x, y) = kx(1 + \gamma Ay) + T_{\text{input}}$ and $W(x, y) = \alpha(x + y) + \delta$, we get that the expression $V(x, y, z)$ for the antigen threshold function follows. $\square$

References

[1] C.-S. Hsieh, Y. Liang, A.J. Tyznik, S.G. Self, D. Liggitt, A.Y. Rudensky, Recognition of the peripheral self by naturally arising $CD25^{+}CD4^{+}$ *T* cell receptors, Immunity 21 (2004) 267–277.
[2] N.J. Burroughs, B.M.P.M. Oliveira, A.A. Pinto, Regulatory *T* cell adjustment of quorum growth thresholds and the control of local immune responses, J. Theoret. Biol. 241 (2006) 134–141.
[3] N.J. Burroughs, B.M.P.M. Oliveira, A.A. Pinto, H.J.T. Sequeira, Sensibility of the quorum growth thresholds controlling local immune responses, Math. Comput. Modelling 47 (2008) 714–725.
[4] A.M. Thornton, E.M. Shevach, $CD4^{+}CD25^{+}$ immunoregulatory *T* cells suppress polyclonal *T* cell activation in vitro by inhibiting interleukine 2 production, J. Exp. Med. 188 (1998) 287–296.
[5] K.S. Schluns, W.C. Kieper, S.C. Jameson, L. Lefrancois, Interleukin-7 mediates the homeostasis of naive and memory CD8 *T* cells in vivo, Nat. Immunol. 1 (2000) 426–432.
[6] C.A. Michie, A. McLean, C. Alcock, P.C.L. Beverley, Life-span of human lymphocyte subsets defined by CD45 isoforms, Nature 360 (1992) 264–265.
[7] D. Moskophidis, M. Battegay, M. Vandenbroek, E. laine, U. Hoffmannrohrer, R.M. Zinkernagel, Role of virus and host variables in virus persistence or immunopathological disease caused by a noncytolytic virus, J. Gen. Virol. 76 (1995) 381.
[8] H. Veiga-Fernandes, U. Walter, C. Bourgeois, A. McLean, B. Rocha, Perturbation theory analysis of competition in a heterogeneous population, Nat. Immunol. 1 (2000) 47–53.
[9] P.M. Anderson, M.A. Sorenson, Effects of route and formulation on clinical pharmacokinetics of interleukine-2, Clin. Pharmacokinet. 27 (1994) 19–31.
[10] John W. Lowenthal, Warner C. Greene, Contrasting interleukine 2 binding properties of the alpha (p55) and beta (p70) protein subunits of the human high-affinity interleukine 2 receptor, J. Exp. Med. (1987) 1155–1169.
[11] R.J. de Boer, P. Hogeweg, Immunological discrimination between self and non-self by precursor depletion and memory accumulation, J. Theoret. Biol. 124 (1987) 343.
[12] A.R. McLean, Modelling *T* cell memory, J. Theoret. Biol. (1994) 63–74.
[13] C. Utzny, N.J. Burroughs, Perturbation theory analysis of competition in a heterogeneous population, Physica D 175 (2003) 109–126.
[14] Robin E. Callard, Jaroslav Stark, Andrew J. Yates, Fratricide: a mechanism for *T* memory-cell homeostasis, Trends Immunol. 24 (2003) 370–375.
[15] K. Leon, R. Perez, A. Lage, J. Carneiro, Modelling T-cell-mediated suppression dependent on interactions in multicellular conjugates, J. Theoret. Biol. 207 (2000) 231–254.
[16] K. Leon, A. Lage, J. Carneiro, Tolerance and immunity in a mathematical model of T-cell mediated suppression, J. Theoret. Biol. 225 (2003) 107–126.