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A stochastic microscopic model for the dynamics of antigenic variation

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HIGHLIGHTS

- We introduce a branching process to model the dynamics of antigenic variation.
- We completely characterize the different phases in the space of parameters in a rather general setting.
- Parameters as random variables allow to capture relevant features observed in nature.
- The model is simple and allows generalizations to more complicated situations.
- The interplay between immune evasion and immune response alone does not lead to persistent oscillatory behavior (parasitemia waves).

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ABSTRACT

We present a novel model that describes the within-host evolutionary dynamics of parasites undergoing antigenic variation. The approach uses a multi-type branching process with two types of entities defined according to their relationship with the immune system: clans of resistant parasitic cells (i.e. groups of cells sharing the same antigen not yet recognized by the immune system) that may become sensitive, and individual sensitive cells that can acquire a new resistance thus giving rise to the emergence of a new clan. The simplicity of the model allows analytical treatment to determine the subcritical and supercritical regimes in the space of parameters. By incorporating a density-dependent mechanism the model is able to capture additional relevant features observed in experimental data, such as the characteristic parasitemia waves. In summary our approach provides a new general framework to address the dynamics of antigenic variation which can be easily adapted to cope with broader and more complex situations.

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1. Introduction

Parasites have evolved a diversity of sophisticated strategies to evade the host's immune response, among which antigenic variation is perhaps one of the most striking ones. This strategy consists of periodically changing a protective coat composed of an abundant and immunogenic protein. In this mechanism the parasites express only one variant antigenic protein copy from a large repertoire of silent genes. The mechanism allows transient immune evasion, since after changing the variable protein that is being expressed, an entirely new parasite population arises that is not recognized by the host's immune system, which has only developed an antibody response directed against the previous antigen. By repeating this cycle during the course of an infection, parasites are able to remain in the host for long periods of time.

Perhaps the most paradigmatic example is that of African trypanosomes (responsible for producing sleeping sickness in humans), but antigenic variation is also observed in *Giardia llambia* and the malaria agents belonging to the *Plasmodium* genus. Some viruses are also able to evade the immune response in a strategy similar to that just described. However in the latter systems, antigenic diversity is generated by the introduction of point mutations in the gene encoding the antigen, rather than by switching the expressed gene. This implies some substantial differences in the dynamics since the new antigen is most likely somewhat similar to the previous one (parental) and perhaps is recognized (yet with lower affinity) by the same antibodies.

Several models have been developed to study and predict the population dynamics of parasites and viruses during the course of an infection within a single host. Most of them are based on a system of coupled differential equations inspired on variations of predator-prey models (see Kosinski, 1980; Barry and Turner, 1991; Agur et al., 1989; Agur, 1992; Antia et al., 1996; Frank, 1999; Nowak and May, 2000.). In short, this approach consists of a set of differential equations describing the dynamical interaction between antigens and the host's

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immune system, in such a way that the outcome of one equation is a modulating parameter of the others, and including in some cases cross-reactive immune responses as well as other possible interactions (Antia et al., 1996). Stochasticity is incorporated ad hoc into the models by the emergence of new variants (which are not recognized by immune system) at random times, usually driven by a Poisson process (see Nowak and May, 2000 and references therein).

Very recently Gurarie et al. (2012) implemented a discrete time computer model for the case of malaria. This modeling approach, termed agent-based, consists of a set of coupled difference equations that describe the transition between successive iterations of the parasite population (i.e. parasite generations) and its interaction with the immune system. According to the authors the advantage of this approach is that, owing to its discrete nature, stochastic components are incorporated more easily by adding random factors to the variables that represent the efficiency of immune system.

In spite of the existence of these models of antigenic variation, in our opinion it is worth re-addressing the problem from a different perspective. Here we present a model that tackles this topic from a microscopic point of view that consists of following the pathway and behavior of its individual elements through a multi-type branching process. Iwasa et al. (2004) already used this methodology to study problems related to the ones presented here; however these authors focused on the evolutionary dynamics of viruses to escape antiviral therapy.

The model presented here has the following advantages: the role of each one of its parameters has a straightforward biological interpretation; its versatility easily permits the incorporation of increasing complexity and realism; the process can be studied backwards in time, as in population genetics' coalescent theory. Finally, its simplicity allowed us to obtain analytical expressions for the critical surface separating subcritical from supercritical regimes in the parameter space, both in the simplest version of the model as well as for its generalizations.

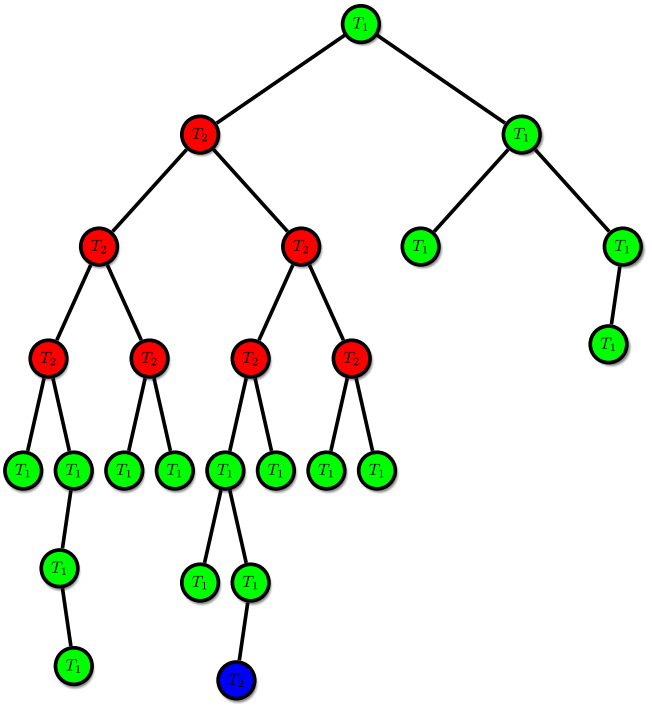

Fig. 1. Green cells are sensitive. Red and blue cells represent clans of antigen variants not recognized by the immune system. (For interpretation of the references to color in this figure caption, the reader is referred to the web version of this paper.)

2. The model

Our model, a discrete-time non-independent multi-type branching process, assumes the existence of two types of *cells/infective particles* (viral particles, parasites, etc.) which are defined according to the host's immune system ability to recognize them; namely, sensitive (type-1) and resistant (type-2) cells.

The model in its simplest version involves three parameters, $\delta, \mu, p \in [0,1]$, that are defined as follows:

The population of cells proliferates by binary division and the offspring of sensitive cells die, independently, with probability δ (and consequently survive with probability $1-\delta$). Surviving cells may become resistant (i.e. start producing a new antigen variant) with probability μ.

A newly arisen resistant cell creates a clan (or lineage) of resistant cells in the following, recursive, way: at a given time the whole progeny of resistant cells divides into resistant cells, which remain as such with probability equal to p. In other words, $1-p$ is the probability that the immune system acquires the ability to recognize this particular clan, i.e. the clan bearing this specific variant antigen.

This means that for a given resistant cell appearing at generation n we take a geometric random variable, N, of parameter $1-p$, and consider the whole dividing resistant clan until generation $n+N$, where resistant cells become sensitive.

Summarizing: parameter δ measures the efficiency of immune response against sensitive cells; parameter μ represents the rate at which new resistant variants appear; and parameter p is related to the delay times spent by the immune systems to recognize a new variant.

Fig. 1 illustrates a realization of the process: a sensitive (green) cell originates a resistant descendant clan (red) which in turn becomes sensitive (green) after three generations. At the bottom of the figure, the emergence of a new resistant variant (blue) is represented. Different clans of resistant cells, and sensitive cells, evolve independently.

We remark that this is not a standard multi-type branching process as for example those considered in Kimmel and Axelrod (2002), in the sense that resistant cells in a given clan do not evolve independently: instead, their destiny is determined by immune system capacity, which does or does not recognize the whole population of cells carrying a specific variant antigen. The model could also be envisaged as a percolation process on the complete binary tree in presence of a random environment (the clans of resistant cells of random sizes).

3. Extinction probability

To compute the extinction/survival probabilities of the process – and thus obtain the critical surface as a function of the parameters – we introduce an additional multi-type branching process which has the same extinction/survival probabilities as the antigenic variation model introduced before. This independent multi-type branching process with two types of cells is obtained from the antigenic variation model by *collapsing* to one generation each clan of resistant cells.

3.1. Independent multi-type branching process

Let us consider two types of cells that evolve independently. The progeny of each cell is as follows: type-1 (sensitive) cells give birth to

- two type-1 cells with probability $(1-\mu)^2(1-\delta)^2$,
- two type-2 cells with probability $\mu^2(1-\delta)^2$,

- one type-1 cell and one type-2 cell with probability $2\mu(1-\mu)(1-\delta)^2$,
- one type-1 cell with probability $2(1-\mu)\delta(1-\delta)$,
- one type-2 cell with probability $2\mu\delta(1-\delta)$,
- no cell with probability δ^2;

type-2 (resistant) cells give birth to

- 2^N type-1 cells with probability $p^{N-1}(1-p)$, $N=1,2,3,\ldots$

3.2. Extinction probability

We get the equation for the extinction probability of this independent multi-type branching process – starting with one type-1 cell, one type-2 cell, or eventually any given initial configuration of cells – following standard procedures that use probability generating functions (pgf) as given, for example, in Kimmel and Axelrod (2002), Jagers (1975), Haccou et al. (2005).

For each $n=0,1,2,\ldots$ we denote $\overline{Z}_1(n)$ (resp. $\overline{Z}_2(n)$) the number of type-1 (resp. type-2) cells present at generation n in the collapsed model. In the particular case $n=1$ we put $\overline{Z}_1=\overline{Z}_1(1)$ (resp. $\overline{Z}_2=\overline{Z}_2(1)$) to simplify the notation.

As it is well known, the distribution of $\overline{Z}_1(n)$ and $\overline{Z}_2(n)$ as well as the probability of extinction of the process could be computed from the pgf's of $\overline{Z}_1$ and $\overline{Z}_2$, which are

$$f_1(s,t)=\mathbb{E}\left[s^{\overline{Z}_1}\,t^{\overline{Z}_2}\,|\,\overline{Z}_1(0)=1,\ \overline{Z}_2(0)=0\right]$$
$$f_2(s,t)=\mathbb{E}\left[s^{\overline{Z}_1}\,t^{\overline{Z}_2}\,|\,\overline{Z}_1(0)=0,\ \overline{Z}_2(0)=1\right],$$
$$s,t\in[0,1]$$

We have

$$f_1(s,t)=\{\delta+(1-\delta)[\mu t+(1-\mu)s]\}^2 f_2(s,t)=(1-p)\sum_{k=1}^{\infty}s^{2^k}p^{k-1}. \tag{1}$$

Let q_1 (resp. q_2) denote the extinction probability for the process starting with a single type-1 (resp. type-2) cell. The following result is adapted from Kimmel and Axelrod (2002), Jagers (1975); Haccou et al. (2005):

Proposition 3.1. *The probability of extinction of the process, $q=(q_1,q_2)$, is the solution of* equation

$$(f_1(s,t),f_2(s,t))=(s,t) \tag{2}$$

that is closest to the origin in $[0,1]^2$.

Note that $f_1(1,1)=f_2(1,1)=1$, so that $(1,1)$ is a solution of (2). Depending on the values of parameters δ, μ and p, it can happen: (i) $(1,1)$ is the only solution of (2) and hence the extinction probability is one; (ii) there exists another solution of (2) in $(0,1)^2$ and the non-extinction probability is positive.

3.3. Critical surface

In order to know which is the set of parameter values that makes the multi-type branching process (and hence the antigenic variation model) becomes extinct with probability one, or survives forever with a positive probability, we look for the values $(s_0,t_0)\in[0,1]^2$ that satisfy

$$\{\delta+(1-\delta)[\mu t_0+(1-\mu)s_0]\}^2=s_0$$
$$(1-p)\sum_{k=1}^{\infty}s_0^{2^k}p^{k-1}=t_0. \tag{3}$$

Note that $s_0\neq 1$ (resp. $s_0\neq 0$) if and only if $t_0\neq 1$ (resp. $t_0\neq 0$). From these equations we get that the probability of non-extinction is positive if and only if there exists $s_0\in(0,1)$ such that

$$\left(\delta+(1-\delta)\left[\mu(1-p)\sum_{k=1}^{\infty}s_0^{2^k}p^{k-1}+(1-\mu)s_0\right]\right)^2=s_0.$$

Let us introduce the function

$$g(s)=\left(\delta+(1-\delta)\left[\mu(1-p)\sum_{k=1}^{\infty}s^{2^k}p^{k-1}+(1-\mu)s\right]\right)^2,$$

$s\in[0,1]$. Note that, for $\delta,\mu,p\in(0,1)$, $g(.)$ is a strictly increasing and convex function with $g(0)=\delta^2$ and $g(1)=1$. Therefore, there exists $s_0\in(0,1)$ satisfying $g(s_0)=s_0$ if and only if $g'(1)>1$. It holds

$$g'(1)=2(1-\delta)\left[2\mu(1-p)\sum_{k=0}^{\infty}(2p)^k+(1-\mu)\right].$$

Case 1: $p\geq 1/2$. $g'(1)=\infty$, and so there exists $s_0\in(0,1)$ such that $g(s_0)=s_0$. The probability of non-extinction is positive; we say the process lies in the *supercritical region.*

Case 2: $p<1/2$. We have

$$g'(1)=\frac{2(1-\delta)}{1-2p}[1+\mu-2p].$$

Critical μ: For each pair (δ,p) we introduce the *critical value*, $\mu_c(\delta,p)$, defined by

$$\frac{2(1-\delta)}{1-2p}[1+\mu_c(\delta,p)-2p]=1,$$

$0\leq\mu_c(\delta,p)\leq 1$.

It is clear that the probability of extinction is one – or strictly smaller than one – depending on $\mu\leq\mu_c(\delta,p)$ – or $\mu>\mu_c(\delta,p)$.

We summarize the analysis in the following result:

Proposition 3.2. *Let*

$$\mu_c(\delta,p)=\begin{cases}\min\left\{1,\ \max\left\{0,\frac{(1-2p)}{2(1-\delta)}(2\delta-1)\right\}\right\} & \text{if } p<\frac{1}{2}\\ 0 \quad \text{if } p\geq\frac{1}{2}.\end{cases}$$

It holds, for each (δ,p,μ):

(i) *The process is subcritical* (*i.e. the extinction probability is one*) *if* $\mu\leq\mu_c(\delta,p)$.
(ii) *The process is supercritical* (*i.e. the probability of non-extinction is strictly positive*) *if* $\mu>\mu_c(\delta,p)$.

Remark 1. The critical surface does not depend on the initial condition of the process, since it starts with a finite number of type-1 and type-2 cells. On the other hand, what it does depend on the initial condition is the extinction probability in the supercritical region.

Fig. 2 shows the critical surface in the non-trivial region: $\delta>1/2$, $p<1/2$.

Numerically solving Eq. (3) in the supercritical region we obtain the extinction probability $q_1=s_0$ (resp. $q_2=t_0$) for the process starting with one type-1 cell (resp. one type-2 cell). In Fig. 3 we show these extinction probabilities computed for $p=0.65$.

4. Generalization of the model

Although in previous sections we introduced the model in a simple version, it can be generalized to include more realistic situations, allowing natural death of resistant cells (an effect driven by a set of parameters $\boldsymbol{\Delta}$) and more efficient mechanisms of immune response to recognize resistant clans (driven by a set of parameters $\mathbf{p}$).

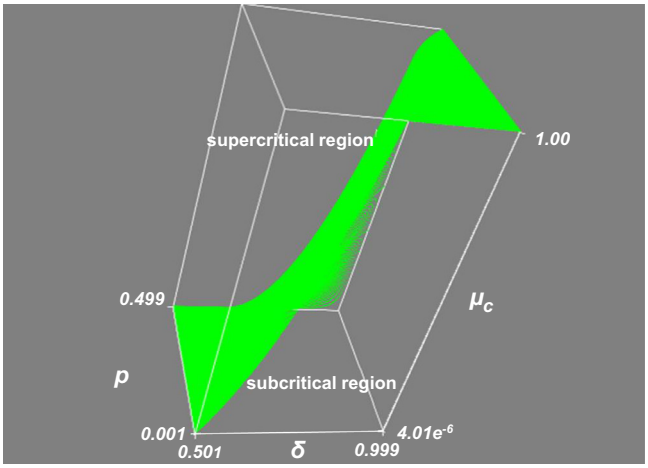

Fig. 2. Critical surface: μ_c as a function of parameters p and δ. μ_c stands for the critical μ as given in Proposition 3.2.

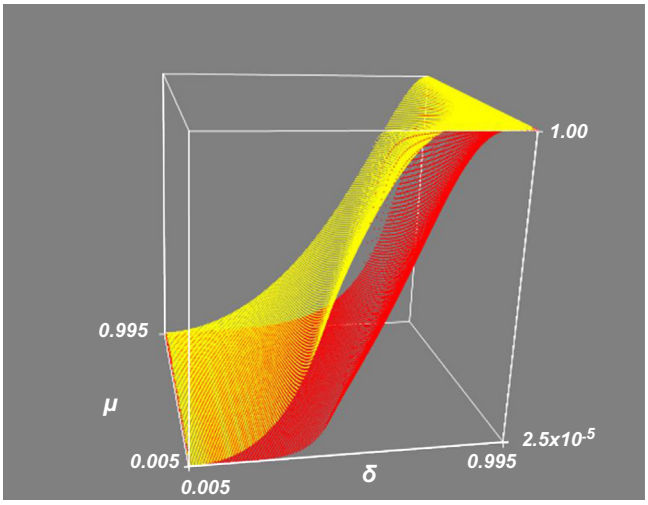

Fig. 3. Extinction probabilities, as functions of μ and δ, for the process starting with one type-1 cell (yellow) and one type-2 cell (red). The parameter p is fixed and equal to 0.65. (For interpretation of the references to color in this figure caption, the reader is referred to the web version of this paper.)

Let us denote $\Theta(\mathbf{\Delta},\mathbf{p})$ the expectation of $\mathcal{N}$, the number of cells that comprise a given resistant clan right in the moment when it becomes sensitive (i.e. recognized by immune system), and $\mathbb{P}_{\mathbf{\Delta},\mathbf{p}}$ the probability distribution of variable $\mathcal{N}$. The case of interest is $\Theta(\mathbf{\Delta},\mathbf{p})>1$. Progenies of type-1 (sensitive) cells are as in Sections 2 and 3. As before, to compute extinction/survival probabilities, each resistant clan can be collapsed to one generation. The pgf's appearing in (1) must be replaced by

$$f_1(s,t)=\{\delta+(1-\delta)[\mu t+(1-\mu)s]\}^2$$

$$f_2(s,t)=\sum_{k=0}^{\infty}s^k\mathbb{P}_{\Delta,\mathbf{p}}(\mathcal{N}=k).$$

The same calculations of Section 3 allow us to compute extinction/survival probabilities. The process becomes extinct with probability one if and only if

$$2(1-\delta)[\mu\Theta(\mathbf{\Delta},\mathbf{p})+(1-\mu)]\leq 1.$$

We get the following general result:

Proposition 4.1. *Let* $\mu_c(\delta,\mathbf{\Delta},\mathbf{p})=0$, *if* $\Theta(\mathbf{\Delta},\mathbf{p})=\infty$ *and*

$$\mu_c(\delta,\mathbf{\Delta},\mathbf{p})=\min\left\{1,\ \max\left\{0,\frac{(2\delta-1)}{2(1-\delta)[\Theta(\mathbf{\Delta},\mathbf{p})-1]}\right\}\right\},$$

if $\Theta(\mathbf{\Delta},\mathbf{p})<\infty$. *It holds, for each* $(\delta,\mu,\mathbf{\Delta},\mathbf{p})$:

i) *The process is subcritical* (*i.e. the extinction probability is one*) *if* $\mu\leq\mu_c(\delta,\mathbf{\Delta},\mathbf{p})$.
ii) *The process is supercritical* (*i.e. the probability of non-extinction is strictly positive*) *if* $\mu>\mu_c(\delta,\mathbf{\Delta},\mathbf{p})$.

In the next subsections we consider two particular cases of interest. Progenies of type-1 cells are as in Section 2.

4.1. Death of resistant cells

Let us assume that, at each generation, type-2 cells of a given clan remain resistant with probability p or become sensitive with probability $(1-p)$. Each resistant cell inside the clan proliferates by binary division and the offspring die, independently, with probability Δ (the case $\Delta<1/2$ gives exponential growth of the mean size of the resistant clan; note that the model introduced in Section 2 corresponds to $\Delta=0$).

Let us denote M the random variable that accounts for the number of generations until a given resistant clan, originated by a type-2 cell, becomes sensitive. Clearly, M is a geometrical random variable of parameter $1-p$. We have

$$\Theta(\Delta,p)=\sum_{m=1}^{\infty}\mathbb{P}(M=m)\ \mathbb{E}(\mathcal{N}|M=m).$$

Note that, until generation M, the clan of descendants of a given type-2 cell behaves as an usual Galton–Watson process in which the mean number of descendants of each cell is $2(1-\Delta)$. Then, as it is well known from the theory of independent branching processes:

$$\begin{aligned}\Theta(\Delta,p)&=\sum_{m=1}^{\infty}\mathbb{P}(M=m)\,[2(1-\Delta)]^m\\&=\sum_{m=1}^{\infty}(1-p)p^{m-1}\,[2(1-\Delta)]^m.\end{aligned}\tag{4}$$

Then

$$\Theta(\Delta,p)=\begin{cases}\frac{2(1-\Delta)(1-p)}{1-2p(1-\Delta)}, & \text{if } 2p(1-\Delta)<1\\ \infty, & \text{if } 2p(1-\Delta)\geq 1.\end{cases}$$

Finally, the critical surface in the space of parameters (δ,μ,Δ,p) separating subcritical from supercritical regimes is given by

$$\mu_c(\delta,\Delta,p)=\min\left\{1,\max\left\{0,\frac{(2\delta-1)[1-2p(1-\Delta)]}{2(1-\delta)(1-2\Delta)}\right\}\right\}$$

if $2p(1-\Delta)<1$; $\mu_c(\delta,\Delta,p)=0$ if $2p(1-\Delta)\geq 1$.

4.2. Growth of immune response probability

Here we introduce a mechanism in which probability of immune response grows with the size of resistant clans. This extension does not affect the intensity of immune response (which is regulated by δ) but the waiting time until response appears. In a general Markovian setting, the number of generations M until the immune system recognizes a new resistant clan is a random variable with $\mathbb{P}(M=1)=(1-p_1)$ and

$$\mathbb{P}(M=m)=p_1p_2\ldots p_{m-1}(1-p_m)\quad m=2,3,\ldots$$

where p_i is the probability that immune system does not recognize the resistant clan at generation i (given that the clan was not

previously recognized). In the (homogeneous) case analyzed in Sections 2 and 3 $p_i = p$ for all $i = 1, 2, \ldots$. In this setting, growing of immune response can be explicitly modeled by choosing a non increasing sequence: i.e. $p_{i+1} \leq p_i$ for all $i = 1, 2, \ldots$. Closed expressions for the critical surface in the space of parameters are difficult to obtain except in particular cases.

Example 1. Uniformly bounded size of resistant clans. Suppose that there are not natural death of type-2 cells. For fixed $N_o > 1$ and $p \in (0, 1/2)$ consider

$$p_i = \begin{cases} p & \text{if } i \leq N_o \\ 0 & \text{if } i > N_o. \end{cases}$$

That is to say, each clan of resistant cells is recognized, with probability one, before generation $N_o + 1$. After some algebra we get

$$\Theta(p, N_o) = \mathbb{E}(2^M) = \frac{2(1-p) - (2p)^{N_o+1}}{(1-2p)},$$

which gives the critical surface

$$\mu_c(\delta, p, N_o) = \min\left\{1,\ \max\left\{0, \frac{(2\delta - 1)(1-2p)}{2(1-\delta)[1-(2p)^{N_o+1}]}\right\}\right\}.$$

It is clear that $\mu_c(\delta, p, N_o) \geq \mu_c(\delta, p)$, the right hand side of inequality being the critical surface of the homogeneous case. Therefore, the effect of this modification is an increase of the subcritical region of case studied in Sections 2 and 3.

Example 2. Exponential growth of immune response probability. Suppose that there are not natural death of type-2 cells. Fix $p, \alpha \in (0, 1)$. Put $p_1 = p$ and $p_{i+1} = \alpha p_i$, for $i = 1, 2, \ldots$. That situation corresponds to an exponential decay of the probability that immune system does not recognize a given clan, as a function of number of generations. We have

$$\mathbb{P}(M = m) = p^{m-1}\alpha^{(m-2)(m-1)/2}(1 - p\alpha^{m-1}), \tag{5}$$

$m = 1, 2, 3, \ldots$, and after some rearrangements

$$\Theta(p, \alpha) = \mathbb{E}(2^M) = 1 + \sum_{m=0}^{\infty} \left(\frac{2p}{\sqrt{\alpha}}\right)^m (\sqrt{\alpha})^{m^2},$$

which is finite for all values of parameters p and α. From this expression we get the critical surface $\mu_c(\delta, p, \alpha)$. One remarkable fact is that the process can be in the subcritical regime even for values of p arbitrarily close to 1. Note that the size of a resistant clan at generation n is $V_n = 2^n$ and that the probability that immune system does not recognize the resistant clan at generation n is $p_n = \alpha^{n-1}p$. We get

$$p_n = p\left(\frac{V_n}{2}\right)^{-\gamma},$$

where $\gamma = \log(1/\alpha)/\log 2 > 0$. This situation represents a size-dependent increasing probability of the immune system to recognize a given resistant clan.

Example 3. Death of resistant cells and exponential growth of immune response probability. One can add to the previous example the effect of death of resistant cells through a parameter Δ as in Section 4.1. All we have to do is place expression (5) for $\mathbb{P}(M = m)$ in (4). We get for the expectation of the number of cells that comprise a given resistant clan when it becomes sensitive:

$$\Theta(\Delta, p, \alpha) = 1 + (1 - 2\Delta)\sum_{m=0}^{\infty} \left(\frac{2p(1-\Delta)}{\sqrt{\alpha}}\right)^m (\sqrt{\alpha})^{m^2}.$$

Remark 2. The generalizations of the model presented above lead to changes in the structure of critical, subcritical and supercritical regions in the space of parameters. As it happens also in the simpler version of the model described in Section 2, the branching process shows instability in the asymptotic limit, meaning that the process explodes exponentially fast (see for example Kimmel and Axelrod, 2002) or it becomes extinct supposedly at an exponential or sub-exponential rate – as suggested by the bounds obtained for percolation and spatial birth-and-death processes in random environment at subcritical regime (see Fernández et al., 2005 and references therein). All states are transient, excepting when the population becomes extinct, and consequently any kind of stable *oscillatory regime* are extremely unlikely. Persistent, or at least prolonged, regime of parasitemia waves are frequently observed in antigenic variation, which often shows a tendency to chronicity and the infections may last for months or years (see Ross and Thomson, 1910; Barry et al., 2012 and MacGregor and Matthews, 2008). Computer simulations (presented in Supp. file 1) show that even a transient oscillatory regime (of a few hundreds generations) is a very rare phenomenon in this context. It follows that it is necessary to explore other aspects in the models presented above in order to determine the conditions that favor population oscillation. This is the subject of the next section.

5. Self-control and parasitemia waves

In this section we show that by introducing some modifications in the model, the oscillatory behavior arises with high probability for a reasonable number of generations (say, n of the order of hundreds or thousands). For concreteness we focus on the simplest version of the model introduced in Section 2 with the addition of death of resistant cells as in Section 4.1.

5.1. Random parameters

Several parasites can control their population density by mechanisms similar to quorum sensing. Such mechanisms favor sustaining a long term infection, thus increasing the chances of transmission. In the case of African trypanosomes it has been proposed that the self-regulation mechanism that prevents fast population explosions is accomplished by inducing cell transformation from dividing to non-dividing forms in a density-dependent manner (Seed and Black, 1997; Vassella et al., 1997; Tyler et al., 2001; Savill and Seed, 2004). This density-dependent cell-cycle arrest (which is very similar to quorum sensing in bacteria) is well characterized from the biochemical and genetic point of view in *Trypanosoma brucei* (Mony et al., 2014). Other parasites that undergo antigenic variation and parasitemia waves, such as *Plasmodium falciparum*, also appear to exhibit a similar mechanism of population growth control (Mutai and Waitumbi, 2010; Pollitt et al., 2010). This population behavior has been modeled before by Savill and Seed (see Savill and Seed, 2004) using a set of deterministic differential equations. These authors built their model to fit experimental results obtained using immunosuppressed mice, where the parasite population reached an upper limit in spite of not having any external control. In the next subsection and in the Appendix we show that the mechanism observed in mice can be also modeled by a logistic-type Galton–Watson process.

This prompted us to incorporate self-regulation mechanisms. For this purpose we consider parameters Δ and δ as random variables (the distribution of which depends on the state of the process – i.e. the sizes of sensitive and resistant cell populations – at every n). These parameters are updated during the evolution of the (microscopic) process. Furthermore, the parameter μ that governs switching rates is also allowed to vary in a population density manner in such a way that, when the population is small the switching rate is as high as that reported experimentally (which makes the extinction less probable), whereas the hyper-use of silent repertoire

which might lead to immune exhaustion during the peaks of parasitemia is prevented.

5.2. Varying Δ and δ

Here we consider parameters regulating the death of resistant and sensitive cells, respectively Δ and δ, as random variables updated with n.

Updating Δ: Fix a number $r_\Delta > 0$ and $0 < \Delta_{min} < 1/2 < \Delta_{max} < 1$. We denote $Q(n)$ the number of cells not recognized by the immune system (that is, resistant or type-2 cells) at generation n. Let $X_\Delta(n)$ be a random variable with Beta distribution of parameters $\alpha = Q(n)/r_\Delta$, $\beta = 1$.

We put

$$\Delta(n) = \Delta_{min} + (\Delta_{max} - \Delta_{min})X_\Delta(n), \quad n = 1, 2, 3, \ldots$$

and interpret $\Delta_{\min}$ as the background probability of natural death of resistant cells; $\Delta_{sc}(n) = (\Delta_{max} - \Delta_{min})X_\Delta(n)$ takes into account the mechanism of self-control of resistant parasite population at generation n: this mechanism regulates the rate of conversion of dividing cells into non-dividing forms.

Fig. 4 shows the effect of Δ_{sc}: the simulations correspond to realizations of Galton–Watson type branching processes (in this context, cells are resistant during the entire process) where cells at generation n give birth to

- two cells with probability $(1-\Delta(n))^2$,
- one cell with probability $2\Delta(n)(1-\Delta(n))$,
- no cell with probability $\Delta(n)^2$.

$\Delta_{min} = 0.20$, $\Delta_{max} = 0.55$ and $r_\Delta = 10^3, 10^4, 10^5$. As it can be seen, as n increases the total number of cells, $Z(n)$, grows exponentially at first and then fluctuates around a value of the order of r_Δ. Realizations start with $Z(0) = 100$. In the Appendix we show that the expectation of $Z(n)$ grows as a logistic-type process with an upper bound for the carrying capacity given by the expression

$$K = \frac{(1-2\Delta_{min})}{(2\Delta_{max}-1)} r_\Delta > 0.$$

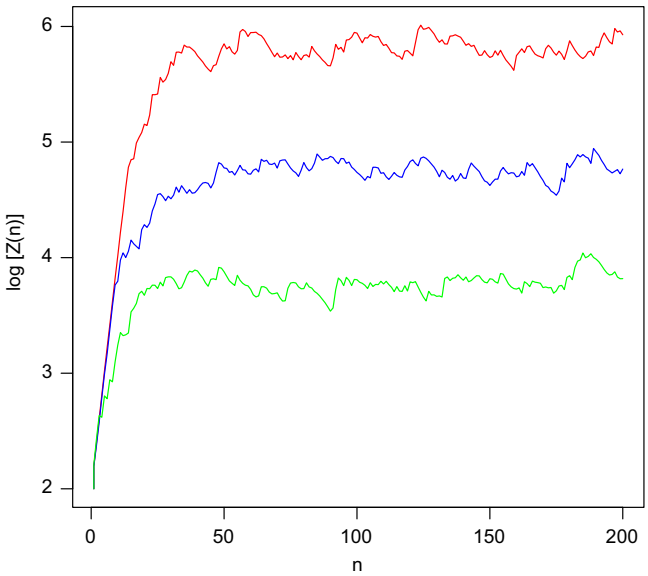

Fig. 4. Self-control: simulations of three Galton–Watson processes. $\Delta_{min} = 0.20$; $\Delta_{max} = 0.55$. $r_\Delta = 10^3$ (green); $r_\Delta = 10^4$ (blue); $r_\Delta = 10^5$ (red); $Z(0) = 100$. (For interpretation of the references to color in this figure caption, the reader is referred to the web version of this paper.)

K is an approximate value for the asymptotic limit of $\lim_{n\to\infty} \mathbb{E}(Z(n))$. For the values used in the simulations $K = 6r_\Delta$.

Whenever we want to restrict ourselves to the case of no death of resistant cells we put $\Delta(n) = 0$ for all n.

Updating δ: Fix a number $r_\delta > 0$ and $1/2 < \delta_{min} < \delta_{max} < 1$. We denote $R(n)$ the number of cells recognized by the immune system (that is, sensitive or type-1 cells) at generation n. Let $X_\delta(n)$ be a random variable with Beta distribution of parameters $\alpha = R(n)/r_\delta$, $\beta = 1$.

As before we put

$$\delta(n) = \delta_{min} + (\delta_{max} - \delta_{min})X_\delta(n), \quad n = 1, 2, 3, \ldots$$

We interpret $\delta_{\min}$ as the background probability of natural deaths and deaths of sensitive cells by the action of the immune system; $\delta_{sc}(n) = (\delta_{max} - \delta_{min})X_\delta(n)$ takes into account the mechanism of self-control of sensitive parasite population at generation n.

There is a crucial difference between the variation of $\Delta(n)$ and $\delta(n)$: $\delta(n) > 1/2$ for all n, something that always leads to a reduction in the number of sensitive cells throughout the entire process; $\Delta(n)$ instead, ranges from values below $1/2$ (which slow down but do not stop population growth), to values above $1/2$ that impose an upper bound and prevent population explosion of the resistant clans as already explained.

5.3. Varying μ

Rates of antigenic variation were measured experimentally in *Trypanosoma brucei* (see Barry and Turner, 1991; Turner, 1997 and references therein). Values of parameter μ range from 10^{-2} to 10^{-3} switches/cell/generation in nature (fly-transmitted); or they can be as low as $10^{-5} - 10^{-7}$ switches/cell/generation in syringe-passaged parasites. The former values of μ are comparable to the switching rates observed in other species undergoing antigenic variation, while the latter are in the range of point mutations. This variation in switching rates has been reported only between the different conditions mentioned before, but not during the course of an individual infection. We postulate, however, that such intra-infection variation is feasible and may be adaptive for the parasite since it contributes to chronicity of the infection. Indeed, lowering switching rates when parasite populations are large might prevent unconstrained hyper-use of the silent repertoire (which may produce immune exhaustion). The biological mechanisms governing this variation would be partially overlapping with those that control the division rate (see next section).

In our approach, we consider μ as a random variable updated with n:

$$\mu(n) = \mu_{min} + (\mu_{max} - \mu_{min})X_\mu(n), \quad n = 1, 2, 3, \ldots$$

where $0 \le \mu_{min} \le \mu_{max} \le 1$ and for fixed $r_\mu > 0$ $X_\mu(n)$ is a random variable with Beta distribution of parameters $\alpha = r_\mu/R(n)$, $\beta = 1$. As in the Δ and δ cases, parameter α is updated with n. The effect of variable $\mu(n)$ is to prevent producing a huge number of switching events during the peak of infection while having, on average, a high switching rate close to that reported experimentally when the population is small (which makes the extinction less probable). Such extreme parasite diversity during the peaks would lead to immune system exhaustion and consequently to render the infection uncontrollable.

5.4. Simulations of parasitemia waves

To visualize the dynamics of the process and particularly the conditions that lead to the emergence of parasitemia waves, we implement simulations in populations of cells using the version of the model presented in Sections 2 and 4.1, with random variables $\Delta(n)$, $\delta(n)$ and $\mu(n)$ updated with n as described before. It is worth mentioning that the search in the space of parameters is greatly

reduced thanks to the analytical results presented in Sections 3 and 4. As mentioned, the undulant behavior can most probably (or uniquely) arise in the supercritical region in the neighborhood of the critical surface, and the latter can be explicitly computed using Proposition IV.1.

We denote $R(n)$ (resp. $Q(n)$) the number of sensitive type-1 (resp. resistant type-2) cells at generation n. For all simulations we compute the total population of cells, $Z(n)=R(n)+Q(n)$. In all cases initial condition is $R(0)=5\times 10^3$, $Q(0)=1$.

In order to study the sensitivity of the process with the parameters and choose them properly, we start with the simple case of no death of resistant cells (i.e. $\Delta(n)=0$ for all n).

Case 1: No death of resistant cells: We put $\Delta(n)=0$ and update variables $\delta(n)$ and $\mu(n)$ as explained in Sections 5.2 and 5.3. Figs. 5 and 6 show typical realizations of the process for different values of parameters. The dependence on p is presented in Fig. 5. The simulations show how the behavior changes from extinction to explosion in the narrow range $0.60-0.70$. Intermediate values of p (around $p=0.65$ for the parameters we have chosen in this context) show the oscillatory regime with high probability.

Because $p>1/2$ the process is always in the supercritical region (whenever $Z(n)>0$). Note that in this context ($0.60\le p\le 0.70$) the vast majority of new variants, which are in order of thousands, do not produce parasitemia peaks but small resistant clans (they are rapidly recognized). These clans however play a decisive role in maintaining the infection.

Fig. 6 shows typical realizations of the process for fixed p ($p=0.65$) changing the different values of the parameters associated with δ ($\delta_{\min}$, $\delta_{\max}$) and μ ($\mu_{\min}$, $\mu_{\max}$).

The examples show that our model is able to capture some aspects of the dynamics of antigenic variation. However, the probability of explosion (say, $Z(n)>10^7$) is still rather high when the number of new variants that appear along the process is in the order of thousands. It is worth wondering whether there exists any mechanistic way to exert such control in switching rates. Although such mechanisms were never characterized experimentally, it is feasible that the same or similar mechanisms that work to control cellular division rate may also play a role in controlling variation rate. Note that many of the enzymatic pathways that participate in recombination overlap with those that make part of the DNA duplication machinery.

Case 2: Death of resistant cells We proceed as in Case 1, but also update variable $\Delta(n)$ as explained in Section 5.2. The simulations presented in Fig. 7 show that when there is self-control of resistant cells, since there is no risk of explosions, the behavior of infection is reasonable even when the times of reaction of immune system are long (using values of $p=0.90-0.95$). As a result the infection can survive even when the switching rates μ are low or very low. In these examples we have chosen very small values for $\mu_{\min}$ and $\mu_{\max}$; but it is also possible to keep μ constant (and small) without significantly affecting the overall behavior (Fig. 7 panels c–d). Furthermore, it is worth remarking that the number of variants that appear is low (in the order of tens) and consequently there is not much risk of immune exhaustion.

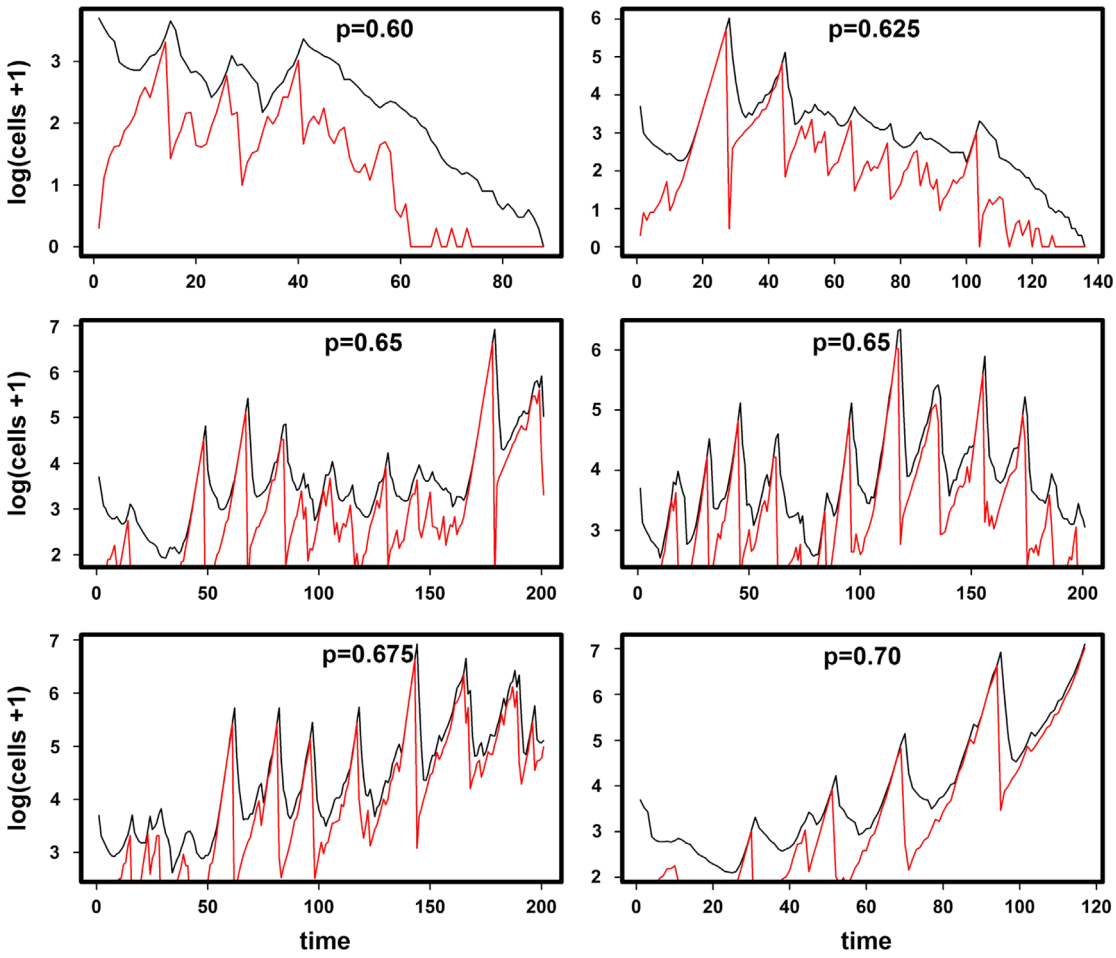

Fig. 5. Numerical simulations of parasitemia waves for different values of p. Black lines represent the total population of cells whereas red lines correspond to resistant cells. The different values of p are indicated in each panel. The minimum and maximum of the other relevant parameters are: $\delta_{min}=0.60$, $\delta_{max}=0.95$, $\mu_{min}=0.004$, $\mu_{max}=0.01$. $r_\delta=10^4$ and $r_\mu=10^2$. Last panel ($p=0.70$) shows an example of what we consider a population explosion: $Z(n)>10^7$ before generation 200. (For interpretation of the references to color in this figure caption, the reader is referred to the web version of this paper.)

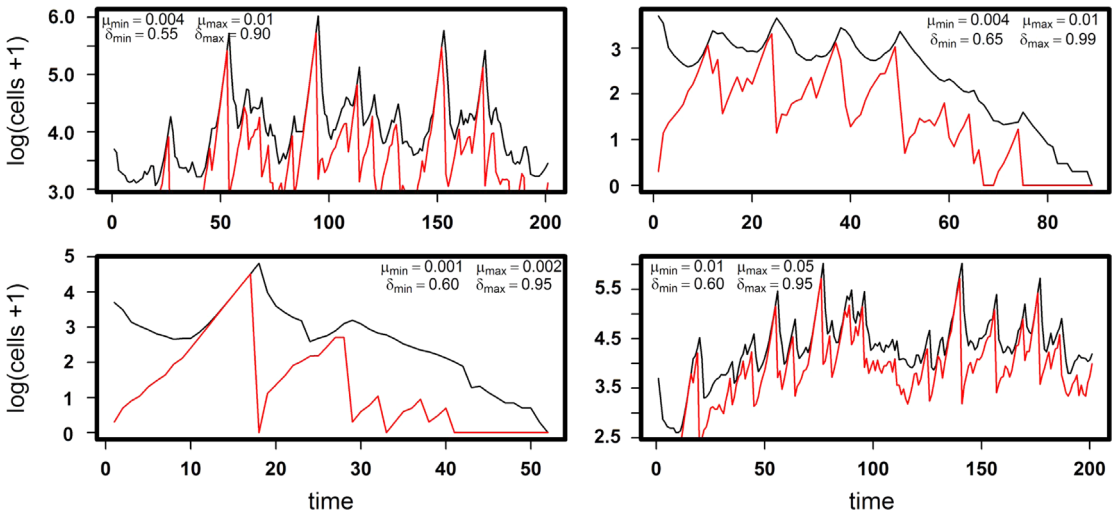

Fig. 6. Numerical simulations of parasitemia waves for different minimum and maximum values of μ and δ, keeping constant $p=0.65$. Black and red lines as in Fig. 5. The different variation ranges for μ and δ are indicated in each panel. $r_\delta = 10^4$ and $r_\mu = 10^2$. (For interpretation of the references to color in this figure caption, the reader is referred to the web version of this paper.)

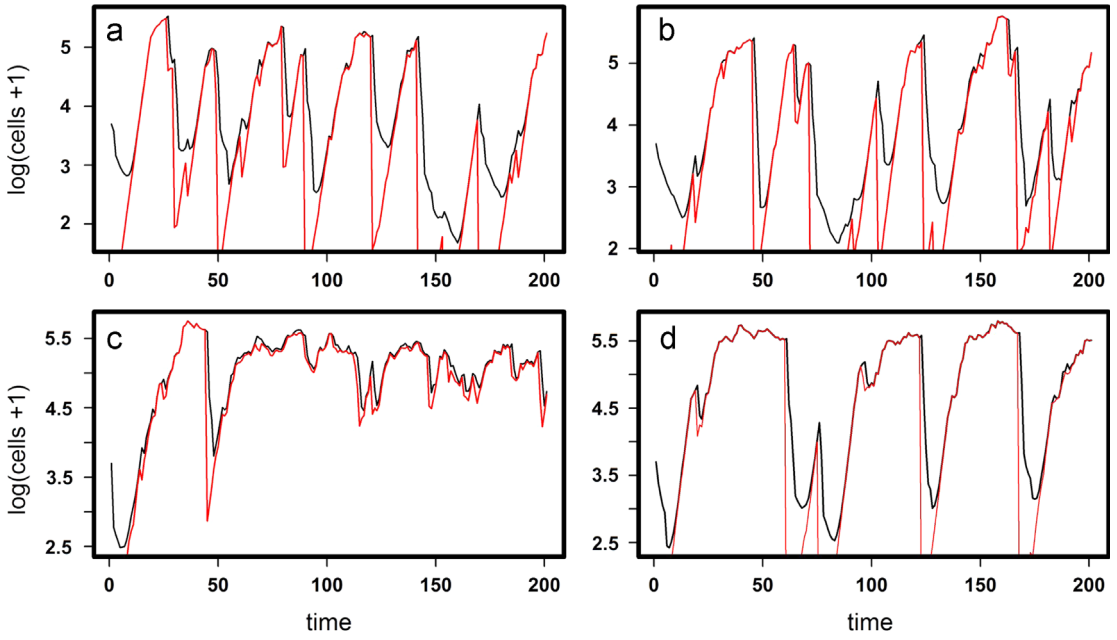

Fig. 7. Numerical simulations of parasitemia waves including self-control of resistant cells. Parameters: $\Delta_{min} = 0.05$, $\Delta_{max} = 0.55$, $\delta_{min} = 0.60$, $\delta_{max} = 0.95$, $r_\Delta = 5 \times 10^4$, $r_\delta = 10^4$ and $r_\mu = 10^2$. Panel a: $\mu_{min} = 0.0001$, $\mu_{max} = 0.004$, $p=0.90$; Panel b: $\mu_{min} = 0.00001$, $\mu_{max} = 0.01$, $p=0.90$; Panel c: $\mu_{min} = \mu_{max} = 0.01$, $p=0.90$; Panel d: $\mu_{min} = \mu_{max} = 0.0001$, $p=0.95$. Black and red lines as in Fig 5. (For interpretation of the references to color in this figure caption, the reader is referred to the web version of this paper.)

6. Discussion

In this paper we introduce a new approach to model the dynamics of antigenic variation using a multi-type branching process. The model considers the following aspects: efficiency of immune response against sensitive cells; rate at which new resistant variants appear; natural death of resistant cells and a random modeling for the delay times spent by the immune system to recognize a new variant.

For the simplest case where the parameters of the model are fixed we characterize analytically the critical, subcritical and supercritical regions on the corresponding spaces. This allows us to draw an interesting conclusion: the undulating behavior typically observed in antigenic variation (parasitemia waves) cannot be explained only as the interplay between new antigens being produced and the immune response removing them. This is because the system is very unstable and goes to extinction or to explosion fast or very fast. Instead, it is necessary to invoke mechanisms of population self-control that can be either similar to quorum sensing (which have been well described in African trypanosomes and they are probably also present in malaria parasites) or simpler ones like those that rely on the limited availability of nutrients and other resources. By appropriately updating the set of parameters we introduce a density-dependent mechanism that accounts for qualitative behavior observed in vivo experiments such as the characteristics peaks of parasitemia along an infection.

The model can be extended to include situations of higher complexity and more realism, as for instance: the delay times to recognize new antigen variants can be modulated by modifying values of parameters p with the size of resistant clan as explained in Section 4.2 (Example 2). In addition, the intensity of immune response can also be controlled with the size of sensitive clan (as explained in Section 5.2, updating δ). Such kind of clan size dependency of immune response efficiency has been discussed

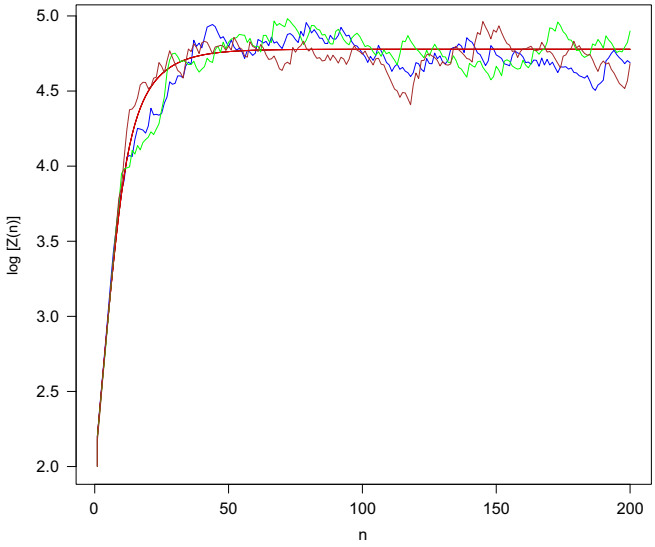

Fig. 8. Simulations of three realizations of the logistic-type Galton–Watson process (blue, green and brown lines) and upper bound for the expectation of the process (red line). $\Delta_{min}=0.20$; $\Delta_{max}=0.55$. $r_o=10^4$. Initial condition: $Z(0)=100$. (For interpretation of the references to color in this figure caption, the reader is referred to the web version of this paper.)

in the literature as a key element to produce predator-prey-like cyclical population dynamics (Nowak and May, 2000; Seger, 1988).

It is worth noting that immune memory can also be incorporated in this model in a relatively straightforward way. This aspect can be interpreted in a broad sense as if some of the new antigenic variants that appear in the course of an infection are not always completely new, something that can result from different situations. One possibility is that the host was already exposed to the same antigen during the current infection and this occurs when the parasite starts re-using antigens. The second alternative occurs when the new variant presents shared epitopes to previously exposed variants, due to sequence or structural similarity. Hence the immune system is able to recognize it, yet with lower affinity. Both situations can be incorporated in the model. The probability of re-using antigenic variants depends on the size of antigenic repertoire and the time elapsed since the infection started. It is evident that it becomes progressively more probable as the infection is more prolonged and also when there are fewer antigenic variants in the repertoire. Therefore this phenomenon is equivalent to decreasing the effective switching rate μ and is a typical urn problem. On the other hand, the case of new variants that present similarity to previously exposed antigens can be modeled by increasing the value of Δ by a fixed number ε (representing partial response of immune system) for all the cells that belong to the clan originated by the new variant. An approach similar to this one is used in Antia et al. (1996).

In our opinion multi-type branching processes have a great potential to model evolution of host-parasite interactions like the particular case of antigenic variation. The approach presented here opens a variety of possibilities for future work.

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Appendix A

Here we show that the density-dependent mechanism of the Galton–Watson process introduced at Section 5.2 gives a logistic-type upper bound for the mean of total number of cells (a situation that represents a self-controlled viral population in the absence of immune response). In our opinion this gives support to the election of a Beta distribution for the variable driven changes of Δ and δ with n.

Let us consider a Galton–Watson branching process in which population of cells at generation n proliferates by binary division and the descendants die, independently, with probability $\Delta(n)$, where

$$\Delta(n)=\Delta_{min}+(\Delta_{max}-\Delta_{min})X(n),\quad n=1,2,3,\ldots$$

Here $X(n)$ is a random variable with Beta distribution of parameters $\alpha=Z(n)/r_o$, $\beta=1$, and $Z(n)$ is the total number of cells at generation n. We fix $r_o>0$, $\Delta_{min}<1/2$ (a parameter's value in the supercritical region of the corresponding homogeneous Galton–Watson process) and $\Delta_{max}>1/2$ (a parameter's value in the subcritical region of the corresponding homogeneous Galton–Watson process).

We obtain a recursive formula for the expectation of $Z(n+1)$ by conditioning on random variables at generation n. Note that $Z(n+1)$ conditioned to $Z(n)$ and $X(n)$ is a Binomial random variable with parameters $2Z(n)$ and $1-\Delta(n)$, so that $\mathbb{E}(Z(n+1))$ can be computed as

$$\begin{aligned}\mathbb{E}(Z(n+1)) &= \sum_{l=0}^{\infty}\int_0^1 dt\ \mathbb{E}(Z(n+1)|Z(n)=l,X(n)=t)\\ &\quad\times\rho_X(t)\mathbb{P}(Z(n)=l)\\ &= \sum_{l=0}^{\infty}\mathbb{P}(Z(n)=l)\int_0^1 dt\rho_X(t)\\ &\quad\times 2l[(1-\Delta_{min})-(\Delta_{max}-\Delta_{min})t],\end{aligned}$$

where $\rho_X(t)$ is the probability density of a random variable, X, Beta distributed with parameters $\alpha=l/r_o$ and $\beta=1$. Using the well-known fact that $\mathbb{E}(X)=\alpha/\alpha+\beta$ we obtain

$$\begin{aligned}\mathbb{E}(Z(n+1)) &= \sum_{l=0}^{\infty}\mathbb{P}(Z(n)=l)2l(1-\Delta_{min})\\ &\quad-\sum_{l=0}^{\infty}\mathbb{P}(Z(n)=l)(\Delta_{max}-\Delta_{min})\frac{2l^2}{l+r_o}.\end{aligned}$$

Finally, denoting $a=(1-2\Delta_{min})$ and $b=2(\Delta_{max}-\Delta_{min})$ (note that $a,b>0$), we obtain the following equation for the increment of expectations:

$$\mathbb{E}(Z(n+1))-\mathbb{E}(Z(n))=a\,\mathbb{E}(Z(n))-b\,\mathbb{E}\left(\frac{Z(n)^2}{Z(n)+r_o}\right)$$

$n=1,2,3,\ldots$ This is a logistic-type difference equation; to make this point more evident we make use of Jensen inequality (see Rudin, 1987) applied to the convex function $f(x)=x^2/(x+r_o)$, $x\geq 0$, to get

$$\mathbb{E}\left(\frac{Z(n)^2}{Z(n)+r_o}\right)\geq\frac{\mathbb{E}^2(Z(n))}{\mathbb{E}(Z(n))+r_o},$$

which gives an upper bound for the increments of expectations:

$$\mathbb{E}(Z(n+1))-\mathbb{E}(Z(n))\leq a\,\mathbb{E}(Z(n))-b\,\frac{\mathbb{E}^2(Z(n))}{\mathbb{E}(Z(n))+r_o}.$$

In Fig. 8 we show the solution of the logistic-type difference equations

$$z_{n+1}-z_n=az_n-b\frac{z_n^2}{z_n+r_o},\quad n=1,2,\ldots$$

with initial condition $z_o=100$. There exists a stationary point at $ar_o/(b-a)=(1-2\Delta_{min})\, r_o/(2\Delta_{\max}-1)>0$, which provides an upper bound for $\lim_{n\to\infty}\mathbb{E}(Z(n))$. Values of parameters are $\Delta_{min}=0.20$, $\Delta_{max}=0.55$ (which give: $a=0.60$ and $b=0.70$) and $r_o=10^4$. In the same plot we present three realizations of the Galton–Watson process. These were numerically obtained using the same values of parameters and initial condition. It is evident that the logistic upper bound fits very well to the observed behavior of the process.

Appendix B. Supplementary data

Supplementary data associated with this paper can be found in the online version at http://dx.doi.org/10.1016/j.jtbi.2015.06.025.

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