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MODELING PARASITE REDUCTION MECHANISMS IN CHAGAS DISEASE

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Various procedures have been proposed to reduce parasitemia during Chagas disease. Here we analyze in detail models developed to predict the possible outcomes of two of these treatments. The model solutions reproduce available experimental population data and can be used as tools to help researchers to find a cure or, at least, an optimal treatment to reduce cardiac cell damage. In particular, we study how the phase diagram describing the infection outcome is modified by the application of the ganglioside GM1, finding the optimum concentration of the drug in terms of the parameters characterizing its influence on the parasite-host interaction. We also investigate the use of the non-pathogenic parasite *T. rangeli* to reduce *T. cruzi* load in a mixed infection, finding that this method is not effective against all *T. cruzi* strains. Furthermore, we compare phase portraits of the evolution of the disease for a single and a mixed infection, and evaluate the cell damage as a function of the time elapsed between both infections, remarking on the temporary protective effect of the reaction to *T. rangeli*.

Keywords: Chagas Disease; *T. cruzi*; *T. rangeli*; GM1; Hormesis.

1. Introduction

Chagas disease is the most important Latin American endemic human parasitosis.[1] It is mainly a vector-borne disease in the Americas, although, due to blood transfusion, it has also been identified in Europe and North America. Vector-borne cases of Chagas disease have likewise been reported in the southern United States.[2] The World Health Organization estimates that 90–100 million people are at risk of infection and that 10–20 million are actually infected, with an annual incidence of 560,000 new cases.[3] It is estimated that there are about 50,000 annual deaths in the world as a result from chronic cardiomyopathies caused by the disease, whose causative agent is the unicellular flagellate *Trypanosoma cruzi*. This protozoan is transmitted to humans and other mammals by an hematophagous insect belonging to the *triatominae* subfamily. *T. cruzi* presents a heterogeneous clonally structured population showing extremely high levels of genetic diversity. In the mammal host, the parasite replicates inside the cells of a variety of tissues (with marked

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preference for cardiocytes and smooth muscles), producing a low, but continuous, cellular damage. After an undetermined phase of 15–20 years, the condition of the infected people worsens due to the loss of up to 80% of the cardiac ganglion cells, ending in chronic cardiomyopathy.

No really effective cure has been developed for Chagas disease. The main commercially available treatment is the parasiticide drug benznidazole, in spite of its low levels of efficacy. Such treatment is thought to prevent pathology during the late chronic stages of the infection, but efficacy and tolerance of benznidazole is inversely related to the age of the patient, its side effects being more frequent in elderly patients.[4] Several new methods have been proposed to reduce *T. cruzi* parasitemia levels. In particular, Cossy Isasi and coworkers[5] reported that the monosialoganglioside GM1 was effective to control parasitemia and enhance survival in mice infected with a lethal amount of parasites. Intramuscular treatment with 0.1 mg of GM1 during the acute phase reduced parasitemia to undetectable levels, while mice survived for more than one year after the infection. In fact, histological examination of different organs failed to show parasite nests after treatment. A generalization of our model[6–8] for the *T. cruzi* — immune system interaction was used to explain the strikingly nonlinear reaction observed in mice inoculated with GM1,[9] which had previously been shown to lead to a strong parasite load reduction at low doses, but to a parasite load increase at high doses.[5] Following experimental observations, we considered that GM1 modulates the physical boundary involved in the processes of host cell invasion and emergence after parasite multiplication,[9] strongly influencing parasite replication rates. Our model reproduces this effect, predicting an optimal dosage of 0.15 mg of GM1, which maximizes parasite removal with minimal cell destruction. This value agrees with the experimental observations.

Another method proposed to reduce or eliminate parasitemia in the acute phase of Chagas disease is by pre-infecting the mammal host with another parasite, in particular, with *T. rangeli*. This is a protozoon non-pathogenic to vertebrate hosts[10] that shares many antigens, endemic areas, and vectors with *T. cruzi* widely across Latin America.[11] As a consequence, mixed *T. cruzi*–*T. rangeli* infections can occur, making a Chagas disease diagnosis difficult. On this basis, several researchers, such as Basso,[11–16] Zuñiga[17] and Palau[18] have proposed a vaccination procedure for Chagas disease. These authors performed experiments in mice and dogs, using *T. rangeli* epimastigotes to generate protection against a subsequent *T. cruzi* infection by inducing an overproduction of antibodies. They found that the *T. cruzi* parasitemia level was lower and lasted a shorter period of time in pre-infected animals.

We have modeled previously infections for both single *T. rangeli*, and mixed *T. rangeli*-*T. cruzi* parasitemias.[19] Considering that there are no observations of intracellular *T. rangeli* replication,[20] our model assumes that *T. rangeli* trypomastigotes reproduce by binary fission in the bloodstream, neglecting the existence of any intracellular form like that characteristic of the *T. cruzi* replication. The model reproduces the mentioned experimental data, and shows that the *T. cruzi* parasitemia can be reduced, and even eliminated, as a consequence of the antibody

level rise due to a pre-infection with *T. rangeli*. But, as the mammal immune system is able to eliminate the single *T. rangeli* infection, the host high antibody levels, needed to resist the Chagas infection, are reduced with time. As a result, the immune system returns to its initial untreated state, demonstrating that a pre-infection with *T. rangeli* induces a protective but only temporary effect against Chagas disease.

Here we further investigate the parasite evolution during the acute phase of the Chagas infection when a host is treated with the proposed methods. In particular, we analyze in detail the mathematical properties of the model, on the basis of the experimental data, presenting informative phase diagrams and phase portraits.

The rest of this paper is organized as follows: In Sec. 2 we review the original *T. cruzi* model, and in Sec. 3, we present the model variations for each antiparasite method, with the corresponding analysis. In Sec. 4 we conclude with a short discussion of the results.

2. The Model

To formulate our basic model,[6,8] we assume that the population of circulating parasites is controlled mainly by antibodies. The number of antibodies evolves following the changes in the parasite density in the plasma. These antibodies mediate parasite removal either through lysis, when antibodies are recognized by complement, or through phagocytosis when they are recognized by phagocytic cells. The model for the interaction between *T. cruzi* and its antibodies is then built upon the following assumptions:

(1) A parasite batch invades or is inoculated into the mammalian host.
(2) Trypomastigotes invade healthy cells, where they mutate into amastigotes and reproduce.
(3) Newborn parasites break out of the cell and restart the invasive process.
(4) Parasites activate antibody generation.
(5) One or more antibody species mediate extracellular parasite removal.

These assumptions are used to formulate coupled first-order equations for the time dependence of the cell and antibody populations. Defining the infectivity ζ as the rate at which a circulating parasite (trypomastigote) penetrates a host cell and the cytotoxicity η as the rate at which infected cells burst, the equation for the number $r(t)$ of infected cells is,

$$\frac{dr(t)}{dt} = \zeta n(t) - \eta r(t). \tag{2.1}$$

If N_r is the mean number of trypomastigotes emerging from a ruptured cell and N is the number of antibody species present, the evolution equation for the number

$n(t)$ of circulating trypomastigotes can be written as,

$$\frac{dn(t)}{dt} = \eta N_r r(t) - \sum_{i=1}^{N} \alpha_i(t) a_i(t) n(t) - \zeta n(t), \tag{2.2}$$

where α_i is the removal efficiency for antibodies of species i and $a_i(t)$ is the number of molecules of the i-th antibody species, normalized by the average number of molecules removed by a single parasite.[6] Since antibody avidity increases with time, we choose $\alpha_i(t)$ to be a smoothly increasing function of time,

$$\alpha_i(t) = \alpha_{Ai} + \alpha_{Bi}[1 - \exp(-t/T_i)], \tag{2.3}$$

where α_{Ai} and $\alpha_{Ai} + \alpha_{Bi}$ are, respectively, the initial and saturating values, the latter being reached after a "learning" time T_i.

If γ_i and τ_i are, respectively, the production rate and the intrinsic lifetime of antibodies belonging to species i, and a_{i0} represents a continuous source exactly compensating for the spontaneously inactivated antibody molecules in the absence of parasites, the antibody numbers evolve according to,

$$\frac{da_i(t)}{dt} = \gamma_i n(t) - \alpha_i(t) a_i(t) n(t) + \frac{1}{\tau_i}[a_{i0} - a_i(t)]. \tag{2.4}$$

By assuming a Malthusian model for the amastigote replication dynamics, we can relate the parameters η and N_r to the mean amastigote duplication time, Ω,

$$\Omega = (1/\eta)(ln2/lnN_r). \tag{2.5}$$

Once the size n_0 of the initial inoculation is known, the set of equations (2.1) to (2.4) can be solved to obtain the time dependences of the various populations. The steady-state solutions have been studied in detail in previous work.[8] In the $N = 1$ case there is a "healing" solution, for which $n = r = 0$ and $a = a_{i0}$, and a chronic disease solution, given by,

$$\bar{a} = \frac{(N_r - 1)\zeta}{\alpha}, \tag{2.6}$$

$$\bar{n} = \frac{(N_r - 1)\zeta - \alpha a_0}{\alpha\tau[\gamma - \zeta(N_r - 1)]}, \tag{2.7}$$

$$\bar{r} = \frac{\zeta}{\eta}\bar{n}. \tag{2.8}$$

Of course, if $N_r < 1$ only the healing solution is possible. The Routh-Hurwitz criterion can be used to study the stability of these solutions.[21] This criterion leads to three possible outcomes: healing, chronic disease, and host death. Death of the host is characterized by a divergence of $n(t)$ at long times.

The results of *T. cruzi* invasion are best described by using a phase diagram in the plane defined by the parameters γ and N_r (Fig. 1). We observe that:

(1) The borderline between the surviving (healing and chronic) and host death cases is given by the straight line $\gamma = \zeta(N_r - 1)$.

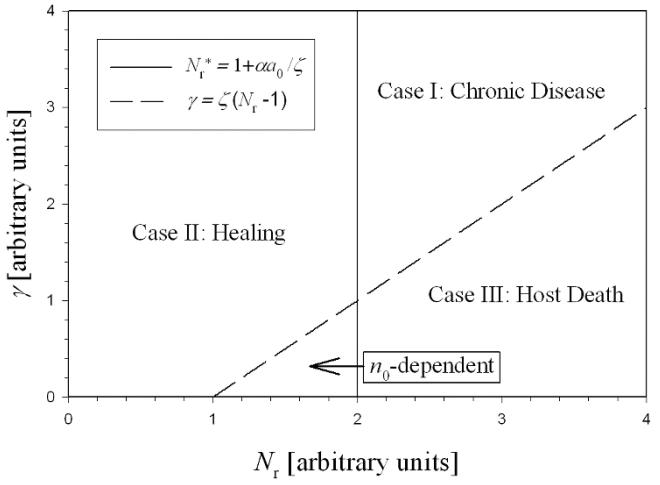


Fig. 1. $\gamma - N_r$ phase diagram representing the possible outcomes of a *T. cruzi* infection with a single antibody species. Parameter values: $\alpha = 0.1$, $\zeta = 1$, and $a_0 = 10$. From Ref. 8.

(2) The phase boundary between the chronic and healing cases is a vertical line located at $N_r^* = 1 + \alpha a_0/\zeta$.
(3) The triple point, where the three cases meet is located at $\gamma = \alpha a_0$.
(4) There is a region (the lower triangle in Fig. 1) where the outcome depends on n_0.

3. Specific Models for Parasite-Reduction Mechanisms

3.1. *GM1 beneficial action*

The beneficial effects of moderate doses of the ganglioside GM1 on infected murine models were investigated in 1999 by Cossy Isasi and coworkers.[5] More recently, fluorescence experiments showed that the addition of GM1 results in an increase in microfluidity in both infected and noninfected cardiocytes.[9] This increase in fluidity is assumed to lead to a decrease in the citotoxicity $\eta = \eta(c)$, where c is the GM1 concentration. Since the amastigote replication time Ω should not be affected by the ganglioside, the number of parasites liberated in a cell breakup, N_r, increases according to the relation $N_r = 2^{\frac{1}{\eta\Omega}}$. If parasite penetration is impaired by GM1 action,[9] the infectivity $\zeta = \zeta(c)$ is also reduced. The simplest way to account for the GM1 effect on infectivity and cytotoxicity is to assume the following functional dependences on c,

$$\zeta' = \frac{1}{1 + \lambda_1 c}\zeta, \tag{3.1}$$

and

$$\eta' = \frac{1}{1 + \lambda_2 c}\eta. \tag{3.2}$$

The parameters λ_1 and λ_2 characterize the influence of GM1 on the parasite and the target cell. Since the model equations are modified by the transformations specified by Eqs. (3.1) and (3.2), the location of the phase diagram boundaries depends on the variable c and on the parameters λ_1 and λ_2. Using Eq. (3.2), we see that the number of parasites emerging per cell burst is increased to $N_r' = N_r^{1+\lambda_2 c}$. Given that N_r' depends on c, it is convenient to continue building and analyzing the phase diagram in the $N_r - \gamma$ plane. This allows for a simple identification of the effects of the ganglioside on the infection outcome and for a straightforward comparison with the untreated case.

The vertical line separating the chronic (I) and healing (II) cases, ${N_r}^* = \alpha a_0/\zeta + 1$, is now given by,

$$N_r^*(c, \lambda_1, \lambda_2) = \left[\frac{\alpha a_0}{\zeta}(1 + \lambda_1 c) + 1)\right]^{\frac{1}{1+\lambda_2 c}}. \tag{3.3}$$

The boundary separating regions I and II from region III (death), which in the case $c = 0$ was a linear function of N_r is now transformed into the convex curve

$$\gamma(c, N_r) = \frac{\zeta}{1 + \lambda_1 c}(N_r^{1+\lambda_2 c} - 1). \tag{3.4}$$

For low values of c the dependence on this variable is linear and the slope of the main term decreases, leading to an expansion of regions I and II at the expense of region III. However, as c grows, the exponential dominates and regions I and II shrink, in agreement with the observed hormesis phenomenon. In the limit $c \to \infty$, the curve $\gamma(c, N_r)$ tends to the vertical line $N_r = 1$. As a consequence, for high GM1 doses, death becomes the disease outcome if $N_r > 1$. With the introduction of GM1, the phase boundaries, which were independent of the cytotoxicity when $c = 0$, become cytotoxicity-dependent through the parameter λ_1.

Next we search for the values of the ganglioside concentration that are most favorable to control the infection. In particular, we would like to maximize the size of region I (healing) in the phase diagram. Therefore, we look for the value c_{opt} of the GM1 concentration that maximizes the rightward shift of the vertical boundary given by Eq. (3.3). This value satisfies the equation,

$$\left.\frac{dN_r}{dc}\right|_{c_{\text{opt}}} = 0. \tag{3.5}$$

Taking the natural logarithm of both sides of Eq. (3.3) and using implicit derivation, we arrive at,

$$\frac{\lambda_2}{1 + \lambda_2 c_{\text{opt}}} ln\left[\delta + (\delta - 1)\lambda_1 c_{\text{opt}}\right] = \frac{(\delta - 1)\lambda_1}{\delta + (\delta - 1)\lambda_1 c_{\text{opt}}}, \tag{3.6}$$

where $\delta = 1 + \alpha a_0/\zeta$. Since we are interested only in nonnegative values of c_{opt}, we can take $c_{\text{opt}} = 0$ in Eq. (3.6) to obtain an upper bound for the values of the ratio λ_2/λ_1 for which a GM1 inoculation may be useful:

$$\frac{\lambda_2}{\lambda_1} \leq \frac{\delta - 1}{\delta ln(\delta)}. \tag{3.7}$$

Given that $\delta > 1$, the right-hand-side of Eq. (3.7) is always smaller than unity, Therefore $\lambda_2/\lambda_1 < 1$ is a necessary condition for GM1 to be beneficial.

By solving Eq. (3.6), we find the maxima of the curves $N_r^*(c)$, which are represented in Fig. 2 for various values of λ_1 and in Fig. 3 for various values of λ_2. Inspection of Eq. (3.3) and of Fig. 2 indicates that an increase of the value of λ_1 has, for fixed c, two beneficial effects:

(1) It increases the parameter range for which host healing is the infection outcome.
(2) It decreases the optimal dosage c_{opt}. This has the advantage of reducing possible unwanted side effects of the drug.

From Fig. 3 we see that, for fixed λ_1 the GM1 benefits decrease monotonically with λ_2; we also see that the size of the optimal dose is reduced with an increase of λ_2.

The effects of a change in the cytotoxicity sensibility parameter λ_2 are clearly exhibited in Fig. 4, where we show the modifications to the phase diagram of Fig. 1 resulting from an application of GM1. If λ_2 is relatively large, as in Fig. 4(a), the beneficial effects of the ganglioside are weak. In the case depicted in that figure, N_r^* changes only from 1.1 to 1.77 under the application of the optimal dose. For

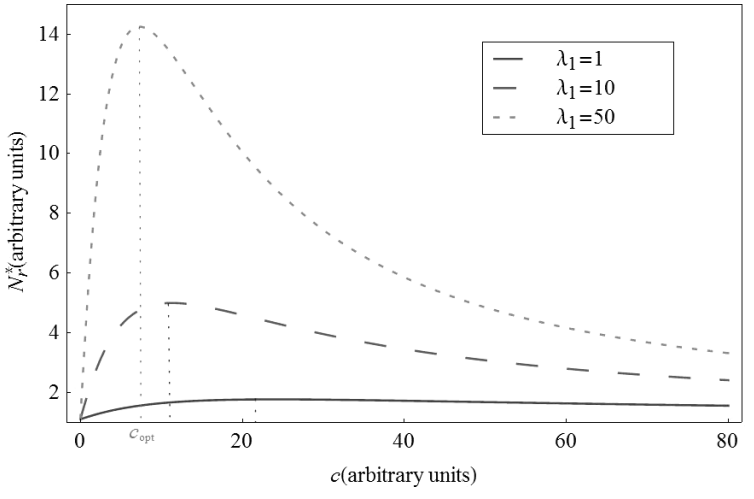


Fig. 2. Location of the separation line, N_r^*, between the healing and chronic regimes as a function of the ganglioside concentration c for the indicated values of the infectivity parameter λ_1. Here $\alpha = 0.01$, $\zeta = 1$, $a_0 = 10$, and $\lambda_2 = 0.05$.

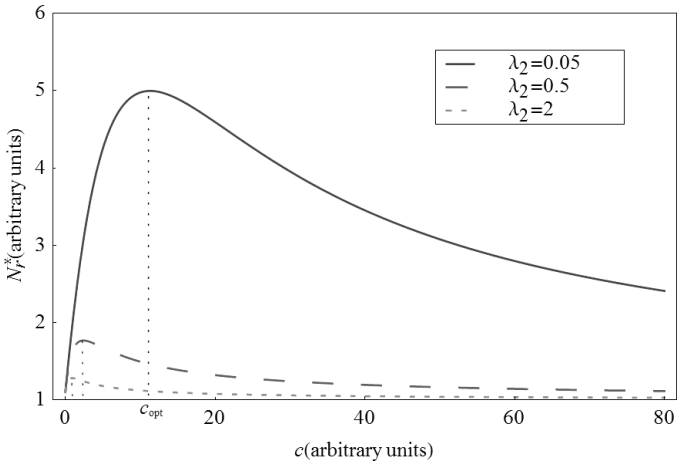
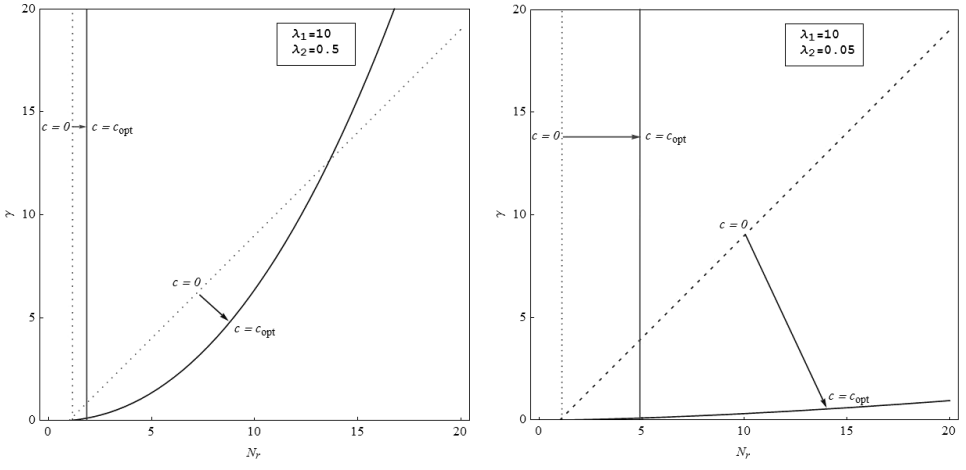


Fig. 3. Location of the separation line, N_r^*, between the healing and chronic regimes as a function of the ganglioside concentration c for the indicated values of the citotoxicity parameter λ_2. Here $\alpha = 0.01$, $\zeta = 1$, $a_0 = 10$, and $\lambda_1 = 10$.

smaller values of λ_2 (see Fig. 4(b)), the drug beneficial effects are much stronger. Note that the space of parameters leading to host death has been confined to very small values of the immune reaction rate γ. The critical parasite burst number N_r^* now changes from 1.1 to 4.90.



(a) (b)

Fig. 4. Phase diagrams for $c = 0$ (dashed lines) and $c = c_{\rm opt}$ (solid lines) for (a) $\lambda_2 = 0.5$ and (b) $\lambda_2 = 0.05$. Note the redistribution of the regions in Fig. 1 and the strong growth of the healing and chronicity regions with a decrease in the ratio λ_2/λ_1. Here $\alpha = 0.01$, $\zeta = 1$, $a_0 = 10$, and $\lambda_1 = 10$.

3.2. *A mixed infection with T. rangeli*

Another method to reduce *T. cruzi* parasitemia levels is by pre-infecting the mammal host with the non-patogenic trypanosoma *T. rangeli*. We thus need to investigate the consequences of a mixed infection with *T. rangeli* and *T. cruzi*.

As a first step we modeled a single *T. rangeli* infection. Considering the characteristics of the immune system-*T. rangeli* interaction we worked on an infection model for trypanosomiasis,[7,22] which only takes into account the extracellular replication of the parasite. In the model, the number $m(t)$ of *T. rangeli* parasites increases at a rate κ_m due to their reproduction by binary fission, and decreases due to the interaction with antibodies. As in the model for *T. cruzi* presented in Sec. 2, *T. rangeli* removal is considered through the set of coefficients $\alpha_{i,m}(t)$. They also have the same learning process until reaching saturation level. The time dependence of the parasite number is then described by the equation

$$\frac{dm(t)}{dt} = \kappa_m m(t) - \sum_{i=1}^{N} \alpha_{i,m}(t) a_i(t) m(t). \tag{3.8}$$

Since the immune system reaction is similar to that corresponding to an infection with *T. cruzi*, the equation for the antibody evolution remains formally the same as Eq. (2.4), but considering the *T. rangeli* population instead.

Although the evolution of the system described by this model has three possible outcomes (healing, chronic and death), in experiments found in the literature for single infections with any of these parasites, the immune reaction is strong enough to effectively reduce the possible outcomes of the infection to two cases, chronic infection or parasite elimination, depending on the size of the parasite replication rate κ compared to αa_{i0}. The reported death cases are due to the big transient parasitemia that may develop before the system reaches its asymptotic state. Particularly, experiments with *T. rangeli* have only been reported to end in a healthy state, indicating that the immune system is able to eliminate the parasite infection, returning the system to the initial conditions ($m = 0, a = a_0$). To illustrate the innocuous nature of *T. rangeli*, compared with the harmful *T. cruzi*, we can associate to each experiment a point in the plane $\alpha a_{i0} - \kappa$ ($\kappa_n = \zeta(N_r - 1)$ for *T. cruzi*, an effective reproduction rate) considering the parameter values obtained in previous works.[8,19,22] Figure 5 shows that in all experiments $\kappa_m < \alpha_m a_{i0}$ for the *T. rangeli* infection, leading to the healthy state, whilst for a *T. cruzi* infection $\kappa_n > \alpha_n a_{i0}$ in agreement with the chronic state reported in murine experiments.[23]

As we mentioned in the Introduction, Basso and coworkers[11–16] showed that *T. rangeli* offers a protective effect against infections with *T. cruzi*. In their experiments, they first pre-infected mice with *T. rangeli*, introducing the *T. cruzi* parasites a certain number of days later. They observed that the *T. rangeli* pre-infection induces some immunity, causing a strong reduction in parasitemia and mortality levels when compared with non-vaccinated or pre-infected mice.

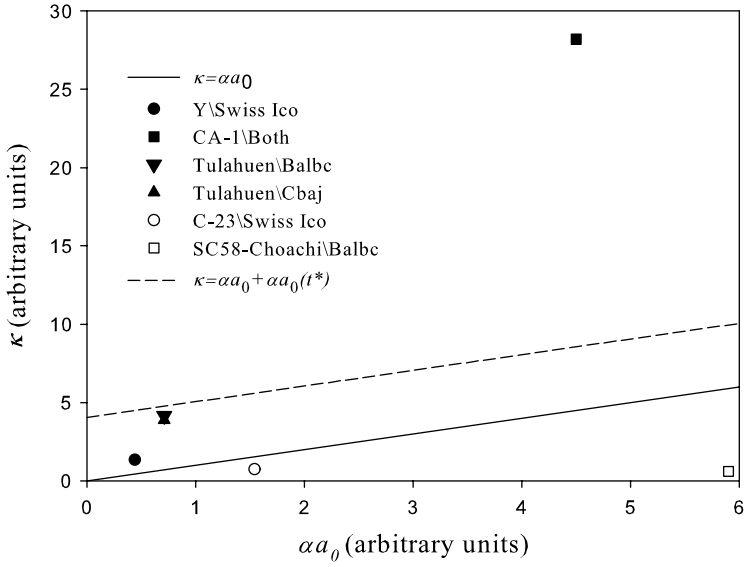


Fig. 5. Comparison between the experimental results for *T. cruzi* and *T. rangeli* in the $\kappa - \alpha a_{i0}$ space. a_{i0} is measured in ml^{-1}, $\alpha_{i,j}$ in (ml/days), and κ_j in days^{-1}. The solid and dashed lines correspond, respectively, to the single infection and the mixed infection with a gap time of 22 days (see text).

The model of the mixed infection must have a differential equation for each parasite, identical to those formulated for single parasite infection models (Eqs. (2.2) and (3.8)), while the antibody time evolution equation include terms for the interaction of the immune system with both *T. rangeli* and *T. cruzi*:

$$\frac{da_i(t)}{dt} = \gamma_{i,m} m(t-\theta_{i,m}) + \gamma_{i,n} n(t-\theta_{i,n}) - \alpha_{i,m}(t) a_i(t) m(t) - \alpha_{i,n}(t) a_i(t) n(t) + \frac{1}{\tau_i}[a_{i0} - a_i(t)]. \tag{3.9}$$

Here, m refers to the *T. rangeli* and n to the *T. cruzi* parasite populations.

To obtain the steady state populations we study the asymptotic ($t \to \infty$) behavior, by setting equal to zero the time derivatives in the system evolution equations, as it was done in Sec. 2 for a single *T. cruzi* infection. The outcomes now depend on two parameter sets, one for each parasite. Due to the broad *T. cruzi* genetic diversity, resulting in a wide range of parameter values, we will analyze the possible outcomes considering the *T. rangeli* parameter values obtained previously from the single infection experiments. Thus we will work under the conditions $\kappa_m < \gamma_m$ and $\kappa_m < \alpha_m a_0$, which yield three outcomes as follows:

(I) *Healing.* If the effective parasite reproduction rate $\kappa_n = (N_r - 1)\zeta$ is lower than $\alpha_n.a_0$, both parasite populations are eliminated at long times and the

system returns to the initial healthy conditions ($r_s = 0$, $n_s = 0$, $m_s = 0$ and $a_s = a_0$), for any value of γ_n.

(II) *Chronic disease caused by T. cruzi infection.* The *T. rangeli* population disappears, but the system reaches a long-time equilibrium state between *T. cruzi* and its antibodies described by:

$$r_s = \frac{\zeta}{\eta} n_s, \quad n_s = \frac{\alpha_n a_0 - \kappa_n}{\tau \alpha_n (\gamma_n - \kappa_n)}, \quad a_s = \frac{\kappa_n}{\alpha_n}. \tag{3.10}$$

The conditions to obtain this state are, $\kappa_n > \alpha_n a_0$ and

$$\gamma_n > \frac{B_0 + B_1 \kappa_n + B_2 \kappa_n^2 + B_3 \kappa_n^3}{A_0 + A_1 \kappa_n + A_2 \kappa_n^2} = \gamma_c, \tag{3.11}$$

where A_i and B_i are constants that depend on the parameters ζ, α_n, α_m, η, a_0 and κ_m.

(III) *Host death.* If $\gamma_n < \gamma_c$ and $\kappa_n > \alpha_n a_0$, the immune system cannot control the infection. This case is divided into two sub-cases:

(III.a) If $\gamma_n > \frac{\alpha_n}{\alpha_m} \kappa_m$, host death is due to the unlimited growth of the *T. cruzi* parasite population alone while *T. rangeli* is eliminated. While $n(t)$ grows without control, the antibody population reaches the asymptotic value,

$$a_s = \frac{\gamma_n}{\alpha_n}. \tag{3.12}$$

(III.b) If $\gamma_n < \frac{\alpha_n}{\alpha_m} \kappa_m$ the host death results from the unlimited growth of both the *T. rangeli* ($m(t)$) and *T. cruzi* ($n(t)$) parasite populations, while the antibody numbers go to the same value of case III.a (expressed by Eq. (3.12)).

The stability of the steady-state solutions was proved using the Routh-Hurwitz criterion.[21] The separation among the different cases can be better observed in a phase diagram defined by the growth rates γ_n and κ_n, assuming one average antibody species. The phase diagram can be found in Ref. 19, where the steady states are shown for different parameter sets.

But what happens if there is a delay between the onsets of both infections? Now the outcome of the *T. cruzi* infection will depend on the transient state of the host immune system due to the previous *T. rangeli* infection. It will produce a priming of the immune system, raising the antibody populations while *T. rangeli* is completely eliminated. When the *T. cruzi* parasites are introduced into the host, the high level of antibodies that they find could be capable of removing them.

In Fig. 5 we observe how several *T. cruzi* strain-mice combinations, belonging to the chronic case for a single infection, switch to a healthy case by the shifting of the borderline from $\kappa = \alpha_{i,n} a_{i,0}$ to $\kappa = \alpha_{i,n} a_{i,0}(t* = 22\,\text{days})$. This is the maximum shift of the boundary since $a_{i,0}(t* = 22\,\text{days})$ is the largest number of antibodies

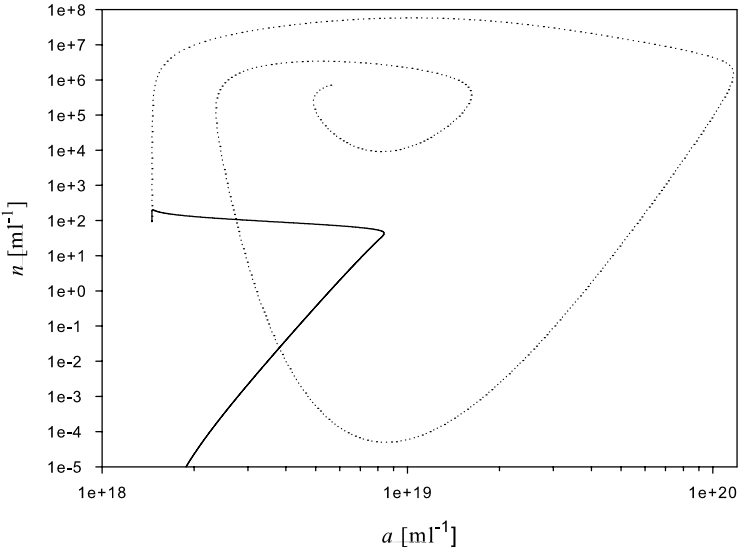


Fig. 6. Phase portraits for the antibody-*T. cruzi* populations obtained with the model, considering the parameters corresponding to fits to el Bouhdidi's data.[23] The dotted line corresponds to a single *T. cruzi* infection, while the full line corresponds to mice pre-infected 22 days earlier with *T. rangeli*.

resulting from the pre-infection. The figure also reveals that one of the strains (CA-1) cannot be eliminated by this method because the antibodies induced by the first infection are insufficient to avoid the chronic condition.

In Fig. 6 we observe phase portraits corresponding to the experiment of el Bouhdidi *et al.*,[23] who infected Balbc mice with the *T. cruzi* Tehuantepec strain. The dotted line represents the chronic case predicted by the model for a single *T. cruzi* infection, which agrees with the experimental results obtained with a small parasite inoculum. For high inocula, the system oscillates until it reaches its attractor, as shown in the figure. The solid line represents how the evolution of the infection is modified if the mice are first pre-infected with *T. rangeli* (considering the parameters obtained from the fit made to the Zuñiga experiment in Ref. 19). The parasitemia is controlled and the system returns to its healthy initial condition $(n = 0, a = a_0)$.

The high level of antibodies is, however, not permanent. Eventually, the immune system eliminates the *T. rangeli* infection returning the system to its initial healthy conditions $(m = 0, a = a_0)$, depriving the host of the acquired immune state. In Fig. 7 we show the total number of lost cells due to the *T. cruzi* infection as a function of the gap time between the start of both infections. Note that, for the parameter values considered and for Chagas infections occurring during the first 3 months after the pre-infection, the immune system will reduce the cell damage by more than four orders of magnitude, providing the host with a chance to heal

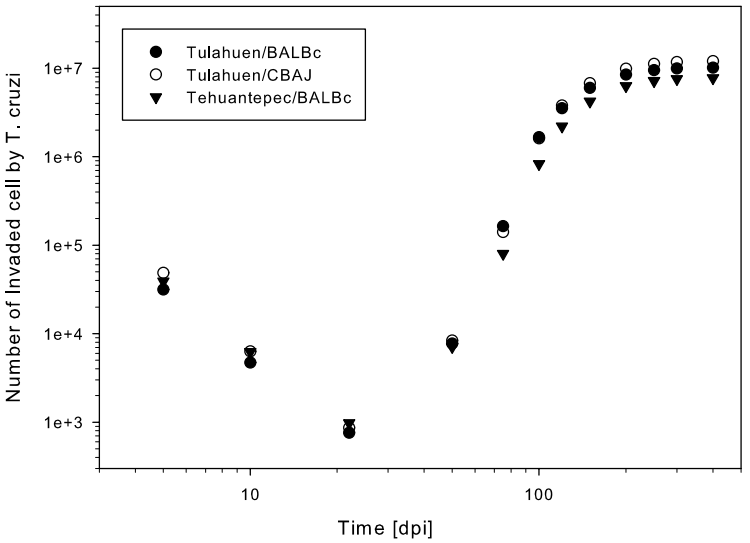


Fig. 7. Total number of lost cells due to the infection as a function of the time elapsed between the onsets of the *T. rangeli* and the *T. cruzi* infections, for different parasite strain/mice species combinations.

(as it was shown previously in the (α_n.a_0-κ) phase diagram). As a result, regular *T. rangeli* pre-infections must be induced in order to keep the immune system prepared to eliminate a Chagas infection. A more practical possibility would be to regularly infect domestic animals with *T. rangeli* three times per year, because these animals are reservoirs for the *T. cruzi* transmission cycle. In turn, it will reduce parasite levels in vectors, preventing a possible Chagas disease in-house contagion.

4. Discussion

In this work we have further analyzed a model for the *T. cruzi* — immune system competition in Chagas disease. In particular, we studied in detail two extensions developed to describe and predict the host's parasitemia levels while it is under one of the proposed treatments. The models yield a very good agreement with observed parasite and immunoglobulin populations, validating the corresponding model assumptions.

The first model extension describes the protective effect of moderate doses of the ganglioside GM1, for which experiments had identified a strong hormesis effect on mammalian hosts: while low doses are clearly beneficial, high doses turn out to be harmful to the host. Here we predict the dependence of the optimal GM1 concentration on the parameters characterizing the effect of the drug on the infectivity and the cytotoxicity of a given host-parasite strain combination. We found that the ganglioside is most efficient when it has a relatively weak effect on the cytotoxicity (small values of the parameter λ_2). We suggest that more experiments

should be carried out using various parasite strains and mammal models to have a comprehensive view of the actual effectiveness of the GM1 inoculation against the infection.

The second model extension describes the induction of a *T-rangeli* pre-infection as a way to reduce the severity of a *T. cruzi* infection. In this case, we calculate for the first time the cellular damage to the host as a function of the time elapsed between the *T. rangeli* pre-infection and the *T. cruzi* infection for the strains for which experimental results are available. Proof of the strength of the *T. rangeli* protective effect is the reduction in the amount of cell damage by up to four orders of magnitude. One limitation of this antichagasic method is its temporary effect. As a practical procedure, we thus suggest periodic *T. rangeli* inoculations of domestic animals, especially dogs, which are the main *T. cruzi* reservoirs in the affected geographic areas. Another limitation is that the temporary protective effect acquired by the host against *T. cruzi* is not observed for all *T. cruzi* strains. For instance, the infection with the CA-1 strain presents a late and mild parasitosis, which corresponds to a very high ζ value in our model, resulting in a higher number of infected cells. The parasites inside the cells do not suffer from the effects of high antibody levels, giving them a chance to evade the temporarily enhanced immune attack.

These results indicate the usefulness of our model as a tool to investigate and predict the effects of various prevention methods and therapies that may be proposed to either heal the infection or at least reduce the damage it generates in the host's cardiac cells.

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