

Dynamics of Co-infection with *M. tuberculosis* and HIV-1

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Since 1985, there has been a renewed epidemic of tuberculosis (TB) that was previously thought to be in check. There is evidence to believe the main factor for this resurgence has been the human immunodeficiency virus (HIV). Co-infection with HIV and *M. Tuberculosis* has profound implications for the course of both diseases. This study represents a first attempt to understand how the introduction of an opportunistic infection, namely *Mycobacterium tuberculosis*, the bacteria that causes TB, affects the dynamic interaction of HIV-1 and the immune system. We create a mathematical model using ordinary differential equations to describe the interaction of HIV and TB with the immune system. It is known that infection with TB can decrease the CD4⁺ T cell counts—a key marker of AIDS progression; thus, it shortens survival in HIV infected individuals. Another main marker for HIV progression is the viral load. If this load is increased due to the presence of opportunistic infections, the disease progression is much more rapid. We also explore the effects of drug treatment on the TB infection in the doubly-infected patient. © 1999 Academic Press

1. INTRODUCTION

Tuberculosis (TB) has been a leading cause of death in the world for centuries. During the period from 1945 to 1985, because of improved medical treatment and hygiene practices, the number of cases of TB steadily declined. However, from 1985 through 1992, in the US alone, there were over 50,000 extra cases of TB (CDC, 1993). Today it remains the leading cause of death by pathogen-induced diseases world wide—3.1 million in 1996. It is believed this recent increased incidence is in large part due to HIV. Other factors certainly include an increase in poverty levels as well as the dismantling of TB control programs. Also, since this resurgence of TB in 1985, TB has been called the “Main opportunistic disease for HIV” (Martin *et al.*, 1995). It is clear that each of these diseases

can have a profound impact on the other (Shafer *et al.*, 1996).

First, it has been shown (Edlin *et al.*, 1992; Selwyn *et al.*, 1989) that HIV-infected individuals are at an increased risk of developing TB in the active form. Second, there is an increasing interaction between those individuals at high risk for TB and those at high risk for HIV: IV-drug users, homeless and inner-city minorities (Torres *et al.*, 1990). Third, TB is the most common HIV-related complication world-wide (Narain *et al.*, 1992). Fourth, HIV infected individuals are not only at a greater risk for acquiring TB (as much as 500 times the normal chance in HIV-negative individuals), but reactivation of latent TB infection is greatly increased due to the fact that the very cells that hold the latent TB in check (the CD4⁺ T lymphocytes) are precisely the cells that are rendered dysfunctional in HIV-infected individuals (Bryt *et al.*, 1994). Fifth, TB decreases the number of CD4⁺ T cells thereby interfering with the best predictor of AIDS

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survivability (Katz *et al.*, 1979). This is important, because the CD4⁺ T cells are the cells that not only become infected with HIV, but orchestrate the immune response against both TB and HIV, as well as other pathogens.

Treatment of TB infection, in the case of non-resistant strains, is well developed. The most common treatment regime is a combination of isoniazid, rifampin and pyrazinamide for 2 months, followed by isoniazid and rifampin for at least 4–7 months until all the bacteria have been completely cleared (Gittler, 1994; WHO, 1983). If adhered to properly, the cure rate is almost 100%. There are many problems, however, with administering a treatment regime that has such a long duration, the main one being compliance. In many cases, patients do not complete therapy, and this not only causes a rebound in the individual's TB, but also aids in the formation of mutant strains that are drug resistant (Iseman and Madsen, 1989; Vareldzis *et al.*, 1989). These “first-line” drugs are the best defense against TB, and in their loss of use, due to multi-drug resistance to some or all of the drugs, treatment becomes difficult, costly, or even impossible with a cure rate of only 60%. Treatment of HIV is not so clear cut, as is true of most viral infections. Presently multiple antiviral agents administered in combination serve only to slow the progression to AIDS, but as yet, there is no cure.

In this study we focus mainly on the effect that TB has on HIV infection, but also comment on how the model may explain the resurgence of TB due to HIV. In particular, we wish to explore the impact of *Mycobacterium tuberculosis*, the bacteria that causes TB, on the HIV-infected immune system.

To date there exists only one other mathematical model of the immune system interacting with mycobacteria (Antia *et al.*, 1994; Antia *et al.*, 1996) and none exist that explore the interaction between virus and bacteria, and in particular, with TB and HIV. The model by Antia *et al.* explores persistence at low densities until the dormant stage ends, when large amounts of *M. tuberculosis* are present. They base their findings on the Hayflick limit, i.e., the role of thymus input into the system. As we will model the adult population, it is well known the thymus involutes in adults, especially during HIV infection, and therefore may play a very minor role in the dynamics we model.

In Section 2 we describe the immunology of both diseases and present a mathematical model encompassing the interaction of the key players in these disease dynamics. Then we create a model describing these events, based on knowledge from previous experience with HIV-immune models. In Section 3 we analyze the model and give

numerical results. Section 4 explores the role of treatment. We study the dynamics, as well as the effects of administering both a complete and an incomplete treatment course against TB. Section 5 concludes; we find that the presence of TB worsens the clinical picture of HIV-infected individuals, and that HIV also activates the TB infection. Treatment of the TB infection can improve survivability of the co-infected patient.

2. THE MODELING PROCESS

Immune System

The human immune system has two main responses to the introduction of foreign antigen into the body: a cellular-mediated response and a humoral response (antibodies). Both TB and HIV primarily affect a cellular-mediated immune response, so a brief description of this process follows. Antigen-presenting cells (APC) called macrophages engulf the pathogen(s) (phagocytosis) and present the processed antigens to the CD4⁺ T cells. These T cells elicit an immune response by issuing commands to the other players in the immune system's cellular response, namely helper CD4⁺ T cells, macrophages and cytotoxic CD8⁺ T cells (the CD4 and CD8 are distinguishing cell surface protein markers). The CD4⁺ T cells stimulate macrophages to engulf foreign particles and kill them, and CD8⁺ T cells are cytotoxic T cells (CTLs) that can kill infected host cells directly. This is a primary immune response. If the immune system sees this pathogen again, it issues a secondary immune response that is usually much faster and more efficient than the first.

The infections by HIV and by TB are similar in their pathogenesis, in that they are both microbes that seem able to evade the immune-system's cellular responses; they are, however, very different in their biology.

Tuberculosis

In the case of infection with the bacterium *M. tuberculosis*, the disease is invasive, is locally toxic, and causes a hypersensitive reaction. The immune response takes anywhere from 48 hours to 14 days to appear. As soon as 24 hours after exposure, however, there is an intense increase in both the T cell and macrophage populations. The cell-mediated immune response to infection with TB occurs in two ways. First, the immune system cells damage the walls of the bacteria; and, second, there is phagocytosis and digestion by macrophages. Tuberculosis is a unique disease, in that 90% of persons infected do not progress to active disease; however, 5% progress

rapidly to active infection and die without treatment, while 5% progress slowly over their lifetime (cf. Blower *et al.*, 1996).

In these 90% of primary infections with *M. tuberculosis*, the immune system is able to hold the infection in check, and the pathogen becomes dormant (or latent). This occurs within six weeks of infection. The problem is, then, that *M. tuberculosis* can be “resistant” to the macrophage killing. In this case, the infection activates and unless treatment is administered the patient will die. There is also bacterial killing by CTLs that may keep the TB latent (Tanaka *et al.*, 1995). Clinical TB is characterized by severe lung damage. This occurs when both the bacteria and the immune cells aggregate to the lungs and nodular masses are formed called *granulomata*. Lung damage results that leads to cavitation and spread of *M. tuberculosis*. T cells play a dual role in this aggregation: they cause the accumulation of immune cells to these areas, and also stimulate the macrophages to phagocytize. If the TB is in the active form, this process continues, leading to the eventual collapse of the lung environment.

HIV-1

In the case of infection with the virus HIV-1, the disease is invasive and potentially toxic to cells that are CD4 positive cells such as helper T cells and macrophages (the CD4 surface protein is necessary for the binding of virus to cell). These cells are subsets of white blood cells. The hallmark of HIV infection is the marked decline in the population of CD4⁺ T cells per mm³ of blood, together with a rise in viral titer. Normal amounts of CD4⁺ T cells in healthy individuals range from 1000–2000 per mm³ of blood. The different stages of HIV infection leading toward Acquired Immune Deficiency Syndrome (AIDS) are classified on the basis of T cell counts below the 1000 per mm³ level (e.g., Walter Reed, 1989). At the same time, however, HIV must use these CD4⁺ T cells to reproduce, something that differs greatly from the *M. tuberculosis* which can reproduce independently. Once a virus infects a cell, the cell can remain quiescent or it can be activated to proliferate; however, while the cell is replicating, it also produces new virus particles. We will count the average number of viral particles a cell produces during its lifespan (known as viral burst size) and denote it by N . This production may occur slowly or very rapidly, but eventually the host cell is destroyed. There is evidence to believe that this is one of the reasons for the eventual collapse of the immune system in AIDS (Conner *et al.*, 1993; Dimitrov *et al.*, 1993; Perelson *et al.*, 1993); although, other ideas such as antigenic diversity (Nowak *et al.*, 1990; Nowak and May, 1991; Nowak and Bangham,

1996); high turnover rates of infected T cells (Perelson *et al.*, 1996); and, the “sink” model (Ho *et al.*, 1995) have also been suggested as plausible explanations. We (Kirschner and Webb, 1996, 1997a, 1997b, 1997c) hypothesize that it is the viral load in the lymph system together with long-living, latently-infected T cells as well as altered circulation patterns that actually accounts for progression to AIDS.

The Model

In this model we incorporate the above mentioned mechanisms to describe the immune system-bacteria-virus interactions. To begin modeling these interactions, let the compartment the dynamics occur in be the lymph tissue. We do so as all HIV model values are taken from peripheral blood data, but it is generally assumed that the lymph and periphery are in parallel (Haase *et al.*, 1996). Infection with *M. tuberculosis* is also known to occur in lymph tissue. A more detailed model encompassing separate compartments (e.g., blood and lung) for these dynamics is needed to better elaborate the results obtained here. We define four populations: $T(t)$ represents the *armed* CD4⁺ and CD8⁺ T cell populations at time t in days; $M(t)$ represents the macrophage population; $V(t)$ represents the HIV population; and, $T_b(t)$ represents the *M. tuberculosis* population. We assume the populations are large enough in size to be modeled deterministically, and that the dynamics can be represented by the following ordinary differential equations:

$$\begin{aligned} \frac{dT(t)}{dt} &= s_T(t) - \mu_T T(t) + r_T T(t) \left[\frac{(V(t) + T_b(t))}{C + (V(t) + T_b(t))} \right] \\ &\quad - k_1 V(t) T(t) \end{aligned} \quad (1)$$

= source/death/immune response growth/
infection & loss

$$\begin{aligned} \frac{dM(t)}{dt} &= \mu_M [M_0 - M(t)] - k_2 M(t) V(t) \\ &\quad + r_M^2 M(t) V(t) + r_M^1 M(t) T_b(t) \end{aligned} \quad (2)$$

= source/death/infection & loss/stimulation/
recruitment

$$\begin{aligned} \frac{dV(t)}{dt} &= V(t) [N_1 k_1 T(t) + N_2 g_v M(t)] \\ &\quad - V(t) [k_3 T(t) + k_4 M(t)] - \mu_V V(t) \end{aligned} \quad (3)$$

= source (T and M)/immune response/
death

$$\begin{aligned} \frac{dT_b(t)}{dt} &= r_{T_b} T_b(t)(K - T_b(t)) - \mu_{T_b} T_b(t) \\ &\quad - T_b(t)[k_5 T(t) + k_6 M(t)] \end{aligned} \quad (4)$$

= logistic growth/death/immune response.

The Model is explained as follows. Equation (1) represents the change in the CD4⁺ and CD8⁺ T cell populations over time. The first term is the source term of new T cells. This is modeled, not as a parameter, but as a function of time, and it is documented that the precursors to these T cells are affected by the presence of HIV (cf. Kirschner *et al.*, 1998). Thus, $s_T(t)$ is a decreasing function of time (we discuss this further in the next section). This is followed by a natural loss term, because T cells have a finite life span. Next is growth of T cells, presented in this form to represent expansion by the presence of antigen.¹ Since T cells do not grow without bound, we choose a saturating growth term of Michaelis-Menten type, where r_T is the maximal growth-response rate. (Similar ideas were used in Perelson *et al.*, 1993; Kirschner and Webb, 1996, 1997a, 1997b.) The final term is a loss of T cells from the uninfected class, due to infection by HIV.

Equation (2) represents the change in the macrophage population over time. The reasons for including the macrophage population are many. Macrophages survive once infected with HIV, and slowly bud new virus particles (Orenstein *et al.*, 1997). They, therefore, play a role as a viral source referred to as a reservoir. Also infected macrophages can infect CD4⁺ T cells through presentation of antigen (Lewis and McGee, 1992). Finally, as mentioned above, macrophages play a major role in TB pathogenesis. The first term of Eq. (2) represents the birth-death process for macrophages. The next term represents the loss of uninfected macrophages due to infection by HIV. The final term is a recruitment of new macrophages to the infection site, governed by the presence of pathogen.

Equation (3) represents the change in the HIV population over time. The first two terms represent source terms for the virus population. This follows from (1) and (2) when the T cells and macrophages, respectively, begin to produce new virus after becoming infected with HIV. Macrophage infection, however, is not well understood, and so we allow for the possibility of a different production

rate of virus (g_v) than infection (k_2). Once a cell becomes infected with HIV, we assume that that cell produces on the average N_i (N_1 for T cells and N_2 for macrophages) new viral particles (Perelson, 1989; Perelson *et al.*, 1993). The following two terms of (3) are immune clearance terms—CD8⁺ T cells and macrophages clear or kill virus and infected cells. The final term of (3) is a natural death/clearance term for HIV, as virus have a finite life span.

Equation (4) represents the change in the *M. tuberculosis* population over time. The first term is a logistic growth term, that represents bacterial growth. This is followed by a natural death term and immune clearance/killing terms, with rate constants k_5 and k_6 , respectively.

Parameter Values

Here, our focus will be to model the latent stage disease dynamics of HIV as this is the longest phase during progression. Although this phase is by no means dormant (Ho *et al.*, 1995; Perelson *et al.*, 1996), it is the phase during which the T cells and virus are present in quasi-steady state amounts and the most likely stage where TB is contracted or reactivated (if latent). We present our choices for parameter values, summarized in Table I, and postpone the discussion of the sensitivity analysis until the next section.

We first estimate the source of new T cells. The thymus, lymph nodes and bone marrow all are sources of T cells, as the CD4⁺ T cells are affected by HIV, and it is conjectured that the other immune cells are as well. A functional source, represented by $s_T(t)$, was chosen as a sigmoidal function of viral load that decreases by half as the viral load increases. This function has been shown to be a good first approximation [cf. Kirschner *et al.*, 1998], and is given by $s_T(t) = (0.5s_1 + 0.5s_2/(c_1 + V(t)))$, where s is the constant source rate in the absence of infection. The natural lifespan of uninfected T cells is not well known, although it is known to be variable and possibly large (McLean and Michie, 1995; Tough and Sprent, 1995). Here, we choose a value of $\mu_T = 0.007$ that corresponds to lifespan of 143 days (Kirschner and Webb, 1996, 1997a, 1997b, 1997c; Perelson *et al.*, 1993). We chose this value so that the steady-state concentration of T cells in the absence of infection would be 1500 per mm³ (i.e., s/μ_T). We assume that activated T cells divide every 12–18 hours. Therefore, the growth rate of an activated T cell is approximately 1 per day. This growth rate must be multiplied by the fraction of T cells. This is probably on the order of 1%, hence we choose $r_T = 0.02$. Infection of CD4⁺ T cells by virus has been modeled extensively (Ho *et al.*, 1995; Perelson *et al.*,

¹ This is usually accomplished through antigen presentation by APC; however, the presentation is certainly correlated with the amount of antigen present (i.e., the concept of “antigen threshold”), so we model the pathogens as stimulating the immune response.

TABLE I

Variables and Parameters

Dependent variables		Values
$T(t)$	= Uninfected CD4 ⁺ and CD8 ⁺ T cell population	1500 mm ⁻³
$M(t)$	= Macrophage population	85 mm ⁻³
$V(t)$	= HIV population	0.001 mm ⁻³
$T_b(t)$	= <i>M. tuberculosis</i> (<i>M. tuberculosis</i>) population	1.0 mm ⁻³
Parameters and constants		Values
c_1	= half saturation constant for growth	1 mm ⁻³
s_2	= rate of change in T-cell supply	10 d ⁻¹
s_1	= source of new T cells in absence of infection	10 d ⁻¹ mm ⁻¹
μ_T	= death rate of uninfected T cell population	0.007 d ⁻¹
μ_M	= death rate of macrophage population	0.003 d ⁻¹
μ_V	= death rate of HIV population	2.4 d ⁻¹
μ_{T_b}	= death rate of <i>M. tuberculosis</i> population	0.5 d ⁻¹
k_1	= rate CD4 ⁺ T cells becomes infected by free virus	2.4×10^{-5} mm ³ d ⁻¹
k_2	= rate macrophages become infected by free virus	2.0×10^{-6} mm d ⁻¹
g_v	= rate macrophages produce virus	1.0×10^{-6} mm ³ d ⁻¹
k_3	= rate CD8 ⁺ T cells kill virus	7.4×10^{-4} mm ³ d ⁻¹
k_4	= rate macrophages kill virus	7.4×10^{-4} mm ³ d ⁻¹
k_5	= rate T cells clear <i>M. tuberculosis</i> .	0.5 mm ³ d ⁻¹
k_6	= rate macrophages clear <i>M. tuberculosis</i> .	0.5 mm ³ d ⁻¹
r_T	= maximal proliferation of the T-cell population	0.02 d ⁻¹
r_{T_b}	= maximal proliferation of the <i>M. tuberculosis</i> population	1.0 d ⁻¹
r_M	= recruitment rate of macrophage population	2.5×10^{-7} d ⁻¹
N_1	= number of free virus produced by infected T cells	100 – 1000
N_2	= number of free viruses produced by macrophages	100 – 1000
C	= half saturation constant of the proliferation process	1000 mm ⁻³
K	= carrying capacity for the <i>M. tuberculosis</i> population	1000 mm ⁻³
Mo	= equilibrium value for the macrophage population	100 mm ⁻³

Note. A list of all parameters used in the Model (1)–(4). The discussion of the choices for these values is presented in Section 2.

1996). Here we use a value from the above mentioned studies of $k_1 = 2.4 \times 10^{-5}$.

Modeling macrophage interactions was first done in (Kirschner and Perelson, 1995). Here, we use a similar idea. The first term represents the equilibrium of the macrophage birth-death process. According to (Meltzer *et al.*, 1990), there are approximately 6000/mm³ white blood cells in a healthy human. It is estimated that between 1–6% of these are in the macrophage population (Lewis and McGee, 1992). Approximately 10% of these cells are CD4⁺ (Pauza, 1989), hence an initial condition for uninfected macrophages would be $M(0) = 100$ mm³. Macrophages can have a long life span, so we take the death rate of uninfected cells to be 3×10^{-3} per day (Delemarre *et al.*, 1990). Finally for recruitment of macrophages via virus and bacteria, there is no data to estimate r_M^1 and r_M^2 , so we choose them to be small (i.e., on the order of 10^{-7}).

Mathematical modeling of viral turnover rates has recently been done (Ho *et al.*, 1995; Perelson *et al.*, 1996).

It was estimated that HIV have a minimal half-life of approximately 6 hours, so we choose $\mu_V = 0.5$. As infected T cells and macrophages produce virus, we use N_i ($i = 1$ for T cell production, $i = 2$ for macrophage production) to count the average number of virus particles produced. The values for N_i have been estimated between 100–1000 (Haase *et al.*, 1996). Since production of virus via infected macrophages is not well studied, we assume that the production rate of new viral particles is half that of the infection rate. Finally, there is clearance of virus via immune-system cells. These rates are not well studied, and so we choose them to be on the same order as infection rates.

The growth and death of *M. tuberculosis* are well studied processes. Values for doubling times etc., can be found directly from data (cf. Pfuente and Rander 1996; Youmans, 1979). Other values, such as rates T cells and macrophages kill *M. tuberculosis*, were estimated and analyzed through numerical simulations (next section).

3. ANALYSIS AND NUMERICAL RESULTS

Analysis

To begin analysis of this system, we examine equilibria of (1)–(4). There are several steady states for this system, some that are not biologically feasible (i.e., negative). It is certainly true that both the viral and bacterial populations can be zero. Let an over-bar denote a steady-state value. Hence, there are at least 3 steady states: an *uninfected steady state*, $SS_U = (\bar{T}_1, \bar{M}_1, 0, 0)$, a *HIV-infected steady state*, $SS_V = (\bar{T}_2, \bar{M}_2, \bar{V}_2, 0)$ and a *TB-infected steady state* $SS_{T_b} = (\bar{T}_3, \bar{M}_3, 0, \bar{T}_{b_3})$. There is also at least one other steady state, a *co-infected steady state*, where none of the populations are zero, namely $SS_I = (\bar{T}_4, \bar{M}_4, \bar{V}_4, \bar{T}_{b_4})$. It is easily verified that the non-negative orthant, $\{x \in \mathbb{R}^4 \mid x \geq 0\}$, is a positively invariant region. Hence, no populations becomes negative or grows without bound. For ease of analysis here, we will assume that $s_T(t) = s_T$ is constant.

Case (1). $SS_U = (\bar{T}_1, \bar{M}_1, 0, 0)$: The uninfected steady state. In this case, both the V and T_b populations are zero, and the steady-state values are:

$$\bar{T}_1 = \frac{s_T}{\mu_T} \quad \text{and} \quad \bar{M}_1 = M_0. \quad (5)$$

If we examine the roots of the characteristic equation from the Jacobean matrix of the system (1–4), the four eigenvalues are $\lambda_1 = -\mu_T$, $\lambda_2 = -\mu_M$, and $\lambda_3 = -(\mu_{T_b} + k_5 \bar{T}_1 + k_6 \bar{M}_1 - K r_{T_b})$, $\lambda_4 = -(k_3 \bar{T}_1 + k_4 \bar{M}_1 + \mu_V - N_1 k_1 \bar{T}_1 - N_2 g_v \bar{M}_1)$. For this uninfected steady state to be locally asymptotically stable, we require the eigenvalues to each be negative. This is true only if

$$K < \frac{1}{r_{T_b}} (\mu_{T_b} + k_5 \bar{T}_1 + k_6 \bar{M}_1) = K_{crit} \quad (6a)$$

and

$$N_1 < \frac{1}{k_1 \bar{T}_1} (k_3 \bar{T}_1 + k_4 \bar{M}_1 + \mu_V - N_2 g_v \bar{M}_1) = N_1^{crit} \quad (6b)$$

arising as conditions from the eigenvalues λ_3 and λ_4 . If either of these conditions are invalid, the steady state becomes a locally unstable saddle point. We represent the bifurcation values, K_{crit} and N_1^{crit} in terms of K and N_1 because these parameters play an important role in the system (note that a similar bifurcation scheme applies

for N_2 , but we discuss only N_1 for brevity). K governs the size of the bacteria population, and N_1 governs T-cell viral production. When these conditions are invalidated, another steady states accept stability.² Therefore, they are *Transcritical Bifurcations* (TC). A two-parameter bifurcation diagram is shown in Fig. 1. If any of the parameters involved in the eigenvalues λ_3 and λ_4 are varied, they each lead to a similar transcritical bifurcation scheme from SS_U to either SS_V or SS_{T_b} .

Case (2). $SS_V = (\bar{T}_2, \bar{M}_2, \bar{V}_2, 0)$: The virally-infected steady state. In this case, the T_b population is zero. This reduces to solving

$$s_T = \bar{T}_2 \left[\mu_T - r_T \left[\frac{\bar{V}_2}{C + \bar{V}_2} \right] + k_1 \bar{V}_2 \right], \quad (7)$$

$$\mu_M \bar{M}_0 = \bar{M}_2 [\mu_M - r_M^2 \bar{V}_2 + k_2 \bar{V}_2], \quad (8)$$

$$\bar{T}_2 = \frac{\mu_V - N_2 g_v \bar{M}_2 + k_4 \bar{M}_2}{N_1 k_1 - k_3}. \quad (9)$$

If we solve these equations for the steady state values, we find

$$\bar{M}_2 = \frac{\mu_V + k_3 \bar{T}_2 - k_1 N_1 \bar{T}_2}{N_2 g_v - k_4} \quad [\text{from Eq. (3)}], \quad (10)$$

$$\bar{T}_2 = \frac{s_T}{\mu_T + k_1 \bar{V}_2 - (r_T \bar{V}_2)/(C + \bar{V}_2)} \quad [\text{from Eq. (1)}]. \quad (11)$$

Since the \bar{T}_2 steady-state value depends only on \bar{V}_2 , and the \bar{M}_2 steady-state value depends on \bar{T}_2 (that only depends on \bar{V}_2) it remains to find the value for the \bar{V}_2 steady state. This entails solving a cubic in \bar{V}_2 that yields at most three positive steady-state values, however our bifurcation analysis shows that at most two positive and stable at any time, t (Regions I&II). In the numerical output for this steady-state, there are oscillations (discussed below). It is the cubic in solving for \bar{V}_2 that introduces imaginary values into the eigenvalues for certain parameter regimes. Because we can not find the steady states explicitly, it is computationally difficult to find the eigenvalues. However, one eigenvalue can be read from the Jacobian matrix directly, and it is identical to the eigenvalue λ_3 (6a) for steady-state one, SS_U . There are other parameter restrictions that arise to insure a positive steady state. For the \bar{M}_2 steady-state value, the

² These parameters were studied using the Xppaut3.0 (Ermentrout, 1992) version of AUTO (Doedel, 1981).

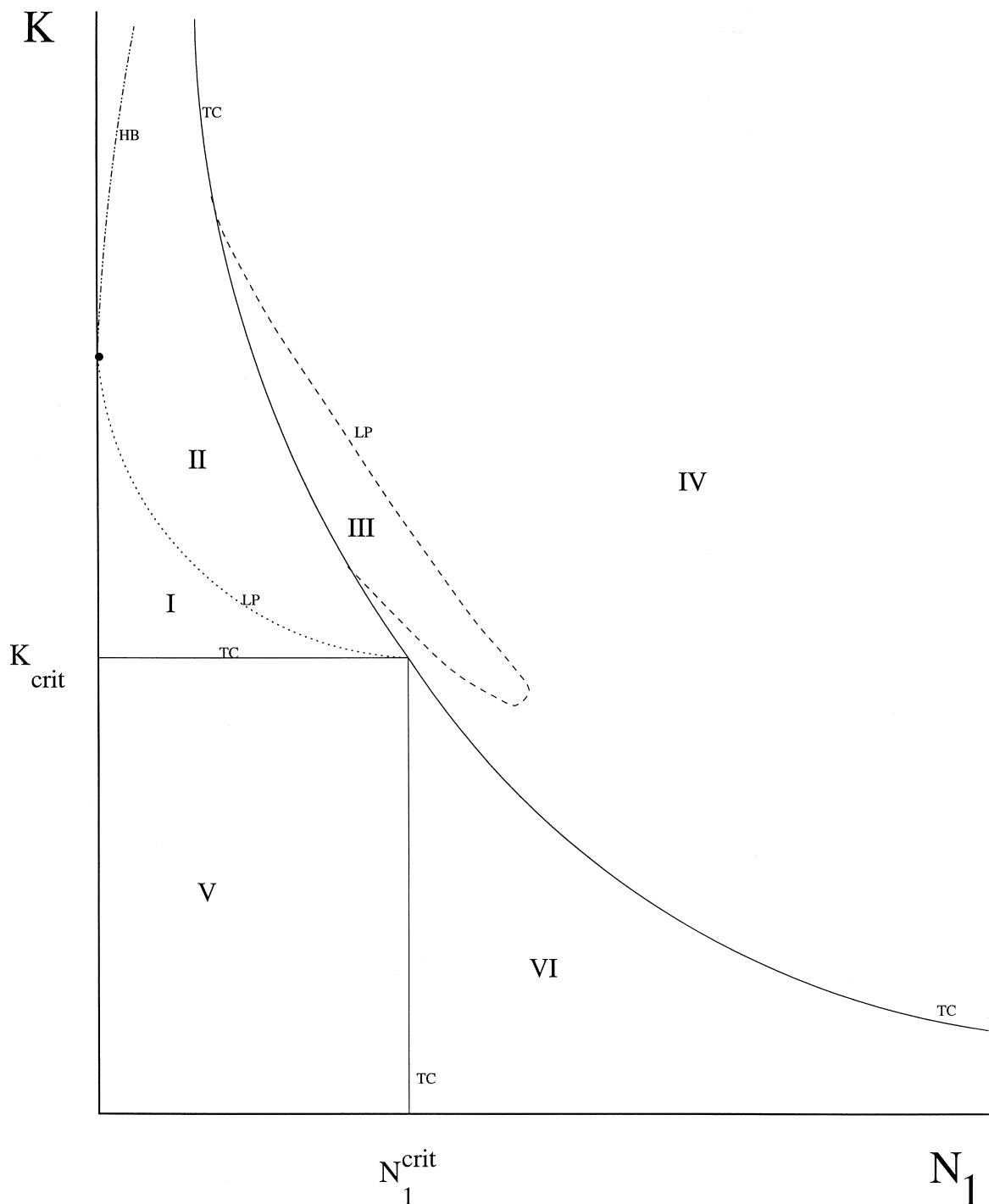


FIG. 1. A 2-parameter bifurcation diagram for K vs. N_1 revealing the steady-state stability. The solid curves indicate a transcritical bifurcation (TC), the dotted and dashed curves indicate limit points (LP), and the dot-dash curve indicates a Hopf bifurcation (HB). The behavior for the system (1)–(4) described in the different regions are as follows. **Region I:** The TB-infected steady state, SS_{T_b} , is a locally stable, improper node; **Region II:** Bistability occurs between the TB-infected and co-infected steady states, SS_{T_b} (improper node) and SS_I (spiral node) are both locally stable; **Region III:** In this region bistability occurs between two different co-infected steady states, SS_I^1 and SS_I^2 , both locally stable spiral nodes; **Region IV:** The co-infected steady state, SS_I , is a locally stable spiral node; **Region V:** The uninfected steady state, SS_U , is a locally stable improper node; and, **Region VI:** The HIV-infected steady state, SS_V , is a locally stable, spiral node.

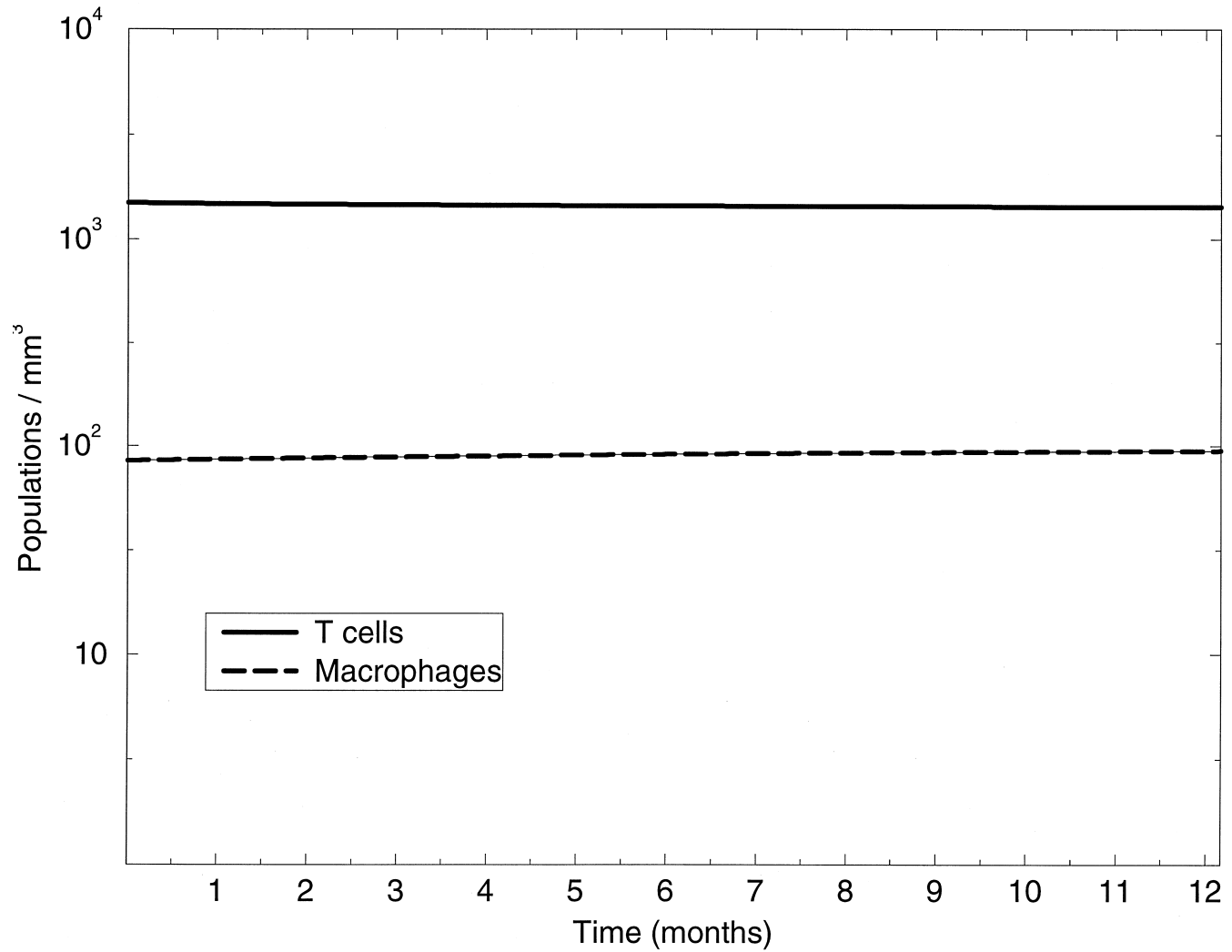


FIG. 2. The uninfected steady state. This is the numerical solution to Eqs. (1)–(4) with the populations HIV and *M. tuberculosis* set to zero. Values are presented on a log-linear plot. Table II gives the corresponding numerical steady-state values. Both T cells, T , and Macrophages, M , converge to a positive, steady-state value. All parameters come from Table I. We begin with initial conditions slightly off from the steady-state value to exhibit the nature of the convergence.

denominator must be positive when the numerator is positive (i.e., $\mu_V > \bar{T}_2(N_1 k_1 - k_3)$ and $N_2 g_v > k_4$). And, if the denominator expression is negative, then the numerator must be negative as well (i.e., $\mu_V < \bar{T}_2(N_1 k_1 - k_3)$ and $N_2 g_v < k_4$). If we examine λ_4 (6b) above, it is clear these inequalities play a key role in the sign of the eigenvalue. Notice, that if any of these conditions are invalid, and there is not a positive steady-state value for all the populations of SS_V , then the SS_U is locally stable. However, when these conditions are all satisfied, and SS_V emerges in the positive cone, then SS_U loses stability via a TC bifurcation and either SS_{T_b} or SS_V becomes stable. Hence, there would be a transcritical bifurcation. The bifurcation diagram in Fig. 1 validates this.

Case (3). $SS_{T_b} = (\bar{T}_3, \bar{M}_3, 0, \bar{T}_{b_3})$: The TB-infected steady state. In this case, the V population is zero. The system reduces in steady state to:

$$s_T = \bar{T}_3 \left[\mu_T - r_T \left[\frac{\bar{T}_{b_3}}{C + \bar{T}_{b_3}} \right] \right], \quad (12)$$

$$\mu_M M_o = \bar{M}_3 [\mu_M - r_M^1 \bar{T}_{b_3}], \quad (13)$$

where the value of \bar{T}_{b_3} is determined by \bar{T}_3 and \bar{M}_3 , namely

$$T_{b_3} = \frac{1}{r_{T_b}} [r_{T_b} K - \mu_{T_b} - k_5 \bar{T}_3 - k_6 \bar{M}_3]. \quad (14)$$

There is only one obtainable eigenvalue in this case as well, and it corresponds to λ_4 , (6b), for steady state one, SS_U . Once again, there are parameter restrictions that ensure stability, namely $K > 1/r_{T_b}(\mu_{T_b} + k_5\bar{T} + k_6\bar{M})$. This corresponds to the eigenvalue λ_3 , (6a), which is needed for stability of the uninfected steady-state, SS_U .

There is an exchange of stability at this point; a transcritical bifurcation occurs.

The values of \bar{T}_3 and \bar{M}_3 , are obtained by solving a cubic equation in \bar{M}_3 . Once a value of \bar{M}_3 is determined, the values of \bar{T}_3 and \bar{V}_3 can be determined from (12)–(13). Again, there is the possibility for at most three

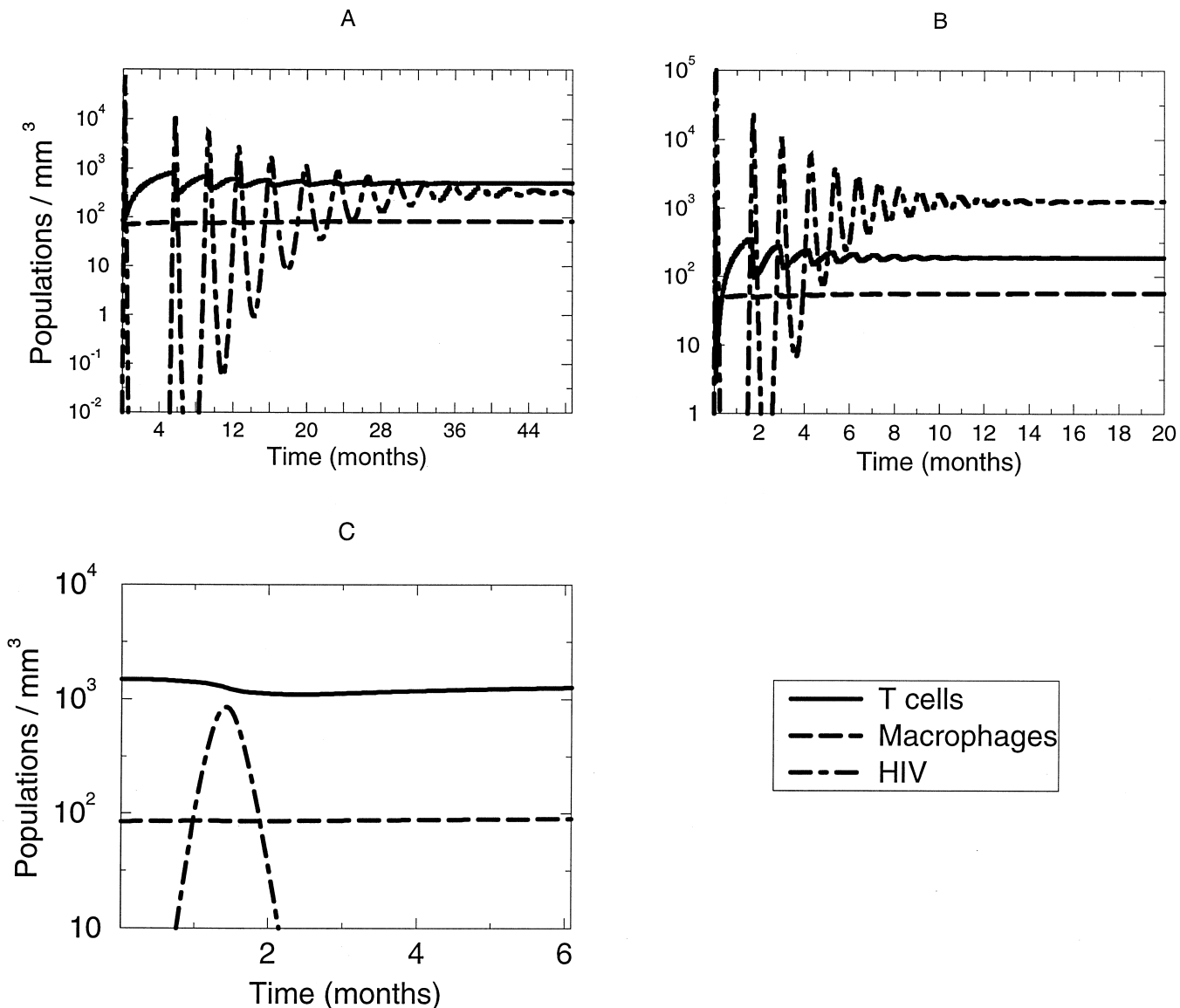


FIG. 3. The HIV-infected steady state. This is the numerical solution to Eqs. (1)–(4) with one of the pathogen populations, HIV, and the other, *M. tuberculosis*, set to zero. Values are presented on a log-linear plot. Table II gives the corresponding numerical steady-state values. The populations of T cells, macrophages and virus, T , M and V respectively, progress to positive steady-state values. All parameters come from Table I. Three cases are shown with different assumptions for the viral production parameter, N_1 . **Panel A** shows the initial transient and the quasi-steady state stage (here, $N_1 = 200 > N_1^{crit}$); **Panel B** shows the immune system crash, where the viral population is very large, and T cell population is low here the viral productions are $N_1 = 500 > N_1^{crit}$; **Panel C** shows the immune system recovering and pushing the viral population to zero (here the viral productions are $N_1 = 50 < N_1^{crit}$).

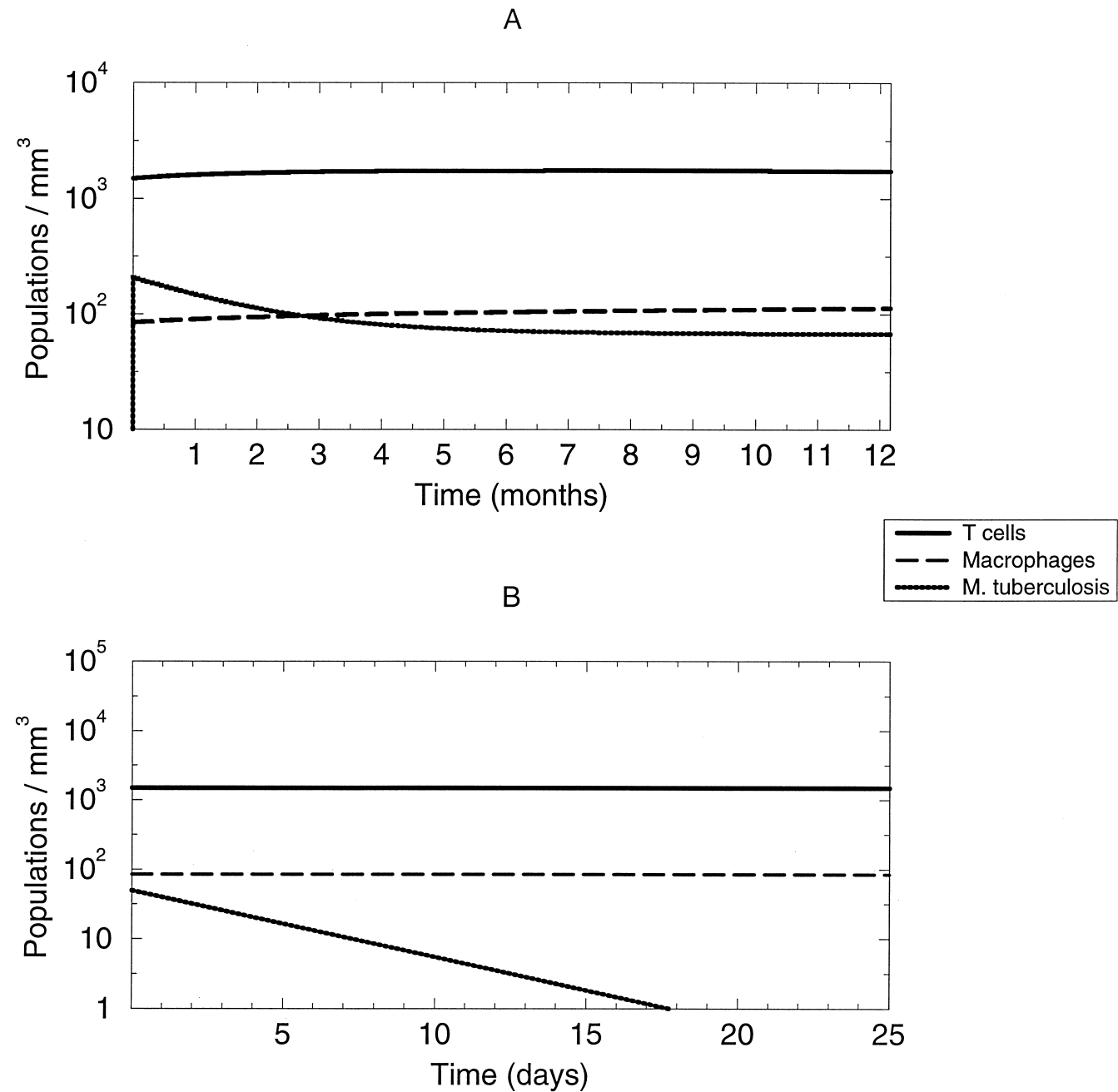


FIG. 4. The TB-infected steady state. This is the numerical solution to Eqs. (1)–(4) showing the TB infection, with the HIV population set to zero. The populations of T cells, macrophages and *M. tuberculosis*, T , M and T_b respectively, progress to positive steady-state values (**Panel A**, $K = 1000 > K_{crit}$), or the infection can be cleared (**Panel B**, $K = 500 < K_{crit}$). All parameters come from Table I. Values are presented on a log-linear plot. Table II gives the corresponding numerical steady-state values.

positive steady states, and from our bifurcation analysis only one is positive and stable at any time, t (Region VI).

Case (4). $SS_I = (\bar{T}_4, \bar{M}_4, \bar{V}_4, \bar{T}_{b_4})$. The co-infected steady state. Here, both pathogens, TB and HIV, are present. This reduces to solving a two equation algebraic

system in M and V and where, the \bar{T}_4 and \bar{T}_{b_4} populations are given as:

$$\bar{T}_4 = \frac{\mu_V - N_2 g_v \bar{M}_4 + k_4 \bar{M}_4}{N_1 k_1 - k_3}$$

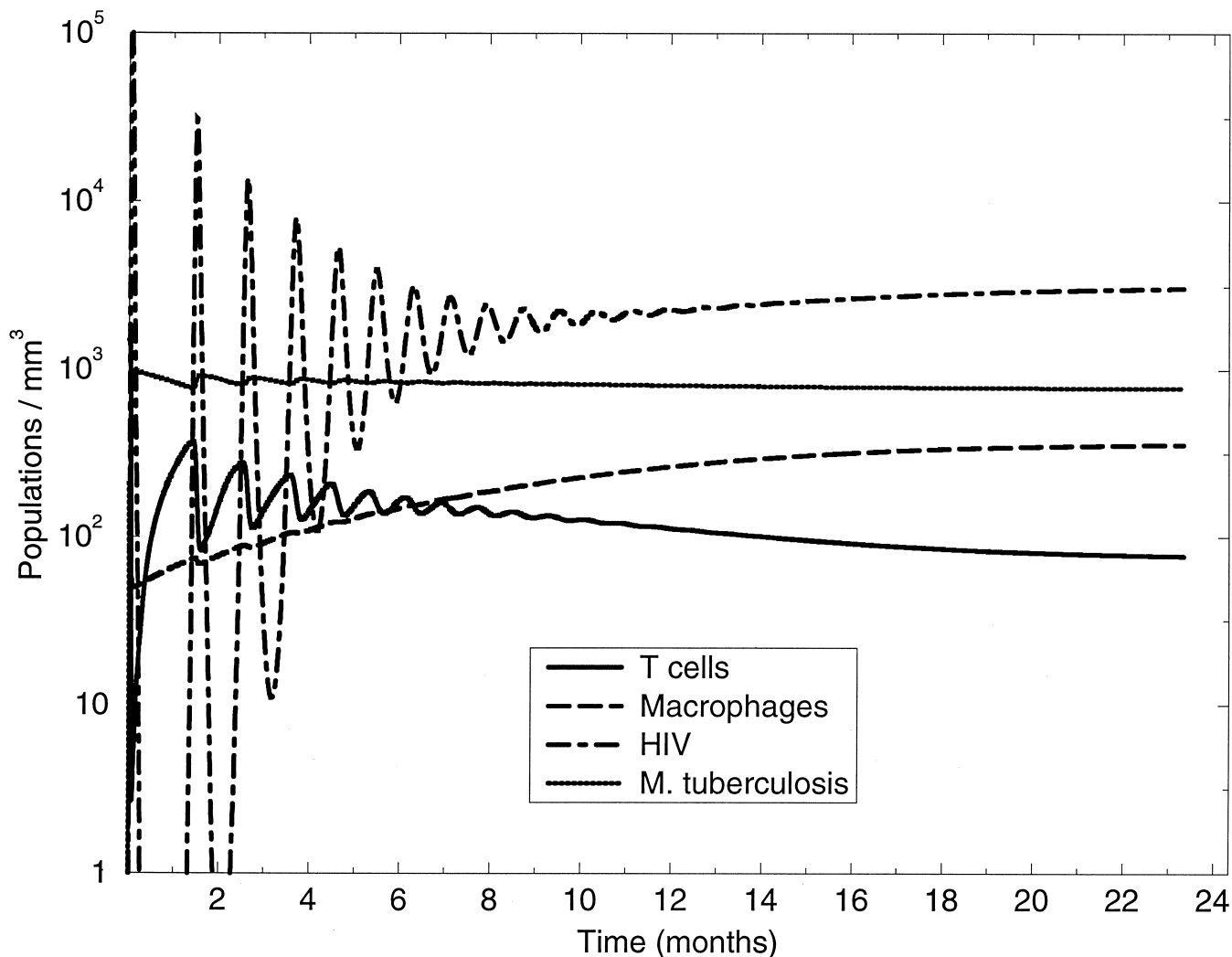


FIG. 5. The co-infected steady state. This is the numerical solution to Eqs. (1)–(4) with both the pathogens, HIV and *M. tuberculosis*, present. Values are presented on a log-linear plot. Table II gives the corresponding numerical steady-state values. All populations progress to a positive steady-state value. All parameters come from Table I.

TABLE II

	Pop	Uninfected	Virus-infected	Virus-infected	TB-infected	Co-infected
	Size	SS_U	SS_{V-H^*}	SS_{V-L}	SS_{T_b}	SS_I
T cell count	$T(t)$	1428.57	191.11	502.79	1738.5	74.69
Macrophage count	$\bar{M}(t)$	100.00	58.25	84.19	127.42	365.97
HIV titer	$\bar{V}(t)$	0.00	1264.7	331.32	0.	3163.4
<i>M. Tuberculosis</i>	$\bar{T}_b(t)$	0.00	0.00	0.00	66.55	779.17
Parameter change	PC	$N_1 = 50, K = 500$		$N_1 = 200$	$N_1 = 50$	

Note. These are data generated by the model using the parameter values in Table I. All units are per mm^3 . The equilibria values are given for each of the steady-states. PC = change in parameter value used to generate if different than in Table I. These correspond to the ending numerical values in Figs. 2–5.

*H, implies high viral infection, L, implies a low viral infection.

and

$$\bar{T}_{b_4} = \frac{1}{r_{T_b}} [r_{T_b} K - \mu_{T_b} - k_5 \bar{T}_4 - k_6 \bar{M}_4]. \quad (15)$$

Quite interesting is that the same parameter restrictions arise here for positivity of this steady state. The eigenvalue problem is intractable analytically; however, there are at most two positive steady states, SS_I^1 and SS_I^2 . Bifurcation analysis reveals that there is bistability between them for a small parameter region (region III of Fig. 1); and, they then merge into SS_I , and this steady state is stable for the largest parameter space (region IV of Fig. 1).

Numerics

We explore the analytical results further through numerical simulations. First, the system should make sense in the absence of pathogens. Figure 2 depicts the numerical solutions for SS_U , namely the uninfected steady-state. Both immune-cell populations are at positive equilibria. Next, each pathogen should be able to sustain its own infection. Infection by HIV (SS_I) can elicit multiple responses. Figure 3a shows the initial transient and steady-state stage of disease. Figure 3b shows the crash to AIDS (i.e., a very low T cell count, and a very high viral load). The T cells drop by 7.5- and 2.9-fold, respectively; and the corresponding macrophage population drop by

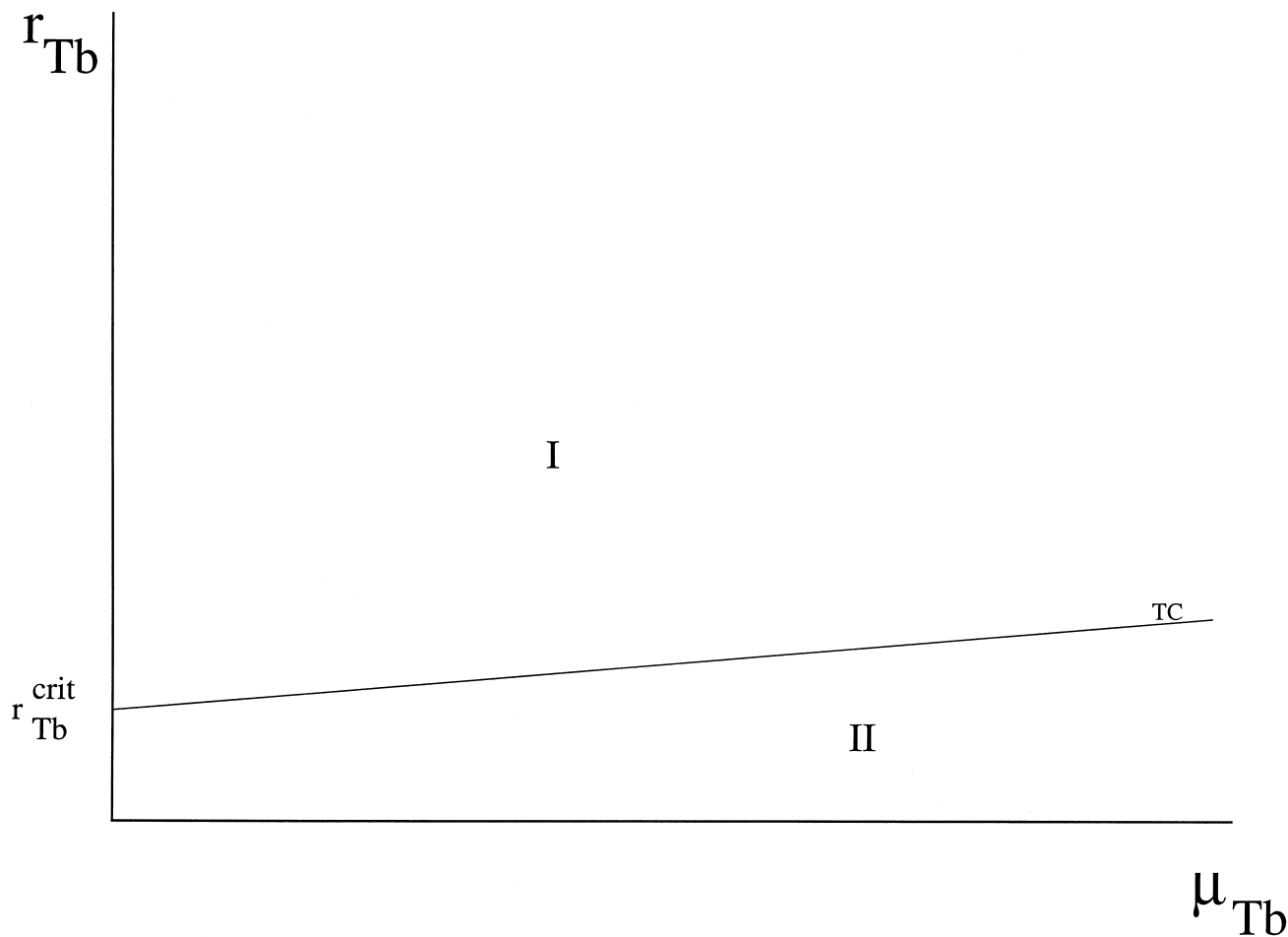


FIG. 6. Bifurcation diagram of treatment parameters r_{T_b} and μ_{T_b} . These are the key parameters that control the effects of treatment of TB infection. There is a smaller region for clearance of *M. tuberculosis* than for maintaining co-infection. In region I the co-infected steady state is stable, and the TB-treatment is not effective. In region II the viral-infected steady state is stable, and the treatment successfully cleared the *M. tuberculosis* infection. TC refers to a transcritical bifurcation.

almost half. The viral loads are between 10^6 – 10^7 /ml. Figure 3c reveals the possibility for recovery once infected. Note that these different phenomena are obtained by varying the viral production, N_1 .

Infection by *M. tuberculosis* (SS_{T_b}) evokes an increase in the immune response by both T cells and macrophages. In this model we differentiate between latent and active forms of TB by the amount of *M. tuberculosis* present. If the bacterial load is low in the body, we assume that infection is latent (Fig. 4a); if infection is active, the bacterial load could be at least 10-fold higher (not shown).

We then assume that the body is able to clear infection with *M. tuberculosis* (Fig. 4b).

Finally in Fig. 5, we explore co-infection (SS_T). When both *M. tuberculosis* and HIV are introduced to the immune system, the initial transient is present, but the T cells approach a much lower level than HIV infection alone (2.6-fold lower than with HIV alone) (see Fig. 3). Almost all cases of co-infection with HIV and TB eventually result in an active form of TB. The *M. tuberculosis* population is 11.8-fold higher (Fig. 5) than TB-infection alone (see Fig. 4). The viral load is 3 times higher in the

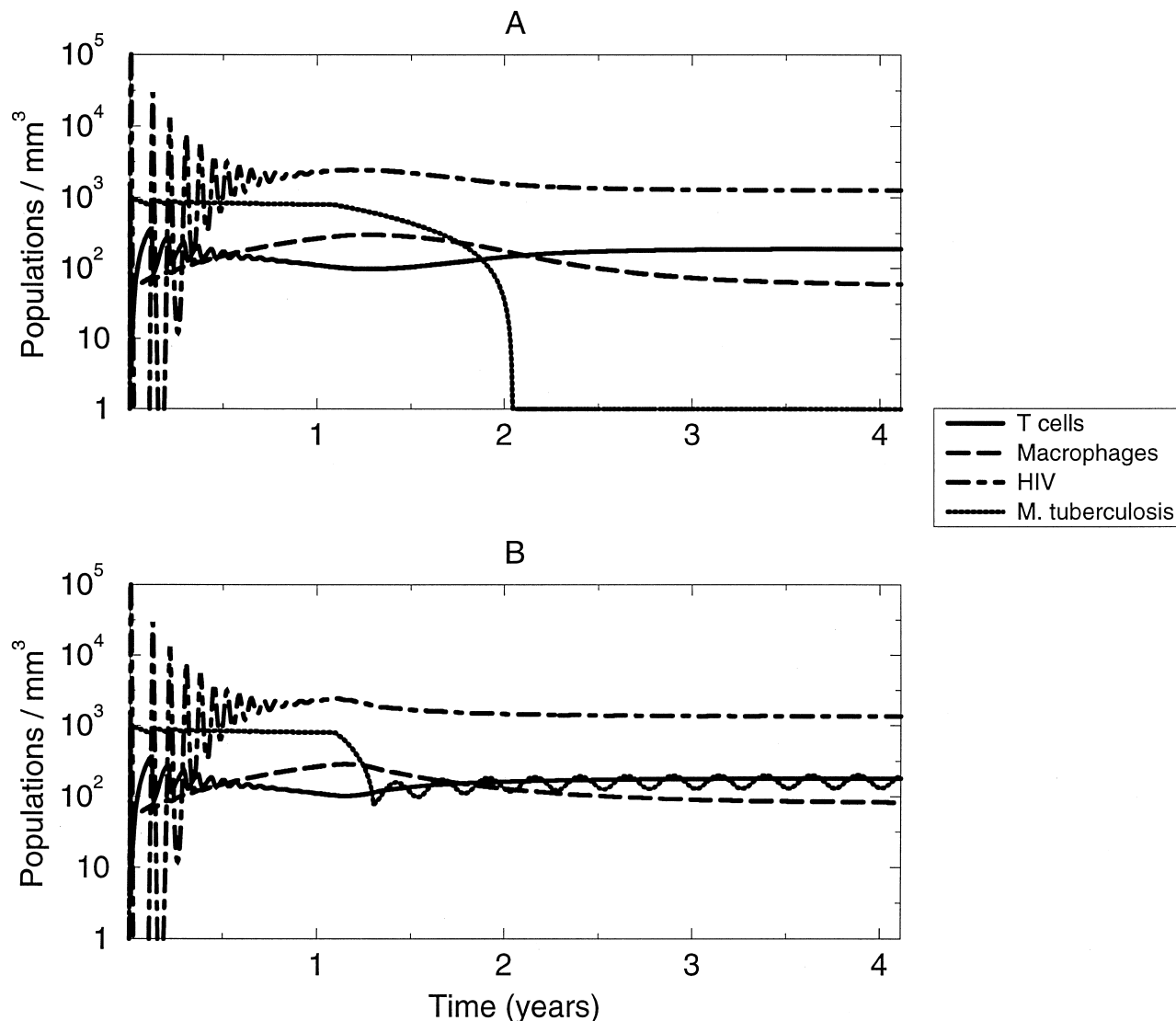


FIG. 7. Treatment simulation. This is the numerical solution to (1)–(4) including the treatment of *M. tuberculosis*. Notice that the macrophage and T-cell populations increase over a period of a few months during treatment and that the viral load decreases **Panel A**. All parameters come from Table I, except the treatment parameters $r_{T_b} = 1$ and μ_{T_b} increases to 600 over 1 year. **Panel B** shows the result of non-compliance after 75 days of complied treatment. Non-compliance is represented by the periodic function $\mu_{T_b} = 35\sin[0.07t] + 700$. Values are presented on a log-linear plot.

co-infected scenario than in HIV alone. Table II summarizes the numerical output.

4. EFFECTS OF TREATMENT

We now explore treatment of *M. tuberculosis* infection in the co-infected individual. We do not model HIV drug treatment here. Most of the HIV and TB co-infection cases worldwide are treated only for TB, because antituberculosis drugs are cheaper and more readily available than antiviral drugs. Other models are concerned with the effects of chemotherapy for HIV, and with the development of resistant mutants (e.g., Kirschner and Webb, 1996, 1997a, 1997b, 1997c).

Recall first the rate equation for *M. tuberculosis*, Eq. (4). When exploring treatment against bacteria, there are two key drug-effect concepts: minimal inhibitory concentration (MIC) and minimal bactericidal concentration (MBC). MIC refers to the drug effect of inhibiting the mycobacterium. This could correspond to restricting the growth via the rate constant, r_{T_b} . MBC refers to the drug-killing action on the bacteria, for example, enhancing the death rate. This would correspond in the model to a larger death-rate constant, μ_{T_b} . As we do not intend to model the pharmacokinetic effects of the drug directly, we explore the whole range of plausible treatment outcomes. A bifurcation diagram showing the effects of altering these parameters is presented in Fig. 6. A key finding of this analysis is that treatment which perturbs the growth rate of bacteria (r_{T_b}) will be more effective than perturbations to the bacterial death rate (μ_{T_b}). A numerical simulation of the effects of a particular drug scenario (where in this case, $r_{T_b} = 1$ and μ_{T_b} increases linearly to

600 over one year) is given in Fig. 7a. Notice how the T cells and macrophages rebound, and the viral load is reduced in response to the treatment of TB. The *M. tuberculosis* population declines to zero over one year.

A recent study (Martin *et al.*, 1995) examined CD4⁺ T cell counts in African patients co-infected with both HIV and TB. They examined the initial T-cell counts and then followed the counts after treatment with standard TB protocol. They found that patients co-infected with HIV and *M. tuberculosis* had a much lower initial CD4⁺ T-cell count than those patients infected only with *M. tuberculosis*. After treatment was administered, the T-cell counts for both study groups rose significantly, although the co-infected patients took more than 3-months to gradually improve, while the TB-only group improved more rapidly (see Table III). In all cases, it is assumed that the therapy regimen was adhered to by the patient. Our model accurately simulates these data.

A problem facing disease treatment today is non-compliance to the therapy regime. This not only affects the immune-system, but may result in resistance to the drug or drugs administered (Iseman and Madsen, 1989; Vareldzis *et al.*, 1989). We briefly explored non-compliance by perturbing the linear treatment function, used in Fig. 7a, after 75 days, with a periodic function. This could represent variable perturbations in compliance. The results are present in Fig. 7b. The *M. tuberculosis* population ceases declining, and begins to slowly rise, while the T cells do not increase to as high as level, and begin to slowly decline. Even in this simple treatment model it is easy to see that non-compliance can cause complications in chemotherapy. A more detailed model encompassing resistance is needed to better explore non-compliance issues.

TABLE III

	CD4 count on Admission	CD4 count Month 1	CD4 count Month 2	CD4 count Month 3	
TB only	241	206	164	124	Sample size
	630	800	880	820	Median
	500–865	630–1080	715–1080	670–1085	Range
HIV&TB	104	73	56	45	Sample size
	230	270	260	380	Median
	90–475	90–540	120–510	200–600	Range

Note. These are data for CD4⁴ T-cell counts as presented in Martin *et al.* (1995). The data represent CD4⁴ T-cell counts of TB-positive and TB/HIV-positive patients from Africa. Values presented when admitted to the study (column 1); values at 1 month (column 2), 2 months (column 3) and 3 months (column 4) after initiation of treatment. The first row are patients infected only with *M. tuberculosis*; the second row are patients infected with both *M. tuberculosis* and HIV.

5. DISCUSSION

The focus of this study was to explore the hypothesis that the presence of infection with *M. tuberculosis* in the body worsens the clinical picture for HIV; and, that the presence of HIV can activate the *M. tuberculosis* infection. A simple mathematical model was developed to describe the interaction of the immune system's key players, T cells and macrophages, with the pathogens HIV and *M. tuberculosis*. It is the first dynamic model to examine HIV together with an opportunistic infection. It incorporates many of the features of our previous models of HIV-immune dynamics (cf. Kirschner and Perelson, 1995; Kirschner and Webb, 1996, 1997a, 1997b, 1997c; Perelson *et al.*, 1993). The first goal was to create a system that captures the immune-system dynamics in the absence of infection, and then to insure that the model responds reasonably well to the introduction of each pathogen. This is judged on the basis of comparisons with clinical data. Figures 2–4 present these findings. The next goal was to explore the interaction of both pathogens with the immune system. We find that the T-cell populations are lower in the presence of both *M. tuberculosis* and HIV (Fig. 5) than in the case of infection with HIV alone (Fig. 3). Also, the viral load and *M. tuberculosis* population are higher in the co-infected patient, than the single-pathogen infection cases.

Since the progression to AIDS is based on the T-cell counts and viral load, we conclude that the presence of *M. tuberculosis* in the HIV-infected individuals worsens the clinical picture. Also, HIV can activate latent TB causing the bacterial load to increase dramatically. This is corroborated in Section 4, when studying treatment of TB in the co-infected case. When treatment is administered, the T-cell counts do improve over a short period of time (Figs. 6, 7, and Table II). Therefore, treatment of TB in HIV-infected individuals can have a profound effect on their progression to AIDS. And, when designing treatment, a drug that suppresses bacterial growth, as opposed to enhancing the bacterial death rate, will likely be more effective. We thus recommend screening HIV-infected individuals at high risk for TB (or showing any clinical signs of TB), and then initiation of a complete course of treatment for TB positive individuals. In the cases of multi-drug resistant TB, this approach becomes difficult. Further work will be needed to examine the role of development of drug-resistance in both TB and TB-HIV infections. A good epidemic model addressing these issues was recently done by (Blower *et al.*, 1996].

The importance of the effects of opportunistic infections on the course of HIV are now being explored.

Our study indicates that co-infection may indeed play a dramatic role in disease.

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