Within Host Dynamics for Treatment of R5 HIV Infection in the Langerhans Cells

Edna Chilenje Manda (edna@aims.ac.za)
African Institute for Mathematical Sciences (AIMS)

Supervised by: Dr Faraimunashe Chirove
University of KwaZulu-Natal, South Africa

22 May 2014

Submitted in partial fulfillment of a structured masters degree at AIMS South Africa

Abstract

Found in the genital mucosa are antigen presenting Langerhans cells that have characteristics and features that attract R5 HIV towards them. Some characteristics include their ability to capture and degrade R5 HIV and the loss of their antigen presenting properties when they are overwhelmed. Hence, they play both inhibitory and mediatory roles for R5 HIV infection. The project investigates the benefits of modelling R5 HIV dynamics in the Langerhans cells during early HIV infection within the host incorporating treatment with maraviroc as an entry inhibitor. Critical thresholds, the reproduction number (R_0), important in determining the invasion of R5 HIV in the Langerhans cells were derived. The results in this study showed that there exist two equilibrium points. These are the disease free equilibrium (DFE) point and the endemic equilibrium point. Stability analysis of the equilibrium points using the Routh-Hurwitz and the center manifold theory techniques was carried out. Our results also showed that there is a supercritical bifurcation that ensures the transition of stability between the two equilibrium points when $R_0 = 1$. The numerical simulation results suggest that entry inhibitors may need to have high efficacies for them to be effective in reducing the infection of Langerhans cells by R5 HIV.

Declaration

I, the undersigned, hereby declare that the work contained in this research project is my original work, and that any work done by others or by myself previously has been acknowledged and referenced accordingly.

Edna Chilenje Manda, 22 May 2014

Contents

1. Introduction

1.1 Background information

A Langerhans cell is a type of white blood cell. It is part of the immune system and subset of dendrite cells that reside in the epidermis. Langerhans cells are antigen-presenting cells that capture invading viruses and induce antiviral immunity. They are found in all layers of the epidermis mostly the dermis (uppermost layer) and around blood vessels in mucosa, foreskin and vagina. They are produced from progenitors in the bone marrow. They contain langerin (protein) which enables the production of birbeck granules that degrades the virus inside the cell (Katriena, 2014).

According to Figdor et al. (2002), Langerhans cells highly contribute to the immune system response and are an integral part of the body's total defence. They are adapted to immune functions in a way that their long dendrites capture antigens in the skin and carry the antigens to the $CD4^+$ T-cells in the lymph nodes allowing the adaptive immune system to respond. The adaptive immune system is a specific immune system composed of highly specialised systemic cells that process and eliminate pathogen growth. This immune system is composed of lymphocytes that are divided into B-cells and T-cells produced in the bone marrow. In addition Langerhans cells help protect the body by keeping dangerous microbes from entering the body by defending the skin from infection and stimulating allergic reactions. In general, Langerhans cells prevent infections and they help stimulate immune reactions from the $CD4^+$ T-cells. However, on one hand an excess production of Langerhans cells in the body leads to damage of skin, bones and other organs causing a disease called Langerhans-cells histiocytosis. On the other hand, Langerhans cells are cellular targets in sexual transmission of human immunodeficiency virus (HIV).

The Human Immunodeficiency Virus (HIV) is a retrovirus which attacks the $CD4^+$ T-cells in the immune system. A retrovirus is a single stranded ribonucleic acid (RNA) virus with nucleic acid in the form of a messenger RNA (mRNA) genome. It uses the host cells as production machines. HIV has ribonucleic acid as its genetic material, and is a devastating human pathogen that causes AIDS. The virus can stay quiescent in an infected host for a long time, allowing its long-term survival in the infection process. HIV is commonly transmitted through contact with blood, semen, vaginal secretions and breast milk (Douek et al., 2009).

Once HIV enters the body, it infects $CD4^+$ T-cells which are crucial for immune response. Millions of $CD4^+$ T-cells are produced by the body to assist with the immunity maintenance and destroy viruses and germs. HIV attaches to the protein $CD4$ present on the surface of $CD4$ T-cells and enters the cell. HIV then duplicates itself increasing its potential to kill the $CD4^+$ T-cells and as a result the infected cells outnumber the healthy $CD4^+$ T-cells making them unavailable to immune defence. Hence, HIV weakens the immune system putting an individual at risk of developing opportunistic infections because of low $CD4^+$ T-cells. Ultimately, the individual develops AIDS which leads to death (HIV and LONG, 2012).

HIV infection has 3 stages of infection which are acute primary infection, clinical latent infection and the AIDS stage. The acute primary infection stage is characterised by large amounts of viruses produced in the body leading to development of flu-like symptoms. The second stage is the chronic stage where HIV reproduce at very low levels. During the chronic stage one may not have symptoms. The use of treatment as an intervention increases the chances of people surviving with clinical latency for several years. The last stage is the AIDS stage where the immune system is overwhelmed and the body becomes

vulnerable to opportunistic infections. A person with $CD4$ count of below $200 cells/mm^3$ of blood is considered to be in the AIDS stage (AIDS.gov).

HIV uses two cell surface chemokine receptors, chemokine co-receptor type 5 (CCR5) and chemokine co-receptor type 4 (CXCR4) as co-receptors for infection, and these viruses are referred to as R5 and X4 HIV respectively. CCR5 is a protein on the surface of white blood cells which is a receptor for chemokines and CXCR4 is a protein encoded by the CXCR4 gene in humans. Initially HIV uses CCR5 to attach and infect host cells and R5 HIV is the one that infects Langerhans cells (Locher et al., 2005).

Langerhans cells are linked to HIV because they are initial targets for the transmission of the virus following sexual exposure, for instance, in vitro studies showed that Langerhans cells are infected by HIV and they can also transmit the infection of the virus to other susceptible cells. In HIV infection, Langerhans cells play a dual role in promoting immunity while also facilitating infections. During antigen presentation, Langerhans cells capture virions at the site of transmission and migrate to the lymphoid tissues to present the virions to naive T-cells and hence, are responsible for the infection of $CD4^+$ T-cells. These interactions suggest that Langerhans cell dynamics play a crucial role in the transmission of HIV to $CD4^+$ T-cells thus infection occurs as Langerhans cells have CCR5 co-receptors which exposes them to R5 HIV (Dusserre et al., 1992).

1.2 Treatment

Currently, available HIV treatment cannot cure HIV but can keep the amount of the virus in the blood low. As HIV has different stages of multiplicity, taking a combination of different types of drugs together is required to help fight the virus. The drugs intercept the virus from multiplying at the different stages. According to INTELENCE, the types of HIV treatment are entry inhibitors, reverse transcriptase inhibitors, integrase inhibitors and protease inhibitors. Entry inhibitors work by stopping HIV entry into the host cells, reverse transcriptase inhibitors help to block, bind and disable the reverse transcriptase proteins that HIV uses to multiply. Integrase inhibitors block the enzyme that HIV needs to infect $CD4^+$ T-cells with its genetic material and the protease inhibitors blocks an enzyme that HIV needs to make copies of itself. This study will focus on maraviroc, a type of entry inhibitor, which is particularly used in Langerhans cells infection prevention.

Maraviroc is a type of anti-HIV drug referred to as a CCR5 receptor antagonist. When HIV binds to the CCR5 receptor to enter cells, maraviroc binds to CCR5 blocking the HIV protein from attaching to the receptor on healthy immune cells thereby preventing HIV infection (Levy, 2009). When used with other anti-HIV drugs, maraviroc lowers the amount of HIV in the blood. Because HIV can also use other co-receptors such as CXCR4, an HIV tropism test such as trofile assay must be performed to determine if the drug will be effective (Biswas et al., 2007). It is recommended that maraviroc should be used only in people with R5 HIV strain.

1.3 Problem statement

The analysis of R5 HIV infection in Langerhans cells has not yet been adequately given attention in as far as the contribution of Langerhans cells towards HIV infection progression is concerned during the early HIV infection stage. Mathematical models have been used to provide insights in HIV dynamics within the host especially on $CD4^+$ T-cells. We develop a mathematical model to investigate the infection of

Langerhans cells during early HIV infection within the host and the benefits of introducing treatment with maraviroc as an entry inhibitor to R5 HIV-Langerhans cell interaction.

1.4 Aim

We seek, in this study, to use mathematical models to investigate the benefits of modelling R5 HIV dynamics in the Langerhans cells during early HIV infection within the host using treatment with maraviroc as an intervention strategy.

1.5 Objectives

The objectives of the study are:

1. To develop a mathematical model that captures the interaction between Langerhans cells and R5 HIV.
2. To analyse the model to determine the qualitative behaviour of solutions of the model.
3. To carry out numerical simulations to get insights on the prognosis of R5 HIV in Langerhans cells in the long run.
4. To validate our results with other findings.

1.6 Significance of study

The study results may be used as a basis of enhancing the understanding of the benefits of introducing preventive treatment of Langerhans cells during early HIV infection to reduce the infectiousness of individuals in this stage.

1.7 Scope of study

This chapter has given an introduction on interactions between Langerhans cells and HIV. Chapter 2 will present a review of the studies on Langerhans cell-HIV interaction that have been done previously by other researchers and some preliminary concepts which will be used for model analysis in the study. In chapter 3, a model will be formulated and detailed analysis of the model will be provided. Chapter 4 will contain numerical simulations and chapter 5 explains a detailed discussion of results, observations, conclusion, strengths of model, weaknesses of model and future work to be done including model extensions.

2. Literature Review and Preliminary Concepts

Introduction

This chapter reviews the work conducted by other researchers to get the general information on HIV infection in Langerhans cells. We also define and explain some major concepts that will be used in the mathematical analysis of the model.

2.1 Literature review

There are a number of clinical studies and few mathematical models on HIV infection in Langerhans cells. We shall select a few that we will review as building blocks to our study.

Wu and Ding (1999) studied HIV-1 dynamics $in\ vivo$. They introduced an application of hierarchical non-linear mixed-effects models to HIV dynamics and showed that a simple model with a sum of exponentials gave a good fit to observed clinical data of HIV-1 dynamics after administering antiviral treatments. They used a biological compartmental model for the interaction between HIV and host cells. A host cell refers to any cell which can be infected by a virus. They however, did not specify the type of host cell they used. In fact different host cells have different roles in the study of HIV infection. For instance Langerhans cells are host cells to HIV but are antigen presenting cells which can kill HIV or infect other cells such as $CD4^+$ T-cells. Their model was not simple and it may be difficult to interpret the results biologically and mathematically.

Sugaya et al. (2004) investigated how HIV-infected Langerhans cells preferentially transmit virus to proliferating $CD4^+$ memory T-cells within Langerhans cells clusters. The objective of their study was to examine the type of the $CD4^+$ T-cells that becomes infected by the HIV-infected Langerhans cells using a number of experiments. Their results showed that close interactions between epidermal Langerhans cells and $CD4^+$ T-cells were crucial for maximum HIV replication inside specific subsets of $CD4^+$ T-cells. Their study did not incorporate strategies such as treatment with entry inhibitors which blocks the infection from HIV after sexual exposure to HIV.

Mbogo et al. (2014) studied a stochastic model for Langerhans cells and HIV dynamics $in\ vivo$. In their study, they derived and analysed a stochastic model explaining the dynamics of HIV, $CD4^+$ T-cells and Langerhans cells interactions under therapeutical interventions $in\ vivo$. Their model results showed that HIV states should not be based on $CD4^+$ T-cells as the target cells infected by HIV. The findings illustrated the role of Langerhans cells as a central hub of interaction and information exchange during HIV infection. But their model did not capture the degradation effects of HIV by Langerhans cells as well as treatment of Langerhans infection by HIV. Furthermore they did not specify the type of HIV that prefer infecting the Langerhans cells.

2.2 Preliminary concepts

2.2.1 Basic reproduction number (R_0). Van den Driessche and Watmough (2002) defined R_0 as the average number of second generation infections produced by a typical infective person in a totally susceptible population. In their approach for computing R_0, they assumed that there are n compartments

of which m are infected, $\overline{x} = (x_1, x_2, ..., x_n)$, where $\overline{x}$ is the disease free equilibrium (DFE) point and x_i denotes the number of individuals in the i^{th} compartment. Let $\mathcal{F}_i(\overline{x})$ be the rate at which new infections appear into compartment i and the transition matrix $\mathcal{V}_i(\overline{x}) = \mathcal{V}_i^-(\overline{x}) - \mathcal{V}_i^+(\overline{x})$, where $\mathcal{V}_i^+$ is the rate of transfer of individuals into compartment i by all other means and $\mathcal{V}_i^-$ is the rate of transfer of individuals out of the i^{th} compartment. We then introduce the next generation matrix (FV^{-1}) from matrices of partial derivatives of $\mathcal{F}_i, \mathcal{V}_i$ evaluated at $\overline{x}$. $F = \left[\frac{\partial \mathcal{F}_i(\overline{x})}{\partial x_j}\right]$ and $V = \left[\frac{\partial \mathcal{V}_i(\overline{x})}{\partial x_j}\right]$, where $i, j = 1, ..., m$. The entries of FV^{-1} give the rate at which infected individuals in x_j produce new infections in x_i, times the average length of time an individual spends in a single visit to compartment j. R_0 is given by the spectral radius (largest eigenvalue) of the matrix FV^{-1}. Thus $R_0 = \rho(FV^{-1})$.

2.2.2 Bifurcation. According to Sastry (1999), a bifurcation is a change in the nature of a solution of trajectories due to a parameter change. A transcritical bifurcation is a bifurcation where there is an exchange of stability between two equilibrium points at a bifurcation point where the stability is transferred from one equilibrium point to another. In infectious disease models, the exchange of stability occurs between a disease free equilibrium (DFE) point and the endemic equilibrium point at $R_0 = 1$ where the DFE point looses its stability and the endemic equilibrium point becomes stable depending on the conditions on R_0. A transcritical bifurcation can be either forward or backward (Britton, 2003). In forward bifurcation, the DFE point looses its stability when it passes through the bifurcation point and endemic equilibrium gains its stability ensuring that the endemic point is locally stable when $R_0 > 1$.

A backward bifurcation in epidemic models occurs when there is existence of two sub-critical endemic equilibria for $R_0 < 1$. The initial direction of the bifurcation curve is such that as we move along it from the bifurcation point, R_0 will decrease as the level of infection increase. The occurrence of backward bifurcation affects disease control since it can be possible for the disease to progress even when $R_0 < 1$. There is need to further reduce R_0 to ensure that the disease is eliminated from a population. At least two sub-critical endemic equilibria exist for which $R^* < R_0 < 1$ where R^* corresponds to the value of R_0 at which a vertical turning point on the bifurcation curve occurs. Thus, to ensure that a disease is cleared from a population we require that $R_0 < R^*$.

2.2.3 Center manifold theory (CMT). The CMT provides a technique to analyse a bifurcation. The theory plays a great role in the study of the stability of non-linear systems when the equilibrium point is not hyperbolic (Hamzi et al., 2004). For the equilibrium point, the dimensions of centre manifolds is determined by the number of roots of the characteristic equation with zero real parts.

If we consider a continuous-time system defined by $\dot{x} = f(x)$, x $\in \mathbb{R}^n$, where f is a sufficiently smooth vector field on $\mathbb{R}^n$ with $f(0) = 0$, we can find the eigenvalues $\lambda_1, \lambda_2, ..., \lambda_n$ of the Jacobian matrix A of the system evaluated at the equilibrium point $x_0 = 0$. Suppose the equilibrium point is not hyperbolic and that there are eigenvalues with zero real parts, we assume that there are n_+ eigenvalues with $Re(\lambda) > 0$, n_0 eigenvalues with $Re(\lambda) = 0$ and n_- eigenvalues with $Re(\lambda) < 0$. We let T^c denote the linear eigen space of matrix A corresponding to the union of the set of n_0 eigenvalues and imaginary axis. We describe the CMT using the theorem below:

2.2.4 Theorem. *Castillo-Chavez and Song (2004)*

Consider a general system of ODE's with a parameter ϕ:

$$\frac{dx}{dt} = f(x, \phi), f : \mathbb{R}^n \times R \to \mathbb{R}^n \quad \textit{and} \quad f \in \mathbb{C}^2(\mathbb{R}^n \times \mathbb{R}). \tag{2.2.1}$$

Without loss of generality, it is assumed that 0 is an equilibrium for system (2.2.1) for all values of the parameter ϕ, that is

$$f(0, \phi) \equiv 0 \quad \textit{for all } \phi. \tag{2.2.2}$$

Assume

1. $A = D_x f(0;0) = \left(\frac{\partial f_i}{\partial x_j}(0,0)\right)$ *is the linearisation matrix of system* (2.2.1) *around the equilibrium* 0 *with* ϕ *evaluated at 0. Zero is a simple eigenvalue of A and all other eigenvalues of* A *have negative real parts.*
2. *Matrix* A *has a non-negative right eigenvector* w *and a left eigenvector* v *corresponding to the zero eigenvalue.*

Let f_k *denote the* kth *component of* f *and*

$$a = \sum_{k,i,j=1}^{n} v_k w_i w_j \frac{\partial^2 f_k}{\partial x_i \partial x_j}(x_0, 0), \qquad b = \sum_{k,i=1}^{n} v_k w_i \frac{\partial^2 f_k}{\partial x_i \partial \varphi}.$$

Then the local dynamics of system (2.2.1) *around* 0 *are totally determined by a and b.*

(i) $a > 0, b > 0$. *When* $\phi < 0$, *with* $|\phi| << 1$, 0 *is locally asymptotically stable and there exists a positive unstable equilibrium; when* $0 < \phi << 1$, 0 *is unstable and there exists a negative and locally asymptotically stable equilibrium.*

(ii) $a < 0, b < 0$. *When* $\phi < 0$, *with* $|\phi| << 1$, 0 *is unstable; when* $0 < \phi << 1$, 0 *is locally asymptotically stable and there exists a positive unstable equilibrium.*

(iii) $a > 0, b < 0$. *When* $\phi < 0$, *with* $|\phi| << 1$, 0 *is unstable and there exists a locally asymptotically stable negative equilibrium; when* $0 < \phi << 1$, 0 *is stable and a positive unstable equilibrium appears.*

(iv) $a < 0, b > 0$. *When* ϕ *changes from negative to positive,* 0 *changes its stability from stable to unstable. Correspondingly, a negative unstable equilibrium becomes positive and locally asymptotically stable.*

2.2.5 Descartes rule of signs . (Murray, Springer, New York, 2002)

Considering the n^{th} polynomial

$$f(\lambda) = \lambda^n + b_1\lambda^{n-1} + ... + b_n = 0 \tag{2.2.3}$$

and without loss of generality $b_n > 0$. Letting N be the number of sign changes in the sequence of coefficients $b_n, b_{n-1}, ..., b_0$, and ignoring those which are zero. Descartes rule of signs states that there are at most N roots of the given polynomial (2.2.3) which are real and non-negative. Further, the rule states that there are $N - 2K, K \geq 0$ and $K \in \mathbb{Z}^+$ real positive roots. If we let $\omega = -\lambda$ and again applying the Descartes rule of signs we obtain $N - 2K$ real negative roots.

2.2.6 Equilibrium point. (Wikipedia, a) The point $\tilde{\mathbf{x}} \in \mathbb{R}^n$ is an equilibrium point for the differential equation

$$\frac{d\mathbf{x}}{dt} = \mathbf{f}(t, \mathbf{x})$$

if $\mathbf{f}(t, \tilde{\mathbf{x}}) = 0$ for all t.

2.2.7 Stability Analysis. (Wikipedia, b) Stability theory is used to analyse the stability of solutions of differential equations and trajectories of dynamical systems under small perturbations of initial conditions. It therefore helps us to understand what happens when we perturb a system. The analysis allows us to determine whether or not a system is stable or will be stable if perturbed.

2.2.8 Routh Hurwitz criterion . (Murray, Springer, New York, 2002) Supposing we have a characteristic polynomial

$$P(\lambda) = \lambda^n + a_1\lambda^{n-1} + ... + a_{n-1}\lambda + a_n,$$

where the coefficients a_i are real constants, $i = 1, \cdots, n$. The n Hurwitz matrices of the characteristic polynomial are given by

$$D_1 = a_1 > 0, D_2 = \begin{vmatrix} a_1 & a_3 \\ 1 & a_2 \end{vmatrix} > 0, D_3 = \begin{vmatrix} a_1 & a_3 & a_5 \\ 1 & a_2 & a_4 \\ 0 & a_1 & a_3 \end{vmatrix} > 0, D_n = \begin{vmatrix} a_1 & a_3 & ... & ... & ... \\ 1 & a_2 & a_4 & ... & ... \\ 0 & a_1 & a_3 & ... & ... \\ ... & ... & ... & ... & ... \\ 0 & 0 & ... & ... & a_n \end{vmatrix} > 0.$$

These conditions are derived using complex variable methods. For example, if we consider a cubic equation

$$\lambda^3 + a_1\lambda^2 + a_2\lambda + a_3 = 0,$$

then the conditions for $Re(\lambda) < 0$ are

$$a_1 > 0, \quad a_3 > 0, \quad a_1a_2 - a_3 > 0.$$

2.2.9 Jacobian matrix and Characteristic equation . Brauer and Castillo-Chavez (2011) defines a Jacobian as a matrix of all first-order partial derivatives of a vector-valued function. Suppose $F : \mathbb{R}^n \longrightarrow \mathbb{R}^m$ is a function, where F is given by m-real-valued component functions $F_1(x_1, \ldots, x_n), \ldots, F_m(x_1, \ldots, x_n)$. The partial derivatives of all these functions with respect to the variables $x_1, \cdots, x_n$ (if they exist) can be organised in an $m \times n$ matrix. The Jacobian matrix J of F is given as follows:

$$J = \begin{pmatrix} \frac{\partial F_1}{\partial x_1} & \cdots \frac{\partial F_1}{\partial x_n} \\ \vdots & \vdots \\ \frac{\partial F_m}{\partial x_1} & \cdots \frac{\partial F_m}{\partial x_n} \end{pmatrix}.$$

A characteristic equation of a matrix is the equation in one variable λ of the form $\det(J - \lambda I) = 0$ where $\det$ is the determinant of a matrix and I is the identity matrix and J is the Jacobian matrix. The solutions of the characteristic equation are precisely the eigenvalues of the matrix J.

2.2.10 Simple eigenvalue. An eigenvalue λ of a matrix A is called simple if its algebraic multiplicity $m_A(\lambda) = 1$ (Queens University) .

Summary

This chapter has reviewed different views that some researchers had on HIV infection in the Langerhans cells and also concepts that are going to be used in the model analysis have been defined and a detailed explanation of how to calculate them . Thus, we now formulate the model in the next chapter.

3. Mathematical Model for Langerhans Cells-R5 HIV Interactions

Introduction

This chapter explains the formulation of an R5 HIV infection in Langerhans cells model. The model incorporates treatment with a drug called maraviroc against R5 HIV. We will show that our model is positively invariant. Then we shall carry out mathematical analysis by calculating the reproduction number, determining the equilibrium points and their stability using the center manifold theory and the Routh-Hurwitz criterion inside the positive region.

3.1 Model formulation

We shall consider 2 populations; Langerhans cells population and virus population at a time t. The Langerhans population is divided into sub-populations of healthy Langerhans cells denoted $L(t)$, which help to defend against infection, the latently infected Langerhans cells, $L_T(t)$, are the Langerhans cells that have been infected by the virus but cannot transmit it to other cells and the actively infected Langerhans cells, $L_I(t)$, are the Langerhans cells that are infected by the virus and can transmit it to other cells. The type of HIV that we consider in our model is the R5 HIV denoted by $V_I(t)$. It infects and replicates in the Langerhans cells.

The healthy Langerhans cells are recruited from the bone marrow at a constant rate π and die naturally at a constant rate μ. The healthy Langerhans cells are removed from their class through infection by R5 HIV and actively infectious Langerhans cells at an infection rate $\beta(1-\epsilon_r)(V_I+\eta L_I)L$ where $0 \leqslant \epsilon_r \leqslant 1$ and $\eta < 1$. $\beta(1-\epsilon_r)(V_I+\eta L_I)$ is called the force of infection which is the probability of a susceptible population to get an infection per unit time. β is the rate of successful infection of the healthy Langerhans cells and ϵ_r is the efficacy that maraviroc drug has against the R5 virus. $\epsilon_r = 0$ means that there is no effect of maraviroc drug against the R5 virus and $\epsilon_r = 1$ means that there is a 100% effect of maraviroc drug against the virus. η is the reduction rate of level of risk of R5 HIV transmission by infectious Langerhans cells compared to the free R5 virus.

All infected healthy Langerhans cells become latently infected and act as a reservoir before they are activated to become actively infectious. The latently infected Langerhans cells cannot transmit the virus to healthy Langerhans cells and cannot produce the virus. They die naturally at a rate μ. They are removed from the latent class through activation and progress to the infectious class at a rate γ.

The actively infected Langerhans cells get their source from activated latently infected Langerhans cells at a rate γ. The viral particles multiply inside the infectious Langerhans cells and when they become mature, they leave the cell by lysing the cell that produces them at a constant rate δ. Each infectious cell produces an average of N viral particles. The new virus produced will move in the blood as a new virus source and infect healthy Langerhans cells at a rate $\beta\eta(1-\epsilon_r)$. They die naturally at a rate μ.

The R5 HIV population grows through a source from bursting actively infectious Langerhans cells at a rate δN. The virus is killed by langerin inside the healthy Langerhans cells at a rate ϕ and by natural death at a rate μ_v. Maraviroc as an entry inhibitor increases the chances of R5 HIV degradation of Langerhans cells by a factor $1+\epsilon_r$. We assume that the degradation of the virus is saturated.

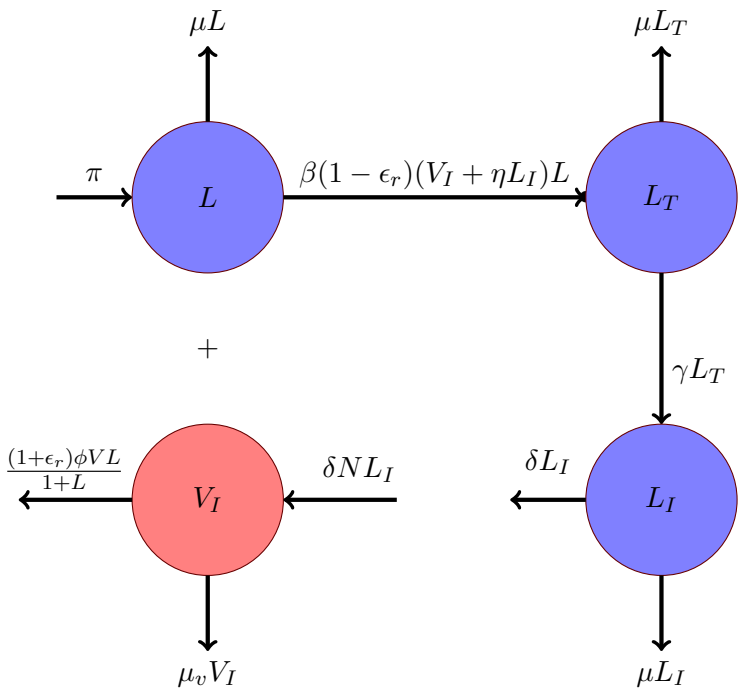

Figure 3.1: Model flow diagram.

The model flow diagram and assumptions result in a system of ordinary differential equations given by;

$$\frac{dL}{dt} = \pi - \beta(1-\epsilon_r)(V_I+\eta L_I)L - \mu L, \tag{3.1.1}$$

$$\frac{dL_T}{dt} = \beta(1-\epsilon_r)(V_I+\eta L_I)L - (\mu+\gamma)L_T, \tag{3.1.2}$$

$$\frac{dL_I}{dt} = \gamma L_T - (\mu+\delta)L_I, \tag{3.1.3}$$

$$\frac{dV_I}{dt} = \delta N L_I - \frac{(1+\epsilon_r)\phi V_I L}{1+L} - \mu_v V_I. \tag{3.1.4}$$

3.2 Model analysis

3.2.1 Positivity and boundedness of solutions. Before we analyse the model (3.1.1) to (3.1.4), we need to prove that all the state variables are positive meaning that the solutions of the system of equations with positive initial conditions remain positive for all $t \geq 0$ and that the solutions are bounded for all $t \geq 0$ in a positive region. The region is called a biologically feasible region where the model will be biologically meaningful. This means that the dynamics of population of Langerhans cells and the virus should remain positive in all our analysis. We shall determine conditions which ensure positivity of solutions.

3.2.2 Theorem. *Let* $L(0) \geq 0$, $L_T(0) \geq 0$, $L_I(0) \geq 0$, $V_I(0) \geq 0$.

Then the solutions of $L(t), L_T(t), L_I(t), V_I(t)$ *are positively invariant for all* $t \geq 0$ *in the region*

$$\Omega = \left\{(L, L_T, L_I, V_I) \in \mathbb{R}^4_+ | L \geq 0, L_T \geq 0, L_I \geq 0, V_I \geq 0\right\}.$$

Proof. Let $\lambda = \beta(1-\epsilon_r)(V_I + \eta L_I)$.

For $t \geq 0$, equation (3.1.1) becomes

$$\frac{dL}{dt} + (\lambda + \mu)L = \pi,$$

whose solution is

$$\begin{aligned} L(t) &= L(0)\mathrm{e}^{\left(-(\mu t + \int_0^t \lambda(s)ds)\right)} \\ &\quad + \mathrm{e}^{\left(-(\mu t + \int_0^t \lambda(s)ds)\right)} \left[\int_0^t \pi \mathrm{e}^{\left(\mu s + \int_0^s \lambda(r)dr\right)} ds\right] \geq 0. \end{aligned}$$

From (3.1.2) and (3.1.3) we obtain

$$\frac{dL_T}{dt} \geq -(\mu + \gamma)L_T \quad \text{and} \quad \frac{dL_I}{dt} \geq -(\mu + \delta)L_I,$$

which become

$$L_T(t) \geq L_T(0)\mathrm{e}^{-(\mu+\gamma)t} \geq 0 \quad \text{and} \quad L_I(t) \geq L_I(0)\mathrm{e}^{-(\mu+\delta)t} \geq 0.$$

Similarly, for (3.1.4) we have

$$\begin{aligned} \frac{dV_I}{dt} &\geq -\left(\frac{(1+\epsilon_r)\phi L(s)}{1+L(s)} + \mu_v\right) V_I, \\ V_I(t) &\geq V_I(0)\mathrm{e}^{-\left(\int_0^t \frac{(1+\epsilon_r)\phi L(s)}{1+L(s)} ds + \mu_v t\right)} \geq 0. \end{aligned}$$

Therefore all state variables are non-negative.

To prove boundedness of solutions, we consider the total population of Langerhans cells as $L+L_T+L_I = L_{tot}$ by adding the right hand sides of (3.1.1) to (3.1.3) such that

$$\frac{dL_{tot}}{dt} = \pi - \mu L_{tot} - \delta L_I \leq \pi - \mu L_{tot}. \tag{3.2.1}$$

Solving (3.2.1) we have

$$L_{tot} \leq \left(L_{tot}(0) - \frac{\pi}{\mu}\right)\mathrm{e}^{-\mu t} + \frac{\pi}{\mu}.$$

Taking the limit supremum of L_{tot} as $t \longrightarrow \infty$ yields

$$\lim_{t\to\infty} \sup L_{tot} \quad \leq \frac{\pi}{\mu} = L_0.$$

This means that

$$L(t) \leq L_0, \quad L_T(t) \leq L_0 \quad \text{and} \quad L_I(t) \leq L_0.$$

For the virus population

$$\begin{aligned} \frac{dV_I}{dt} &= \delta N L_I - \left(\frac{(1+\epsilon_r)\phi L}{1+L} + \mu_v\right) V_I \leq \delta N L_I - \mu_v V_I, \\ &\leq \delta N L_0 - \mu_v V_I, \end{aligned}$$

which yields

$$V_I \leq \frac{\delta N L_0}{\mu_v} + C\mathrm{e}^{-\mu_v t}.$$

Taking the limit supremum of V_I as $t \longrightarrow \infty$ yields

$$\lim_{t\to\infty} \sup V_I(t) \leq \lim_{t\to\infty} \sup \left(\frac{\delta N L_0}{\mu_v} + C\mathrm{e}^{-\mu_v t} \right) = \frac{\delta N L_0}{\mu_v} = M_0.$$

Since

$$0 \leq L \leq L_0, \quad 0 \leq L_T \leq L_0, \quad 0 \leq L_I \leq L_0 \quad \text{and} \quad 0 \leq V_I \leq M_0,$$

choosing $M = max\left\{L_0, M_0\right\}$, we have that $L, L_T, L_I, V_I \leq M$. Thus all state variables are bounded. Since all state variables are positive and bounded in $\mathbb{R}^4_+$, then the region Ω is positively invariant. □

3.2.3 Reproduction Number (R_0)**.** The disease free equilibrium point which is the equilibrium point which occurs when there are no infected Langerhans cells in the body is obtained by setting the infectious classes $V_I^* = L_I^* = L_T^* = 0$ and is given by

$$E_0 = (L_0, 0, 0, 0), \quad L_0 = \frac{\pi}{\mu}.$$

Detailed calculations of equilibrium points are given in section 3.2.4 .

Using the Van den Driessche and Watmough (2002) method we have 3 infection classes and hence the matrix of new infections is

$$\mathcal{F} = \begin{pmatrix} \beta(1-\epsilon_r)(V_I + \eta L_I)L \\ 0 \\ 0 \end{pmatrix}.$$

We proceed to find the Jacobian matrix of $\mathcal{F}$, $D(\mathcal{F}(E^*))$, where E^* is an equilibrium point of the system.

$$D\left(\mathcal{F}(E^*)\right) = \begin{pmatrix} 0 & \beta(1-\epsilon_r)\eta L^* & \beta(1-\epsilon_r)L^* \\ 0 & 0 & 0 \\ 0 & 0 & 0 \end{pmatrix}. \tag{3.2.2}$$

The Jacobian matrix (3.2.2) evaluated at the disease free equilibrium gives rise to a matrix F given by

$$F = \begin{pmatrix} 0 & \beta(1-\epsilon_r)\eta L_0 & \beta(1-\epsilon_r)L_0 \\ 0 & 0 & 0 \\ 0 & 0 & 0 \end{pmatrix}. \tag{3.2.3}$$

The matrix $\mathcal{V}$, which is expressed as $\mathcal{V} = \mathcal{V}^- - \mathcal{V}^+$ is

$$\mathcal{V} = \begin{pmatrix} (\mu+\gamma)L_T \\ (\mu+\delta)L_I - \gamma L_T \\ \frac{(1+\epsilon_r)\phi L V_I}{1+L} + \mu_v V_I - \delta N L_I \end{pmatrix}.$$

The Jacobian matrix for $\mathcal{V}$ is

$$D\left(\mathcal{V}(E^*)\right) = \begin{pmatrix} \mu+\gamma & 0 & 0 \\ -\gamma & \mu+\delta & 0 \\ 0 & -\delta N & \frac{(1+\epsilon_r)\phi L^*}{1+L^*} + \mu_v \end{pmatrix}. \tag{3.2.4}$$

Evaluating the Jacobian matrix (3.2.4) at the disease free equilibrium point yields

$$V = \begin{pmatrix} \mu+\gamma & 0 & 0 \\ -\gamma & \mu+\delta & 0 \\ 0 & -\delta N & \frac{(1+\epsilon_r)\phi L_0 + \mu_v(1+L_0)}{1+L_0} \end{pmatrix}. \tag{3.2.5}$$

The inverse matrix of V, denoted by V^{-1} is given as

$$V^{-1} = \begin{pmatrix} \frac{1}{\mu+\gamma} & 0 & 0 \\ \frac{\gamma}{(\mu+\gamma)(\mu+\delta)} & \frac{1}{\mu+\delta} & 0 \\ \rho_1 & \rho_2 & \rho_3 \end{pmatrix},$$

where

$$\begin{aligned} \rho_1 &= \frac{N\gamma\delta(1+L_0)}{(\mu+\gamma)(\mu+\delta)(\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0)}, \\ \rho_2 &= \frac{N\delta(1+L_0)}{(\mu+\delta)(\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0)}, \\ \rho_3 &= \frac{1+L_0}{\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0}. \end{aligned}$$

The next generation matrix denoted FV^{-1} is

$$FV^{-1} = \begin{pmatrix} R_1 & R_2 & R_3 \\ 0 & 0 & 0 \\ 0 & 0 & 0 \end{pmatrix},$$

where

$$\begin{aligned} R_1 &= \frac{\beta(1-\epsilon_r)\eta L_0\gamma}{(\mu+\gamma)(\mu+\delta)} + \frac{\beta(1-\epsilon_r)L_0N\delta\gamma(1+L_0)}{(\mu+\delta)(\mu+\gamma)\left[\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0\right]}, \\ R_2 &= \frac{\beta(1-\epsilon_r)\eta L_0}{(\mu+\delta)} + \frac{\beta(1-\epsilon_r)L_0N\delta(1+L_0)}{(\mu+\delta)\left[\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0\right]}, \\ R_3 &= \frac{\beta(1-\epsilon_r)L_0(1+L_0)}{\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0)}. \end{aligned}$$

R_1 is the number of secondary infections caused by one latently infected Langerhans cell to come up with new latently infected Langerhans cells. R_2 is the number of secondary latently infected cells produced by actively infected Langerhans cells. R_3 is the number of latently infected Langerhans cells produced by each infectious virus.

The basic reproduction number denoted R_0 which is the spectral radius of FV^{-1} is given by

$$R_0 = \frac{\beta(1-\epsilon_r)\eta L_0\gamma}{(\mu+\gamma)(\mu+\delta)} + \frac{\beta(1-\epsilon_r)L_0N\delta\gamma(1+L_0)}{(\mu+\delta)(\mu+\gamma)\left[\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0\right]}.$$

In the reproduction number, the first term represents the number of secondary infections caused by the actively infectious Langerhans cells while the second term represents the number of secondary infections caused by the free virus.

3.2.4 Equilibrium points. The equilibrium points are obtained by setting the right hand side of equations (3.1.1) to (3.1.4) to zero. To facilitate the ease with which we calculate the equilibrium points, we let

$$\lambda^* = \beta(1-\epsilon_r)(V_I^* + \eta L_I^*). \tag{3.2.6}$$

The right-hand side of equation (3.1.1) becomes

$$\pi - \lambda^* L - \mu L \quad = 0,$$

which gives

$$L^* \quad = \frac{\pi}{\mu+\lambda^*}.$$

Equation (3.1.2) becomes

$$\lambda^* L^* - (\mu+\gamma)L_T^* \quad = 0,$$

yielding

$$L_T^* \quad = \frac{\pi\lambda^*}{(\mu+\gamma)(\mu+\lambda^*)}.$$

Similarly, equations (3.1.3) and (3.1.4) become

$$L_I^* = \frac{\gamma\lambda^*\pi}{(\mu+\gamma)(\mu+\lambda^*)(\mu+\delta)},$$

and

$$V_I^* = \frac{\delta N\gamma\pi\lambda^*(\mu+\lambda^*+\pi)}{(\mu+\gamma)(\mu+\lambda^*)(\mu+\delta)\left[(1+\epsilon_r)\phi\pi+\mu_v(\pi+\mu+\lambda^*)\right]},$$

respectively.

Substituting the expressions of V_I^* and L_I^* into (3.2.6) yields;

$$\lambda^* = \beta(1-\epsilon_r)\left(\frac{\delta N\gamma\pi\lambda^*(\mu+\lambda^*+\pi)}{(\mu+\gamma)(\mu+\lambda^*)(\mu+\delta)\left[(1+\epsilon_r)\phi\pi+\mu_v(\pi+\mu+\lambda^*)\right]}+\frac{\eta\gamma\lambda^*\pi}{(\mu+\gamma)(\mu+\lambda^*)(\mu+\delta)}\right),$$

which reduces to

$$\lambda^*\left(1-\frac{\beta(1-\epsilon_r)\delta N\gamma\pi(\mu+\pi+\lambda^*)}{(\mu+\gamma)(\mu+\lambda^*)(\mu+\delta)\left[(1+\epsilon_r)\phi\pi+\mu_v(\pi+\mu+\lambda^*)\right]}-\frac{\beta(1-\epsilon_r)\eta\gamma\pi}{(\mu+\gamma)(\mu+\lambda^*)(\mu+\delta)}\right) = 0.$$

Either

$$\lambda^* = 0,$$

or

$$1-\frac{\beta(1-\epsilon_r)\delta N\gamma\pi(\mu+\pi+\lambda^*)}{(\mu+\gamma)(\mu+\lambda^*)(\mu+\delta)\left[(1+\epsilon_r)\phi\pi+\mu_v(\pi+\mu+\lambda^*)\right]}-\frac{\beta(1-\epsilon_r)\eta\gamma\pi}{(\mu+\gamma)(\mu+\lambda^*)(\mu+\delta)} = 0. \quad (3.2.7)$$

$\lambda^* = 0$ corresponds to the disease free equilibrium where $L^* = \frac{\pi}{\mu}$, when $V_I^* = L_I^* = L_T^* = 0$.

To find the other equilibrium point, we consider (3.2.7) which when expressed in terms of λ^* gives

$$a_2\lambda^{*2}+a_1\lambda^*+a_0 = 0, \quad (3.2.8)$$

where

$$\begin{aligned}
a_2 &= \mu_v(\mu+\gamma)(\mu+\delta),\\
a_1 &= \mu(\mu+\gamma)(\mu+\delta)\left((1+\epsilon_r)\phi L_0+\mu_v(1+L_0)\right)(1-R_0)\\
&\quad+\frac{\mu\beta(1-\epsilon_r)\gamma L_0\left[\delta N\left(1+\epsilon_r\right)\phi L_0+2\mu_v(1+L_0))+\eta\left((1+\epsilon_r)\phi L_0+\mu_v(1+L_0)\right)^2\right]}{(1+\epsilon_r)\phi L_0+\mu_v(1+L_0)},\\
a_0 &= \mu^2(\mu+\gamma)(\mu+\delta)\left((1+\epsilon_r)\phi L_0+\mu_v(1+L_0)\right)(1-R_0).
\end{aligned}$$

Solving for λ^* in (3.2.8) we have two roots

$$\lambda_1^* = \frac{-a_1+\sqrt{a_1^2-4a_2a_0}}{2a_2} \quad \text{and} \quad \lambda_2^* = \frac{-a_1-\sqrt{a_1^2-4a_2a_0}}{2a_2}.$$

Case 1 If $R_0 = 1$, then $a_0 = 0$, $a_1 > 0$, $a_2 > 0$.

$$a_2\lambda^{*2} + a_1\lambda^* = 0,$$
$$\lambda(a_2\lambda^* + a_1) = 0,$$

therefore

$$\lambda_1^* = 0, \quad \text{and} \quad \lambda_2^* = -\frac{a_1}{a_2} < 0.$$

$\lambda_1^* = 0$ corresponds to the disease free case and $\lambda_2^* = -\frac{a_1}{a_2}$ represent a non feasible equilibrium point.

Case 2 If $R_0 > 1$ then $a_0 < 0$, $a_2 > 0$. According to Descartes rule of signs, there is only one sign change in the coefficients of (3.2.8). So there is one unique positive equilibrium point, $\lambda_1^* > 0$. The root $\lambda_2^* < 0$ represent a non feasible equilibrium point.

Case 3 If $R_0 < 1$, then $a_0 > 0$, $a_1 > 0$, $a_2 > 0$. According to Descartes rule of signs, there is no change in sign from the coefficients of (3.2.8) and thus no positive equilibrium point.

Therefore, the only positive equilibrium point occurs when $R_0 > 1$ and corresponds to λ_1^*. We disregard $\lambda_2^* < 0$ since it leads to non-feasible solutions. We summarise the existence results in the following theorem:

3.2.5 Theorem. *The endemic equilibrium point exists only when* $R_0 > 1$.

3.3 Stability analysis of equilibrium points

3.3.1 Stability analysis of the disease free equilibrium point. According to Van den Driessche and Watmough (2002), the stability of the disease free equilibrium point is determined by the stability of the matrix $F - V$ given by

$$\begin{pmatrix} -(\mu+\gamma) & \beta(1-\epsilon_r)\eta L_0 & \beta(1-\epsilon_r)L_0 \\ \gamma & -(\mu+\delta) & 0 \\ 0 & \delta N & -\dfrac{(1+\epsilon_r)\phi L_0 + \mu_v(1+L_0)}{1+L_0} \end{pmatrix}.$$

The eigenvalues of $F - V$ are determined by solving the equation

$$|\,(F-V) - \zeta I\,| = \begin{vmatrix} -(\mu+\gamma)-\zeta & \beta(1-\epsilon_r)\eta L_0 & \beta(1-\epsilon_r)L_0 \\ \gamma & -(\mu+\delta)-\zeta & 0 \\ 0 & \delta N & -\left(\frac{(1+\epsilon_r)\phi L_0 + \mu_v(1+L_0)}{1+L_0}\right) - \zeta \end{vmatrix} = 0,$$

which gives the characteristic equation of the form

$$\zeta^3 + b_1\zeta^2 + b_2\zeta + b_3 = 0,$$

where;

$$b_1 = (\mu+\gamma)+(\mu+\delta)+\left(\frac{(1+\epsilon_r)\phi L_0+\mu_v(1+L_0)}{1+L_0}\right),$$
$$b_2 = (\mu+\gamma)(\mu+\delta)+(\mu+\gamma)\left(\frac{(1+\epsilon_r)\phi L_0+\mu_v(1+L_0)}{1+L_0}\right)+(\mu+\delta)\left(\frac{(1+\epsilon_r)\phi L_0+\mu_v(1+L_0)}{1+L_0}\right)$$
$$-\beta\epsilon_r\eta\gamma L_0,$$
$$b_3 = (\mu+\gamma)(\mu+\delta)\left(\frac{(1+\epsilon_r)\phi L_0+\mu_v(1+L_0)}{1+L_0}\right)(1-R_0).$$

Using the Routh-Hurwitz criterion, to determine the conditions for $Re(\zeta)<0$ for 3rd order polynomial we require

$$b_1>0, \quad b_3>0, \quad b_1b_2-b_3>0.$$

Clearly,

$$b_1>0,$$
$$b_3>0 \quad \text{if} \quad R_0<1$$

$$b_1b_2-b_3=\xi_1\xi_2(\xi_1+\xi_2)(1-R_0)+\xi_1\xi_2(\xi_1+\xi_2)\xi_4+\xi_3(\xi_1+\xi_2)(\xi_1+\xi_2+\xi_3),$$

where,

$$\xi_1=(\mu+\gamma), \quad \xi_2=(\mu+\delta), \quad \xi_3=\frac{(1+\epsilon_r)\phi L_0+\mu_v(1+L_0)}{1+L_0},$$
$$\xi_4=\frac{\beta(1-\epsilon_r)\gamma\delta N L_0(1+L_0)}{(\mu+\gamma)(\mu+\delta)\frac{(1+\epsilon_r)\phi L_0+\mu_v(1+L_0)}{1+L_0}}.$$

$$b_1b_2-b_3>0 \quad \text{if} \quad R_0<1.$$

Since all conditions for Routh-Hurwitz are satisfied, therefore all eigenvalues are negative or have negative real parts and the disease free equilibrium point is locally asymptotically stable when $R_0<1$ and unstable when $R_0>1$. This result suggests that R5 HIV can be cleared from the Langerhans cells population when the basic reproduction number is less than unity since an infected Langerhans cell will produce less than one new infected Langerhans cell and the virus will die out.

3.3.2 Stability analysis of the endemic equilibrium point. We shall use the centre manifold theory to investigate the stability analysis of the endemic equilibrium point for the case $R_0>1$. We consider equilibrium solutions of the model near the bifurcation point when $R_0=1$. Choosing β as a bifurcation parameter from the expression of R_0 as $\beta=\beta^*$, we obtain

$$\beta^*=\frac{(\mu+\gamma)(\mu+\delta)\left[\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0\right]}{(1-\epsilon_r)L_0\gamma\left[\eta\left[\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0\right]+N\delta(1+L_0)\right]}.$$

The Jacobian matrix for the system of equations (3.1.1)-(3.1.4) at the disease free equilibrium point when $\beta = \beta^*$ is

$$J(E_0, \beta^*) = \begin{pmatrix} -\mu & 0 & C_1 & C_2 \\ 0 & -(\mu+\gamma) & C_3 & C_4 \\ 0 & \gamma & -(\mu+\delta) & 0 \\ 0 & 0 & \delta N & -C_5 \end{pmatrix}, \tag{3.3.1}$$

where

$$\begin{aligned}
C_1 &= -\frac{(\mu+\gamma)(\mu+\delta)\left[\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0\right]}{\gamma\left[\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0\right]+N\delta(1+L_0)}, \\
C_2 &= -\frac{(\mu+\gamma)(\mu+\delta)\left[\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0\right]}{\gamma\eta\left[\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0\right]+N\delta(1+L_0)}, \\
C_3 &= \frac{(\mu+\gamma)(\mu+\delta)\left[\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0\right]}{\gamma\left[\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0\right]+N\delta(1+L_0)}, \\
C_4 &= \frac{(\mu+\gamma)(\mu+\delta)\left[\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0\right]}{\gamma\eta\left[\mu_v(1+L_0)+(1+\epsilon_r)\phi L_0\right]+N\delta(1+L_0)}, \\
C_5 &= \frac{(1+\epsilon_r)\phi L_0+\mu_v(1+L_0)}{1+L_0}.
\end{aligned}$$

The eigenvalues of the Jacobian matrix (3.3.1) are $\zeta_1 = -\mu$ and the rest are obtained from the characteristic equation

$$\begin{vmatrix} -(\mu+\gamma)-\zeta & C_3 & C_4 \\ \gamma & -(\mu+\delta)-\zeta & 0 \\ 0 & \delta N & -C_5-\zeta \end{vmatrix} = 0,$$

which gives

$$-\zeta\left[(\mu+\gamma)(\mu+\delta)+(\mu+\gamma)C_5+(\mu+\gamma)\zeta+C_5(\mu+\delta)+\zeta(\mu+\delta)+\zeta C_5+\zeta^2+C_3\gamma\right]=0$$

Therefore, $\zeta_2 = 0$ or $(\mu+\gamma)(\mu+\delta)+(\mu+\gamma)C_5+(\mu+\gamma)\zeta+C_5(\mu+\delta)+\zeta(\mu+\delta)+\zeta C_5+\zeta^2+C_3\gamma=0$ which reduces to

$$\zeta^2+\alpha_1\zeta+\alpha_2=0$$

where

$$\begin{aligned}
\alpha_1 &= (\mu+\gamma)+(\mu+\delta)+C_5>0, \\
\alpha_2 &= (\mu+\gamma)(\mu+\delta)+(\mu+\gamma)C_5+(\mu+\delta)C_5+C_3\gamma>0.
\end{aligned}$$

Thus, $\zeta_{3,4} = \dfrac{-\alpha_1 \pm \sqrt{\alpha_1^2-4\alpha_2}}{2}$ which have negative real parts.

Since $\zeta_2 = 0$ is a simple eigenvalue, we proceed to apply the center manifold theory to analyse the stability of the equilibrium point. We do this by calculating the left and right eigenvectors associated with the eigenvalue $\zeta_2 = 0$. To do this, we introduce the variables

$$L = x_1, \quad L_T = x_2, \quad L_I = x_3, \quad V_I = x_4$$

and rewrite the system of the equations (3.1.1) to (3.1.4) as

$$\frac{dx_1}{dt} = \pi - \beta^*(1-\epsilon_r)(x_4+\eta x_3)x_1 - \mu x_1 = f_1, \tag{3.3.2}$$
$$\frac{dx_2}{dt} = \beta^*(1-\epsilon_r)(x_4+\eta x_3)x_1 - (\mu+\gamma)x_2 = f_2, \tag{3.3.3}$$
$$\frac{dx_3}{dt} = \gamma x_2 - (\mu+\delta)x_3 = f_3, \tag{3.3.4}$$
$$\frac{dx_4}{dt} = \delta N x_3 - \frac{(1+\epsilon_r)\phi x_4 x_1}{1+x_1} - \mu_v x_4 = f_4. \tag{3.3.5}$$

The non-zero second derivatives of the system (3.3.2) to (3.3.5) evaluated at the disease free equilibrium are

$$\frac{\partial^2 f_1}{\partial x_1 \partial x_3} = \frac{\partial^2 f_1}{\partial x_3 \partial x_1} = -\beta^*(1-\epsilon_r)\eta, \quad \frac{\partial^2 f_2}{\partial x_4 \partial x_1} = \frac{\partial^2 f_2}{\partial x_1 \partial x_4} = \beta^*(1-\epsilon_r),$$
$$\frac{\partial^2 f_2}{\partial x_1 \partial x_3} = \frac{\partial^2 f_2}{\partial x_3 \partial x_1} = \beta^*(1-\epsilon_r)\eta, \quad \frac{\partial^2 f_1}{\partial x_4 \partial x_1} = \frac{\partial^2 f_1}{\partial x_1 \partial x_4} = -\beta^*(1-\epsilon_r),$$
$$\frac{\partial^2 f_4}{\partial x_1 \partial x_4} = \frac{\partial^2 f_4}{\partial x_4 \partial x_1} = -\frac{(1+\epsilon_r)\phi}{(1+L_0)^2}, \quad \frac{\partial^2 f_2}{\partial x_1 \partial \beta^*} = (1-\epsilon_r)(x_4+\eta x_3),$$
$$\frac{\partial^2 f_2}{\partial x_3 \partial \beta^*} = (1-\epsilon_r)\eta L_0, \quad \frac{\partial^2 f_2}{\partial x_4 \partial \beta^*} = (1-\epsilon_r)L_0.$$

The left eigenvector is computed using the equation $(J(E_0,\beta^*)^\top - \zeta I)v = \mathbf{0}$ where $v = (v_1, v_2, v_3, v_4)$, $\mathbf{0}$ is the zero matrix and $\top$ is the transpose of a matrix. Thus,

$$v = \left(0, \frac{\gamma}{\mu+\gamma}, 1, \frac{\beta^*(1-\epsilon_r)L_0\gamma(1+L_0)}{(\mu+\gamma)\left[(1+\epsilon_r)\phi L_0 + \mu_v(1+L_0)\right]}\right).$$

The right eigenvector is computed using the equation $(J(E_0,\beta^*) - \zeta I)w = \mathbf{0}$ where $w = (w_1, w_2, w_3, w_4)$

$$w = \left(\frac{\beta^*(1-\epsilon_r)\left[\eta L_0(1+\epsilon_r)\phi L_0 + \mu_v(1+L_0) + L_0\delta N\right]}{\mu(1+\epsilon_r)\phi L_0 + \mu_v(1+L_0)}, \frac{\mu+\delta}{\gamma}, 1, \frac{\delta N(1+L_0)}{(1+\epsilon_r)\phi L_0 + \mu_v(1+L_0)}\right).$$

To determine the type of transition exhibited at $R_0 = 1$, we compute the parameters a and b (Castillo-Chavez and Song, 2004) where

$$a = \sum_{k,i,j=1}^{n} v_k w_i w_j \frac{\partial^2 f_k}{\partial x_i \partial x_j}(x_0, 0)$$

and

$$b = \sum_{k,i=1}^{n} v_k w_i \frac{\partial^2 f_k}{\partial x_i \partial \beta^*}.$$

3.3.3 Computing a**.** We calculate 64 terms of the form $v_k w_i w_j \frac{\partial^2 f_k}{\partial x_i \partial x_j}(x_0, 0)$ where $k = n = 4$ and choose the non-zero terms.

$$a = v_2 w_3 w_1 \left(\frac{\partial^2 f_2}{\partial x_1 \partial x_3} + \frac{\partial^2 f_2}{\partial x_3 \partial x_1} \right) + v_2 w_4 w_1 \left(\frac{\partial^2 f_2}{\partial x_4 \partial x_1} + \frac{\partial^2 f_2}{\partial x_1 \partial x_4} \right) + v_4 w_1 w_4 \left(\frac{\partial^2 f_4}{\partial x_1 \partial x_4} + \frac{\partial^2 f_4}{\partial x_4 \partial x_1} \right),$$
$$a = -2\left(\vartheta_1 + \vartheta_2 + \vartheta_3\right) < 0.$$

where

$$\vartheta_1 = \frac{\gamma \beta^{*2} (1-\epsilon_r)^2 \left[\eta^2 L_0 \left((1+\epsilon_r)\phi L_0 + \mu_v (1+L_0)\right) + L_0 \delta N\right]}{(\mu+\gamma)\mu \left[(1+\epsilon_r)\phi L_0 + \mu_v (1+L_0)\right]},$$
$$\vartheta_2 = \frac{\gamma \delta N \beta^{*2} (1-\epsilon_r)^2 \left[\eta L_0 \left((1+\epsilon_r)\phi L_0 + \mu_v (1+L_0)\right) + L_0 \delta N\right]}{(\mu+\gamma)\mu \left[(1+\epsilon_r)\phi L_0 + \mu_v (1+L_0)\right]^2},$$
$$\vartheta_3 = -\frac{\gamma L_0 (1+L_0)^2 \beta^{*2} (1-\epsilon_r)^2 \left[\eta L_0 \left((1+\epsilon_r)\phi L_0 + \mu_v (1+L_0)\right) + L_0 \delta N\right] \delta N (1+\epsilon_r)\phi}{(\mu+\gamma)\mu \left[(1+\epsilon_r)\phi L_0 + \mu_v (1+L_0)\right]^3 (1+L_0)^2}.$$

3.3.4 Computing b**.** We calculate 16 terms of the form $v_k w_i \frac{\partial^2 f_k}{\partial x_i \partial \beta^*}(x_0, 0)$. We consider the non-zero terms which are

$$b = v_2 w_1 \frac{\partial^2 f_2}{\partial x_1 \partial \beta^*}(x_0, 0) + v_2 w_3 \frac{\partial^2 f_2}{\partial x_2 \partial \beta^*}(x_0, 0) + v_2 w_4 \frac{\partial^2 f_2}{\partial x_4 \partial \beta^*}(x_0, 0),$$
$$b = \frac{\gamma \beta (1-\epsilon_r)^2 \left[\eta L_0 \left((1+\epsilon_r)\phi L_0 + \mu_v (1+L_0)\right)\right] + L_0 \delta N}{(\mu+\gamma)\mu \left((1+\epsilon_r)\phi L_0 + \mu_v (1+L_0)\right)}$$
$$+ \frac{\gamma (1-\epsilon_r)\eta L_0}{(\mu+\gamma)} + \frac{\gamma \delta N (1+L_0)(1-\epsilon_r) L_0}{(\mu+\gamma)(1+\epsilon_r)\phi L_0 + \mu_v (1+L_0)} > 0.$$

Thus, we have $a < 0, b > 0$ that corresponds to condition (iv) of Theorem 2.2.4. Since $a < 0$ and $b > 0$, it follows that the system will undergo a transcritical bifurcation at $R_0 = 1$. The system exhibits a supercritical bifurcation which is a type of transcritical bifurcation. In a supercritical bifurcation there is an exchange of stability such that the disease free equilibrium looses its stability and the endemic equilibrium point becomes stable when $R_0 > 1$ (Britton, 2003). In this case all solutions starting inside Ω converge to the endemic equilibrium point and each infected Langerhans cell produces more than one new infected Langerhans cell and the virus persists in an individual. We summarise this result using the following theorem,

3.3.5 Theorem. *The endemic equilibrium point that exists when* $R_0 > 1$ *is locally asymptotically stable when* $R_0 > 1$.

Summary

We formulated an R5 HIV infection in Langerhans cells model and proved that region Ω was biologically feasible. We calculated the basic reproduction number and equilibrium points. Our system had a disease free equilibrium point and an endemic equilibrium point. Stability analysis of the two equilibrium

points was carried out. We used the Routh- Hurwitz criterion conditions to prove that the disease free equilibrium point is locally asymptotically stable when $R_0 < 1$ and unstable when $R_0 > 1$. Using the center manifold theory, we proved that the endemic equilibrium point was locally asymptotically stable when $R_0 > 1$. We also showed that when $R_0 = 1$, there was an exchange of stability between the disease free equilibrium point and endemic equilibrium point, a phenomenon known as supercritical bifurcation where the disease free equilibrium point lost its stability to the endemic equilibrium point.

4. Numerical Simulations

This chapter presents numerical simulations to enhance the understanding of the predictions of the analytical results. Data was obtained from published papers. The real data required to test the model predictions is not easy to obtain due to the nature of the study that requires data from human subjects and also the duration of the project. The prediction from the simulation are therefore theoretical in nature.

The initial conditions were assumed to be $1000mm^{-3}$ healthy Langerhans cells, no latently infected cells, $1mm^{-3}$ infected cell and a population of $0.001mm^{-3}$ virus meaning one virus was introduced in a population of $1000mm^{-3}$ healthy Langerhans cells. The model parameter values used in the numerical solutions and their sources are given in table 4.1.

In our numerical simulations, we first illustrate a scenario with no treatment. We use the no treatment scenario as a basis to introduce treatment. We use a hypothetical scenario where after 8 weeks an individual is diagnosed R5 HIV infected and immediately put on treatment with maraviroc. We investigate the effects of treatment in all the Langerhans cells population and the virus population at different treatment efficacies. In addition, we use a contour plot to check how the degradation rate for R5 HIV by the healthy Langerhans cells, ϕ and the efficacy of the maraviroc drug against R5 HIV, ϵ_r affect the basic reproduction number, R_0. We also plot a $3D$ surface plot that shows us the extent to which ϕ and ϵ_r reduces R_0 to values less than 1. We illustrate the simulation results using graphs which show the trends of each of the variables over a period of time.

Table 4.1: Table of parameter values

Parameter	Values	Units	References
π	[2,20]	$cells/day^{-1}$	Culshaw and Ruan (2000).
β	[0.0001,0.02]	day^{-1}	Blauvelt (2008).
μ	0.002	day^{-1}	assumed.
γ	0.035	day^{-1}	assumed.
δ	0.025	day^{-1}	assumed.
N	[200,1000]	$virions/day^{-1}$	Chirove and Lungu (2013).
ϕ	[2,9]	day^{-1}	Perelson and Nelson (1999).
μ_v	2.4	day^{-1}	Perelson et al. (1996)
η	$0 < \eta < 1$	day^{-1}	see text.
ϵ_r	[0,1]	day^{-1}	see text.

4.1 No treatment

We want to observe the impact of R5 HIV-infection on Langerhans cells in the absence of maraviroc. From figure (4.1), we observe that in the absence of treatment, the healthy Langerhans cells will start decreasing due to infection. Their decrease is associated with an immediate increase in latently infected Langerhans cells followed by the increase in R5 HIV and then the increase of infectious Langerhans cells.

The low level of healthy Langerhans cells is associated with the infectious populations reaching their peaks. The decrease in the infectious populations triggers a slight growth in healthy Langerhans cells at a later stage of the infection. Thus the figure suggests that Langerhans cells infection in the early stages

leaves a residual of HIV that could add to the viral load from other immune cells which collectively may overwhelm the immune system. It also suggests that an increase in the infectious Langerhans cells which decreases the healthy Langerhans cells may cause an individual to ultimately develop AIDS and die.

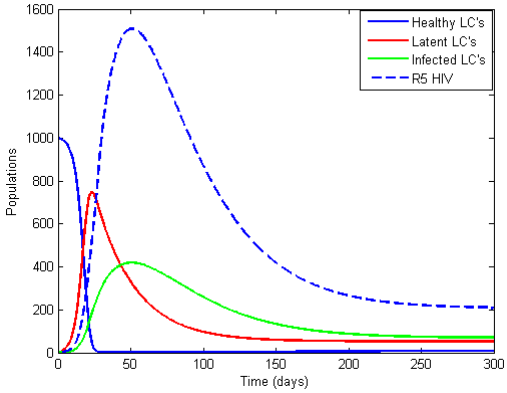

Figure 4.1: No treatment in both populations (Langerhans cell population and R5 HIV population)

4.2 Treatment with maraviroc

Having observed the trends with no treatment we also investigated the benefits which treatment with maraviroc has towards reducing the effects of Langerhans infection by R5 HIV.

Treatment with maraviroc at day 56 was introduced using five different efficacies with $\epsilon_r = 0.2, 0.5, 0.75, 0.9, 0.99$. Treatment was introduced at day 56 assuming the virus was detected in the body after 8 weeks of infection. 8 weeks of infection also fall under the early HIV infection phase. The choice of 56 days is purely hypothetical. We acknowledge that if one chooses a different starting time, the simulation values may change, but the qualitative behaviour of solutions is preserved. Figures (4.2) to (4.5) show the effects of these efficacies towards reducing R5 HIV infection. We observe in figure (4.2) that as the treatment efficacy increases, the number of healthy Langerhans cells starts increasing as well. In fact, a significant increase is observed when the efficacy of maraviroc is very high. In figures (4.3),(4.4) and (4.5), we notice that an increase in the treatment efficacy decreases the number of latently infected Langerhans cells, actively infected Langerhans cells and the virions respectively. In all the plots the drug is shown to be more effective when its efficacy is very high.

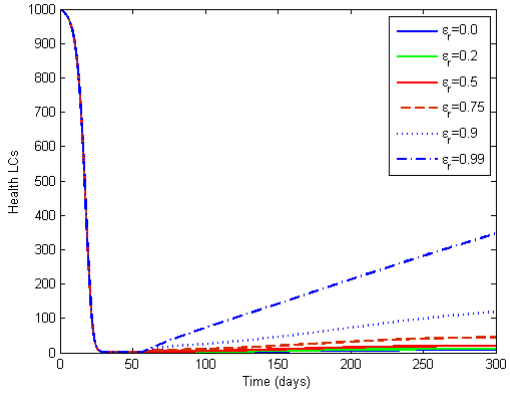

Figure 4.2: Treated healthy LCs

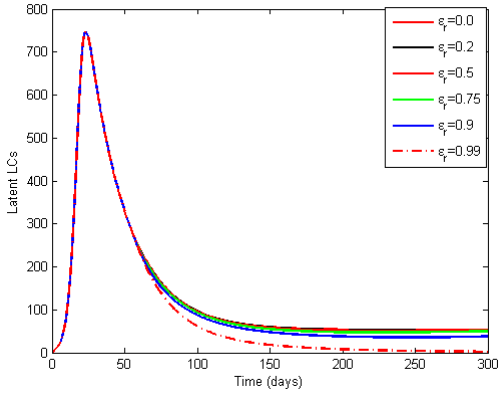

Figure 4.3: Treated latently infected LCs

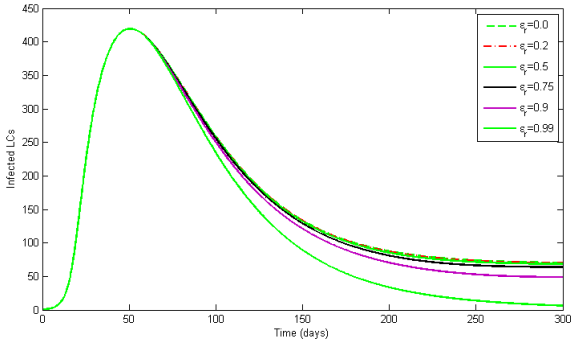

Figure 4.4: Treated actively infected LCs

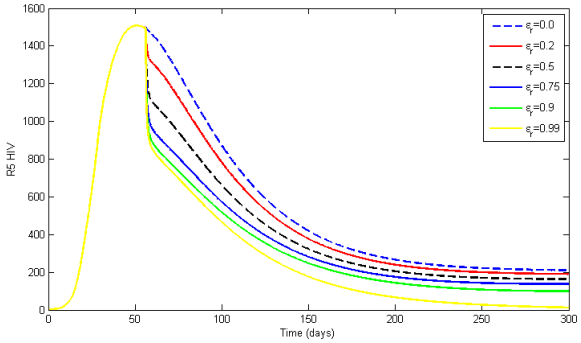

Figure 4.5: Treated R5 HIV

Increasing both the efficacy of maraviroc (ϵ_r) and the degradation rate of R5 HIV by Langerhans cells (ϕ) reduces the basic reproduction number (R_0) as shown in figure (4.6) and ultimately efficacy of maraviroc is the dominant strategy that significantly reduces R_0. The surface plot in (4.7) shows that it takes as much as 99% of maraviroc to reduce the value of R_0 to less than unity. Hence, this scenario suggests that any policy regarding the use of maraviroc should focus on improvement of maraviroc efficacy and R5 HIV degradation rate to achieve a significant control level of R5 HIV-infection in the Langerhans cells.

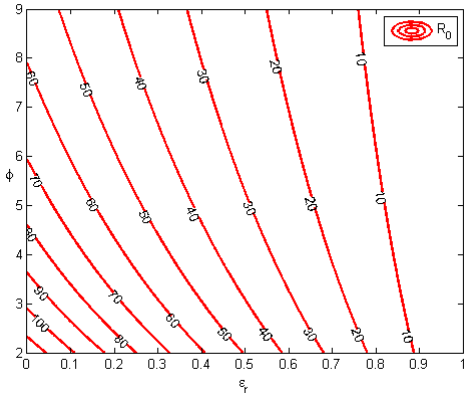

Figure 4.6: Contour

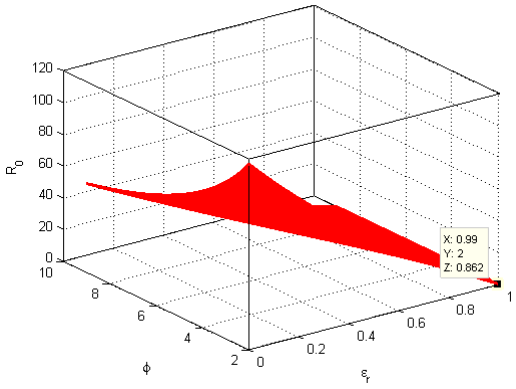

Figure 4.7: Surface

Our model revealed the existence of stability between the disease free equilibrium and the endemic equilibrium points through a supercritical bifurcation. Using the quadratic expression (3.2.8) we plotted the equilibrium points of infected Langerhans cells against the basic reproduction number. Figure (4.8) shows the presence of transcritical bifurcation at $R_0 = 1$. The figure shows that when $R_0 < 1$ then only one equilibrium point, the disease free equilibrium point exists. The figure also shows that when $R_0 > 1$, we have two equilibrium points; one which corresponds to the unstable disease free equilibrium point indicated by the dotted line and the other solid line corresponding to an R5 HIV endemic case.

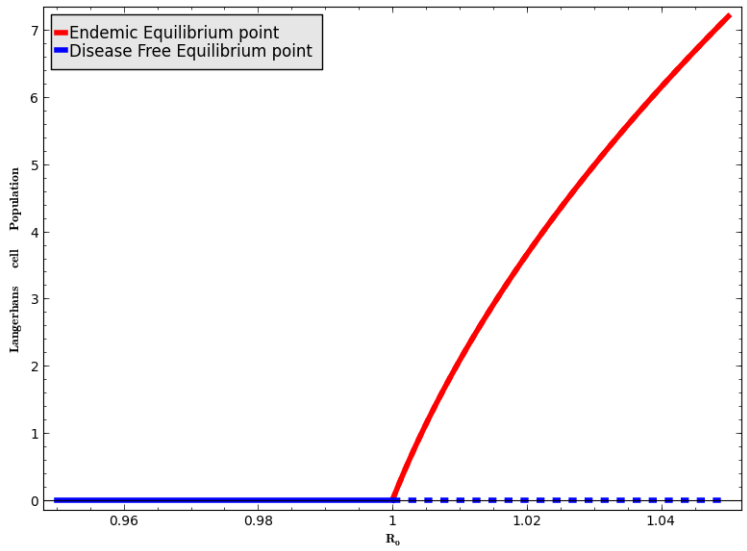

Figure 4.8: Supercritical bifurcation

4.3 Summary

This chapter has provided insights on the predictions of our analytical results. The numerical simulation results were presented using graphs that showed the trends of Langerhans cells populations and virus

population over a period of time. The results have helped us to make observations towards understanding of R5 HIV infection in the Langerhans cells without treatment and with treatment of maraviroc with different efficacies. We noticed that the viral load, which is the amount of R5 HIV in the blood increases with no treatment and is reduced when treatment of high efficacy is introduced.

5. Conclusion and Discussion of Results

5.1 Conclusion/Discussion of results

To investigate the effects of R5 HIV infection in the Langerhans cells we formulated a basic mathematical model capturing the dynamics of Langerhans cells and R5 HIV. We first proved that the state variables of the model were positive and bounded to make sure that the region which the model was analysed was biologically feasible. We then obtained the basic reproduction number and identified that it was influenced by secondary infections caused by the actively infected Langerhans cells and the free virus. The mathematical analysis carried out showed the existence of a disease free equilibrium point and an endemic equilibrium point. Threshold condition for stability of the equilibrium points were established through the use of the basic reproduction number. We showed that the disease free equilibrium point existed for values of $R_0 < 1$ and the the endemic equilibrium point existed when $R_0 > 1$. In the analysis we used the Routh-Hurwitz criterion to prove that the disease free equilibrium point was stable when $R_0 < 1$ and unstable when $R_0 > 1$. The center manifold theory was used to prove that the endemic equilibrium point is locally asymptotically stable when the basic reproduction number is greater than one.

Numerical simulations were carried out to enhance the understanding of the theoretical results. Firstly, we observed the effect of R5 HIV infection in the Langerhans cells with no treatment and the results showed that in the absence of treatment, the R5 virus infects the healthy Langerhans cells, leading to production of more viruses thus increasing the viral contribution of Langerhans cells. Secondly, we observed the effects of treatment of the R5 HIV infection in the Langerhans cells using maraviroc, an entry inhibitor which has generated considerable interests in research. We tested the maraviroc drug on the Langerhans cells and R5 HIV using different efficacies. The results showed that the increase in maraviroc efficacy reduces the population of the infectious Langerhans cells and R5 HIV and increases the population of the healthy Langerhans cells.

Our results suggest that giving R5 HIV infected individuals the maraviroc drug with high efficacy may help reduce the effects of R5 HIV in the early HIV infection period. However, the emergence of viral resistance is one of the greatest challenges in the treatment of HIV-infection and research according to Lieberman-Blum et al. (2008) showed that when maraviroc is used in combination with other antiretroviral agents it appears to be a promising agent for treatment.

Undoubtedly our model provided useful information and insights into the potential impact of reducing R5 HIV in the Langerhans cells using maraviroc.

5.2 Strengths and Weaknesses

The model analysis and numerical simulations helped us to see the impact that R5 HIV infection has on Langerhans cells in the absence of treatment and also the benefits of treatment with maraviroc towards the role of Langerhans cells in HIV infection.

Nevertheless, we were unable to obtain real data to test the model and hence the results in this work are purely theoretical and predictions are of the numerical solutions and purely qualitative. Some parameter values not readily found in published literature were estimated. The effects of changing these parameters needed to be checked using various techniques of uncertainty and sensitivity analysis.

5.3 Future work

This work can be developed further to include the use of real data in case it can be obtained. The model can also be extended to include other host cells infected by HIV with their specific types of treatment. The various immune cells and the virus evolve within different time scales so the study can be extended to capture these time scales.

Acknowledgements

First and foremost, I would like to thank the Almighty God for bringing me this far. It has not been easy but by his grace I am where I am today. My greatest appreciation goes to my supervisor Dr Faraimunashe Chirove who exposed me to the field of mathematical biology, the valuable guidance, advice, inspirations and for always being there throughout the period of the project.

Special thanks to the AIMS family, Prof. Neil Turok, Prof. Barry Green, Prof. Jeff Sanders, Jan Groenewald, A'eeda Rhodes for the opportunity to study at AIMS. Lecturers from all over the world for the wonderful mathematical science experience and for giving me a different perspective of science. Tutors, especially my essay tutor Eva Liliane Ujeneza, for the guidance and assistance during classes and assignments and not forgetting Igsaan, the kitchen staff and class of 2013/2014 for the cooperation and care which made my stay at AIMS bearable. I salute you all.

Lastly, my profound gratitude goes to my handsome man-George, thanks for your patience and support. My brothers Yollam, Wellington and Steve, my grandparents, aunties, uncles, cousins, sisters-in-law, close friends and relatives for the prayers, encouragement and best wishes. God bless you all.

References

AIDS.gov. Stages of HIV Infection, December 2013. URL http://aids.gov/hiv-aids-basics/just-diagnosed-with-hiv-aids/hiv-in-your-body/stages-of-hiv/.

P. Biswas, G. Tambussi, and A. Lazzarin. The status of co-receptor inhibition to counter HIV entry. pages 923–933, 2007.

A. Blauvelt. T-helper 17 cells in psoriatic plaques and additional genetic links between IL-23 and psoriasis. *Journal of Investigative Dermatology*, 128(5):1064–1067, 2008.

F. Brauer and C. Castillo-Chavez. *Mathematical models in population biology and epidemiology*. Springer,Verlag, Berlin-Heidelberg-New York, 2011.

N. F. Britton. *Essential mathematical biology*. Springer, Verlag London Limited, 2003.

C. Castillo-Chavez and B. Song. Dynamical models of tuberculosis and their applications. *Mathematical, Biosciences and Engineering*, 1(2):361–404, 2004.

F. Chirove and E. M. Lungu. Effects of replicative fitness on competing HIV strains. *Biosystems*, 113 (1):28–36, 2013.

R. V. Culshaw and S. Ruan. A delay-differential equation model of hiv infection of $CD4^+$ T-cells. *Mathematical biosciences*, 165(1):27–39, 2000.

D. C. Douek, M. Roederer, and R. A. Koup. Emerging concepts in the immunopathogenesis of AIDS. *Annual review of medicine*, 60:471,84, 2009.

N. Dusserre, C. Dezutter-Dambuyant, F. Mallet, P. Delorme, F. Philit, A. Ebersold, C. Desgranges, J. Thivolet, and D. Schmitt. In vitro HIV-1 entry and replication in Langerhans cells may clarify the HIV-1 genome detection by PCR in epidermis of seropositive patients. *Journal of investigative dermatology*, 99(5):120, 1992.

C. G. Figdor, Y. van Kooyk, and G. J. Adema. C-type lectin receptors on dendritic cells and langerhans cells. *Nature Reviews Immunology*, 2(2):77–84, 2002.

B. Hamzi, W. Kang, and A. J. Krener. Stabilization of discrete-time systems with a fold or period doubling control bifurcation. In *Proceedings of the 16th IFAC World Congress*, 2004.

C. HIV and H. LONG. AIDS. *Medical Care*, 916:874–7720, 2012.

INTELENCE. Types of HIV Treatment, March 2014. URL http://www.intelence.com/understanding-hiv-aids/hiv-treatments.

K. Katriena. What is the Langerhans cell?, 2014. URL http://www.wisegeek.com/what-is-the-langerhans-cell.htm#.

J. A. Levy. HIV pathogenesis: 25 years of progress and persistent challenges. *Aids*, 23(2):147–160, 2009.

S. S. Lieberman-Blum, H. B. Fung, and J. C. Bandres. Maraviroc: a CCR5-receptor antagonist for the treatment of HIV-1 infection. *Clinical therapeutics*, 30(7):1228–1250, 2008.

C. P. Locher, S. A. Witt, R. Kassel, N. L. Dowell, S. Fujimura, and J. A. Levy. Differential effects of R5 and X4 human immunodeficiency virus type 1 infection on $CD4^+$ cell proliferation and activation. *Journal of general virology*, 86(4):1171–1179, 2005.

W. R. Mbogo, L. S. Luboobi, and J. W. Odhiambo. Stochastic model for langerhans cells and hiv dynamics in vivo. *ISRN Applied Mathematics*, 2014(Article ID 594617,10 pages), 2014.

J. D. Murray. Mathematical biology I: An Introduction, vol. 17 of Interdisciplinary Applied Mathematics, Springer, New York, 2002.

A. S. Perelson and P. W. Nelson. Mathematical analysis of HIV-1 dynamics in vivo. *SIAM review*, 41 (1):3–44, 1999.

A. S. Perelson, A. U. Neumann, M. Markowitz, J. M. Leonard, and D. D. Ho. HIV-1 dynamics in vivo: virion clearance rate, infected cell life-span, and viral generation time. *Science*, 271(5255):1582–1586, 1996.

Queens University. Simple eigenvalue. URL http://www.mast.queensu.ca/~math211/m211oh/m211oh94.pdf.

S. Sastry. *Nonlinear systems: analysis, stability, and control*, volume 10. Springer, New York, 1999.

M. Sugaya, K. Loré, R. A. Koup, D. C. Douek, and A. Blauvelt. HIV-infected Langerhans cells preferentially transmit virus to proliferating autologous $CD4^+$ memory T cells located within Langerhans cell-T cell clusters. *The Journal of Immunology*, 172(4):2219–2224, 2004.

P. Van den Driessche and J. Watmough. Reproduction numbers and sub-threshold endemic equilibria for compartmental models of disease transmission. *Mathematical Biosciences*, 180(1):29–48, 2002.

J. G. Villavicencio-Pulido, I. Barradas, and J. C. Hernández-Gómez. A basic backward bifurcation model in epidemiology. *Applied Mathematical Sciences*, 7(107):5327–5340, 2013.

Wikipedia. Equilibrium point, February 2014a. URL http://en.wikipedia.org/wiki/Equilibrium_point.

Wikipedia. Stability analysis, April 2014b. URL http://en.wikipedia.org/wiki/Stability_theory.

H. Wu and A. A. Ding. Population HIV-1 dynamics in vivo: Applicable models and inferential tools for virological data from AIDS clinical trials. *Biometrics*, 55(2):410–418, 1999.