Mathematical Modelling of the Role of Mucosal Vaccine on the Within-host Dynamics of *Chlamydia trachomatis*

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Abstract

A mathematical model of the within-host replicative dynamics of *C. trachomatis* infection and its interactions with the immune system, in the presence of a mucosal vaccine, is presented. Our aim is to estimate the requisite efficacy of an efficacious mucosal vaccine that could promote a stable disease-free state *in vivo*. Sensitivity analysis was used to quantify how variability in the model parameters influence the value of the disease threshold $\mathcal{R}_0$. This shows that the two most important factors to be considered for achieving a disease-free state state *in vivo*, based on their influence on $\mathcal{R}_0$, are the efficacy of the *Chlamydia* vaccine, and the rate at which the humoral immune response protects healthy epithelial cells from infection. Numerical simulations of the model show that a vaccine with a minimum efficacy of 86% may be required for the *in vivo* control of *Chlamydia* burden. Such effective but imperfect *Chlamydia* vaccine could confer long-term protective immunity to genital *Chlamydia* infections. Conditions under which lower vaccine efficacies may suffice are also explored.

Keywords: *Chlamydia trachomatis* infection, Mucosal *Chlamydia* vaccine, Ordinary differential equations, Uncertainty and sensitivity analysis, Vaccine efficacy

1. Introduction

Chlamydia trachomatis is a gram-negative obligate intracellular bacteria that infects mucosal epithelial cells and causes ocular, genital, and respiratory infections [1, 2, 3, 4]. *C.trachomatis* genital infection is the most common bacterial sexually-transmitted infection worldwide, which infects over 105 million people annually (of which over a quarter of all incidences occur in the Americas, and more than two-thirds occur in developing countries) [5, 2, 1, 6, 7, 8]. In the United States alone, over 1.4 million cases occur annually since 2011 (with a 2.8% increase in incidence rate between 2013 and 2014 [9, 10], and with health costs amounting to about $2 billion annually) [11, 12]. *C. trachomatis* is still one of the major causes of morbidity and mortality worldwide [8]. It is often asymptomatic in women, in whom the primary site of infection is the columnar epithelium of the endocervix (the urethra in men) [1, 13, 14]. It is an important cause of tubal factor infertility (TFI), life-threatening ectopic pregnancy, endometritis, and mucopurulent endocervicitis in women [1, 9, 15, 16, 2, 8], epididymitis, infertility, and non-gonococcal urethritis (NGU) in men [1, 17], and neonatal pneumonia and opthalmia in infants born to genitally-infected women [9, 17, 18]. Its ocular serovars are responsible for trachoma, the world's leading cause of preventable blindness [7, 19, 2, 1]. The effects of maternal blindness due to trachoma, which include infant mortality, and the severe sequelae of

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genital chlamydial infections on reproduction, make *C. trachomatis* one of the most serious bacterial diseases of major global public health importance [19, 20, 6].

C. trachomatis has a unique and complex biphasic developmental cycle [1, 21]. It has two distinct morphological forms namely the infectious, extracellular, metabolically inert elementary bodies (EBs) and the non-infectious, intracellular, metabolically active reticulate bodies (RBs) [7, 1]. When infectious EBs attach to the host epithelium, they become internalised into a vacuole termed an inclusion [7, 21]. Within 2-6 hours post infection (PI), the internalised EBs differentiate into RBs. These RBs then exponentially replicate themselves by binary fission [7]. They re-differentiate and condense back into EB forms after several hours, and are, consequently, released to infect neighbouring host epithelial cells when the infected epithelial cell lyses (that is, their cell plasma membrane ruptures) [1, 7]. This cycle is referred to as the chlamydial developmental cycle (CDC).

The female reproductive tract has both innate and adaptive immune systems, which detect and respond to invading microbial pathogens [13, 1]. The innate immune system, which is non-specific, is responsible for the first line of defense against pathogens [13, 1]. When *Chlamydia* infects the epithelial cells of the endocervix of women, an intense inflammation occurs at the site of infection because the system is resisting *Chlamydia* by recruiting protective cells of the female reproductive tract to the infection site. These cells include neutrophils, dendritic cells, natural killer (NK) cells, B cells, T-lymphocytes, and inflammatory cells (such as macrophages), which cause the early release of pro-inflammatory cytokines (such as IFN-γ) and chemokines [3, 22, 13, 1]. The adaptive immune system, which is specific, is triggered in response to a foreign antigen [13]. After a naturally-occurring *Chlamydia* infection, protective immunity is developed by an infected individual [15, 3]. However, this immunity is imperfect, as it only offers partial protection against reinfection [3, 1, 6, 5], which often leads to severe immunopathology [5].

As noted by Schachter [15] and Loomis and Starnbach [3], the humoral arm of the immune response is believed to offer some protection against reinfection in an immune host. It does this by binding its mucosal and circulating antibodies to the pathogen. Antibodies also directly destroy the pathogen, thereby inactivating extracellular EBs [13, 15, 3]. Natural killer (NK) cells have also been commonly observed to lyse infected cells in a non-specific way [15, 3]. Neutralising or bactericidal antibodies secreted by B cells, and targeted against major outer membrane protein (MOMP) (an important antigen in the clearance and control of *Chlamydia* infections) play important roles in the clearance and control of *Chlamydia* infection [15, 3, 22]. However, neutralising bodies have not been confirmed to be able to proffer protective immunity in human [19, 15]. Their major role is in the enhancement of T helper-1 (Th1) activation [22, 13].

The cell-mediated immune system removes established infection [15, 3, 23]. Both immune responses appear to be aimed at the sites of infection [15, 3]. Although the mechanism behind the immuno-pathogenesis of *Chlamydia* infection is not fully understood [13], studies have, however, shown that the IFN-γ-secreting Th1-like $CD4^+$ T cell-mediated immune responses, as compared to the humoral immune response, plays the dominant role in protective immunity [19, 15, 3, 16, 22].

C. trachomatis infection can be treated with antibiotics, including azithromycin and doxycycline [17, 8, 18, 5]. Nevertheless, it is supposed that antimicrobial treatments reduce natural immunity to chlamydial infection and this facilitates the transmission of infection in the population [20, 24, 25, 1, 5, 26, 27]. Despite *azithromycin*'s in vitro efficacy of about 85-95% [28, 29], more treatment failures have been recorded in practice than originally thought, with failure rates ranging from 8% to 23% [30, 5, 31, 32, 33, 34, 35, 36]. Consequently, and due to the morbidity and high health costs associated with chlamydial infection, the development of a prophylactic *Chlamydia* vaccine has become crucial, and is considered to be the only feasible solution to the effective population level control of chlamydial infections (and its associated complications) [1, 5, 19, 20, 12, 37, 4].

Although no *Chlamydia* vaccine has been approved for use in humans, a number of candidate vaccines have been previously identified and tested in various delivery systems [16, 22, 12, 6, 38]. These candidate vaccines are largely based on the use of defined recombinant proteins [5]. Tested vaccine candidates include inactivated, live whole organisms, subunit vaccines, various chlamydial antigens, recombinant proteins and peptide vaccines [16, 22, 39, 5, 5, 12, 40, 41, 38]. The types of potential anti-*Chlamydia* vaccines recommended by the World Health Organisation (WHO) are those with high efficacy and that confer both sterilising and (long-term) protective immunity [42, 41, 4, 5]. Such a vaccine mimics the natural immune response to the

infection, while not inducing the severe inflammatory reactions often associated with *Chlamydia* infection [1]. Correlates of a vaccine that will induce protective immunity are primarily IFN-γ-secreting CD4+ T cells and accessory antibodies of IgA and IgG isotype response in the associated mucosal regions of the genital areas [43]. It has been suggested that for the development of such an efficacious chlamydial vaccine, more effective delivery systems need to be advanced, and effective immunomodulation should be used [16, 12, 1, 6, 39]. This is because effective delivery systems are expected to boost the induction of adequate levels of mucosal T-cells and antibody responses that mediate long-term protective immunity [16, 12]. A first-in-human clinical trial of an adjuvanted intramuscular *Chlamydia* vaccine coupled with some intranasal boosts based on a recombitant protein unit (CTH522) was recently assessed for safety and humoral immunogeneity [44]. The vaccine, which induced high serum neutralising antibodies and cell-mediated immune responses, is very promising and probably the future of an efficacious *Chlamydia* vaccine, but it has not been assessed for a long-term protective immunity. None of the candidate vaccines have been assessed for efficacy, with efficacy here defined as "the extent to which a specific intervention, procedure, regimen, or service produces a beneficial result under ideal conditions. Ideally, the determination of efficacy is based on the results of a randomized controlled trial" [45]. Studies have shown that *Chlamydia* vaccines delivered *via* mucosal routes are promising (as they induce high levels of *Chlamydia*-specific IFN-γ, and consequently, an enhanced protective immunity) [16, 12, 22, 2, 6]. However, while most *Chlamydia* vaccine trials have only evaluated protective immunity up to 4 weeks post vaccination [41], a recent experimental study by Stary *et al.* [2], which constitutes a major advancement in *C. trachomatis* immunobiology [37], have been able to identify some bio-profiles of a vaccine that can confer long-lived protective immunity [12, 41, 37, 2].

Stary *et al.* [2] report that mucosal vaccination of mice with ultraviolet light-inactivated *C. trachomatis* conjugated to charge-switching synthetic adjuvant nanoparticles (UV-Ct-cSAPs) brought about a robust *C. trachomatis*-specific antibody response that was equivalent to that elicited by *C. trachomatis* infection. The UV-Ct-cSAPs vaccine induces a wave of effector T cells (T_{EFF}) which seeded the uterine mucosa during the first week after vaccination. These T_{EFF} then established tissue-resident memory cells (T_{RM}) (in both resting and inflamed mucosal surfaces) thereafter, which persisted for at least six months even in the absence of local *Chlamydia* antigens. These cells were also referred to as a first wave of mucosal-tropic memory cells. Non-mucosal vaccines do not induce this first wave.

There was also a second wave of vaccine-induced circulating memory T cells (T_{CM}) which preferentially resided in blood and lymphoid tissues, from where they survey the body for *Chlamydia* antigens. The concentration of these T_{CM} is more than that of T_{RM}. These T_{CM} were produced irrespective of the mucosal route. They however do not traffic to resting uterine mucosa. In the absence of pre-existing T_{RM}, they were slow to access the uterus when vaccinated mice were rechallenged with *C. trachomatis* via the uterus. However, in mucosal-vaccinated mice, upon *Chlamydia* rechallenge, uterine-T_{RM} instantly responds to chlamydial infection and they initiate the speedy recruitment of *Chlamydia*-specific T_{CM} [2] to peripheral tissues, including uterine mucosa. For optimal clearance of *C. trachomatis*, T_{RM} must be established [2, 26], otherwise, the clearance would be sub-optimal even in the presence of abundant circulating memory cells. The two waves of memory T cells are however crucial to optimal clearance of *C. trachomatis* infection. Interestingly, when humanised mice (mice that have been genetically reconstituted with a human immune system) who were vaccinated (*via* mucosal routes) with UV-Ct-cSAP were rechallenged with intrauterine *C. trachomatis* infection, a vigorous mucosal T_H1 response that cleared *C. trachomatis* infection was elicited. This suggests that such a vaccine may also elicit such protective immunity in humans [2].

Mathematical models have been used to study the within-host dynamics of *Chlamydia trachomatis* infection and the associated immune responses (see [23, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 25, 56], and some of the references therein). In particular, Wilson [46] developed a deterministic mathematical model for chlamydial infections, that implicitly incorporates the immune response. Craig *et. al.* [51] used a mathematical model and experimental data to estimate the percentage of target (healthy) host epithelial cells that can be infected during the course of a typical chlamydial infection. Hoare *et. al.* [52] developed a model of the CDC which attempts to explain the mid-to-late stages of the cycle, thereby proving some hypothesis on chlamydial developmental events. Using delay differential equations, Burns *et al.* [53] estimated parameters associated with various aspects of the CDC. Akinlotan *et. al.* [56] used optimal control theory tools to investigate the effects of different treatment combinations on the within-host dynamics of chronic genital

chlamydial infections characterised by the presence of IFN-γ-induced chlamydial persistence.

Different aspects of the immune response to, and spatial progression of, chlamydial infections have also been modelled [46, 48, 54, 55, 25, 57, 58, 59, 60, 61, 56]. Using a deterministic approach, Wilson *et al.* [23] investigated the response of the cell-mediated arm of the immune system over the CDC in the control of chlamydial infections, while also tracking the number of chlamydial particles within infected host cells over the CDC. Although some within-host models for assessing the impact of a vaccine have been developed and used in the literature (such as the in-host malaria model developed by Niger and Gumel [62]), no such model has been designed for a potential *C. trachomatis* vaccine.

This study extends prior *Chlamydia* within-host modelling studies by theoretically assessing the potential role of an efficacious anti-*Chlamydia* vaccine on the within-host dynamics of *C. trachomatis* and prognosis of genital chlamydial infection. Since vaginal chlamydial shedding is positively correlated with the development of the severe sequelae of the upper genital tracts such as PID, of which the evaluation of the latter has been proposed to be the most robust endpoint for the evaluation of a chlamydial vaccine efficacy [26], we hypothesise that an efficacious *Chlamydia* vaccine will reduce or eliminate vaginal chlamydial shedding and population-level transmission consequently [26, 63]. This investigation uses a prototype vaccine that induces similar protective immunity to that described by the study of Stary *et al.* [2]. The paper is outlined as follows. In Section 2, a deterministic model for the within-host dynamics of *C. trachomatis* infection in the presence of a mucosal vaccine is presented. Basic properties of the model and its stability analyses are presented in Sections 2.1 and 2.2, respectively.

Basic properties of the model, including the positivity of solutions, and the definition of a positively invariant and attracting region for the model, are presented in Section 2.1. In Section 2.2, the existence of a unique Chlamydia-free equilibrium is shown. We also show that the model system (1) has a globally asymptotically stable *Chlamydia*-free equilibrium (CFE) whenever the basic reproduction number $\mathcal{R}_0 \leq 1$, and a unique *Chlamydia*-present equilibrium (CPE) when $\mathcal{R}_0 > \mathcal{R}_0^c > 1$, where $\mathcal{R}_0^c$ is a threshold called the *critical* $\mathcal{R}_0$. In Section 3.1, the uncertainty and sensitivity analysis of the basic reproduction number $\mathcal{R}_0$ to selected model predictors is presented. Results of the numerical simulations of the model are presented in Section 3.3. In this section, the requisite efficacy of an efficacious mucosal *Chlamydia* vaccine that could promote a stable disease-free state was investigated. The critical vaccine efficacy was estimated in Section 3.2. The discussion is presented in Section 4.

2. Model Formulation

The model to be developed is that of the dynamics of *C. trachomatis* within the body of an infected host subject to a mucosal anti-*Chlamydia* vaccine. The model, which builds on the model presented by Sharomi and Gumel [55], is designed as follows. Let $H_e(t)$ represent the concentration of healthy epithelial cells, $H_h(t)$, the concentration of healthy epithelial cells protected by the humoral immune response against EB attachment, and $I(t)$, the concentration of infected epithelial cells. Furthermore, let $T_r(t)$ and $T_c(t)$ represent the concentrations of *Chlamydia*-specific mucosal resident memory T cells ($\mathrm{T_{RM}}$) and *Chlamydia*-specific circulating memory T cells ($\mathrm{T_{CM}}$), respectively. Let $F(t)$ be the concentration of IFN-γ molecules secreted by T cells, $E_b(t)$ be the concentration of chlamydial elementary bodies, and $R_b(t)$ be the concentration of reticulate bodies.

The model for the in-host dynamics of *C. trachomatis*, subject to a mucosal anti-*Chlamydia* vaccine, is given by the following non-linear system of differential equations:

$$
\begin{aligned}
\frac{\mathrm{dH}_e}{\mathrm{dt}} &= \Pi_h + \omega H_h - \gamma(1-\varepsilon_v)H_e E_b - \phi\varepsilon_h H_e - \mu_h H_e,\\
\frac{\mathrm{d}H_h}{\mathrm{dt}} &= \phi\varepsilon_h H_e - \omega H_h - \mu_h H_h,\\
\frac{\mathrm{d}I}{\mathrm{dt}} &= \gamma(1-\varepsilon_v)H_e E_b - \kappa I - \rho I F,\\
\frac{\mathrm{d}T_r}{\mathrm{dt}} &= \varepsilon_r \Lambda_v + \tau_1 F T_r - \mu_t T_r,\\
\frac{\mathrm{d}T_c}{\mathrm{dt}} &= \varepsilon_c \Omega_v + \tau_2 F T_c - \mu_c T_c,\\
\frac{\mathrm{d}F}{\mathrm{dt}} &= \psi_1 T_r E_b + \psi_2 T_c E_b - \rho I F,\\
\frac{\mathrm{d}E_b}{\mathrm{dt}} &= N_E \kappa I - \gamma(1-\varepsilon_v)H_e E_b - \varepsilon_a \alpha E_b - \mu_e E_b,\\
\frac{\mathrm{d}R_b}{\mathrm{dt}} &= N_{R1}\kappa I + N_{R2}\rho I F - \mu_e R_b.
\end{aligned}
\tag{1}
$$

Healthy epithelial cells are replenished by non-differentiated stem cell precursors [64], at a rate Π_h, and are naturally protected against EB attachment by the humoral immune response at a rate ϕ [23, 55], with efficacy $0 < \varepsilon_h \leq 1$ (where $\varepsilon_h = 0$ means a totally ineffective humoral immune response, and $\varepsilon_h = 1$ represents a perfect humoral immune response). This immunity is assumed to wane at a rate ω. EBs infect unprotected healthy epithelial cells at a rate γ. The presence of a *Chlamydia* vaccine is expected to boost the body's defense mechanism (*via* antibody blocking) against *Chlamydia* infection. Thus, it is assumed that the number of newly-infected epithelial cells depends largely on the efficacy of the vaccine (with a 100% vaccine efficacy implying that new epithelial cell infections are prevented). The efficacy of the vaccine is represented by $0 < \varepsilon_v \leq 1$) (where $\varepsilon_v = 0$ means a totally ineffective vaccine, and $\varepsilon_v = 1$ represents a perfectly efficacious vaccine). The natural mortality rate of epithelial cells is μ_h. We assume that the *Chlamydia* vaccine works by reducing the probability of host infection by an (infectious) EB form of *Chlamydia*.

C. trachomatis infection triggers the rapid release of cytokines (IFN-γ in particular) by the tissue-$\mathrm{T_{RM}}$ and $\mathrm{T_{CM}}$ [2, 65, 13, 1, 3]. Thus, it is plausible to assume that the rate of production of IFN-γ is proportional to the number of EB forms and the concentration of $\mathrm{T_{RM}}$ and $\mathrm{T_{CM}}$. The production rates of IFN-γ by $\mathrm{T_{RM}}$ and $\mathrm{T_{CM}}$ are ψ_1 and ψ_2, respectively. In an uninfected host, the genital tract mucosa contains relatively few lymphocytes. Thus, the recruitment of circulating lymphocytes is a very important component of the immune response [1]. The concentration of *Chlamydia*-specific $\mathrm{T_{CM}}$ has been observed to be significantly higher than that of *Chlamydia*-specific (tissue) $\mathrm{T_{RM}}$ [2]. Thus, it is assumed that the rate of production of $\mathrm{T_{CM}}$ (Ω_v) is greater than the rate of production of $\mathrm{T_{RM}}$ (Λ_v). That is, $\Omega_v > \Lambda_v$. Furthermore, it is assumed that there are are vaccine-induced increases in the production of *Chlamydia*-specific $\mathrm{T_{RM}}$ and $\mathrm{T_{CM}}$, which are accounted for by the modification parameters $\varepsilon_r > 1$ and $\varepsilon_c > 1$, respectively. The tissue-$\mathrm{T_{RM}}$ confers substantial protection against *Chlamydia* infection even when the influx of $\mathrm{T_{CM}}$ is impeded [2]. Thus, it is assumed that $\varepsilon_r > \varepsilon_c$. Cytokines, such as IFN-γ, account for the enhancement of cellular proliferation [13, 64]. Thus, there is an IFN-γ-induced proliferation of T cells - $\mathrm{T_{RM}}$ (T_r) and $\mathrm{T_{CM}}$ (T_c), represented by the rates τ_1 and τ_2, respectively. The natural mortality rates of $\mathrm{T_{RM}}$ and $\mathrm{T_{CM}}$ are μ_t and μ_c, respectively. We assume that they are equal, thus, $\mu_t = \mu_c$.

Infected epithelial cells lyse (after maturation of their intracellular inclusions) at a rate κ, and are lysed/destroyed prematurely (that is, before the CDC is completed) by IFN-γ at a rate ρ. Since these processes occur at a much higher rate than the 'natural' death rate of epithelial cells (see Table 3), we assume that these processes account fully for the death/destruction of infected cells. While antibodies are involved in the signalling of macrophages for the engulfment of bound pathogen [46], IFN-γ activation of macrophages empowers them more to destroy phagocytosed EBs [66, 1]. Thus, it is assumed that there is a vaccine-induced increase in the rate at which antibodies destroy phagocytosed EBs. This is accounted for by the modification parameter $1 < \varepsilon_a \leq 2$. The rate at which macrophages engulf free extracellular EB forms

is α. The natural mortality rates of EBs and RBs are μ_e and μ_r, respectively. Since within-cell chlamydial replication is inhibited at the RB stage in the presence of IFN-γ [15, 66], it is assumed that there will be an IFN-γ-induced increase in the number of RBs that will be released on infected epithelial cell lysis [66]. It is expected that the presence of IFN-γ will lead to a marked decrease in the amount of EB forms that will be released from IFN-γ-induced cell lysis [66]. Furthermore, it is assumed that infected cells in the advanced stage of the CDC mainly contain EBs [23]. Hence, the number of EBs released when infected cells lyse (N_E) is greater than both the number of RBs released when "mature" infected cells lyse (N_{R1}) and the number of RBs released when infected cells lyse due to the inhibitory action of IFN-γ on the CDC (N_{R2}). That is, $N_E > N_{R1}$ and $N_E > N_{R2}$. However, if the inhibitory effect of IFN-γ on the CDC is very potent (which is one of the goals of a potentially effective anti-*Chlamydia* vaccine), it can be expected that $N_{R2} > N_E$ and $N_{R2} > N_{R1}$.

State Variables	**Description:** Concentration of
$H_e(t)$	Healthy epithelial cells
$H_h(t)$	Epithelial cells protected by humoral immune response
$I(t)$	Infected epithelial cells
$T_r(t)$	Resident memory T-cells (T_{RM})
$T_c(t)$	Circulating memory T-cells
$F(t)$	IFN-γ molecules produced by the cell-mediated immune response
$E_B(t)$	Chlamydial elementary bodies
$R_B(t)$	Chlamydial reticulate bodies

Table 1: Description of state variables of the model (1).

The new within-host model system (1) is an extension of the within-host *Chlamydia* models by Sharomi and Gumel [55]. In particular, in addition to the incorporation of the effects of a potentially efficacious mucosal *Chlamydia* vaccine, the model system (1) extends the model in [55] by, *inter alia*,

(i) adding a new compartment for the dynamics of *Chlamydia*-specific resident memory T cells (T_r);
(ii) adding a new compartment for the dynamics of *Chlamydia*-specific circulating memory T cells (T_c);
(iii) allowing for the proliferation of resident memory T cells (at a rate τ_1), and circulating memory T cells (at a rate τ_2);
(iv) allowing for a vaccine-induced (additional) protective immunity against infection of healthy epithelial cells by *Chlamydia* via antibody blocking (binding of mucosal and circulating antibodies to EBs, thereby neutralising the antigen of some EBs and blocking their ability to enter the mucosa [13, 15, 3]), modelled by the term $\gamma(1-\varepsilon_v)H_eE_b$;
(v) adding a compartment for the inhibitory action of IFN-γ (F).

A schematic representation of the above dynamics is presented in Figure 1. The description of the state variables and parameters of the model are shown in Tables 1 and 2 respectively. In the section to follow, we give the basic properties of model (1).

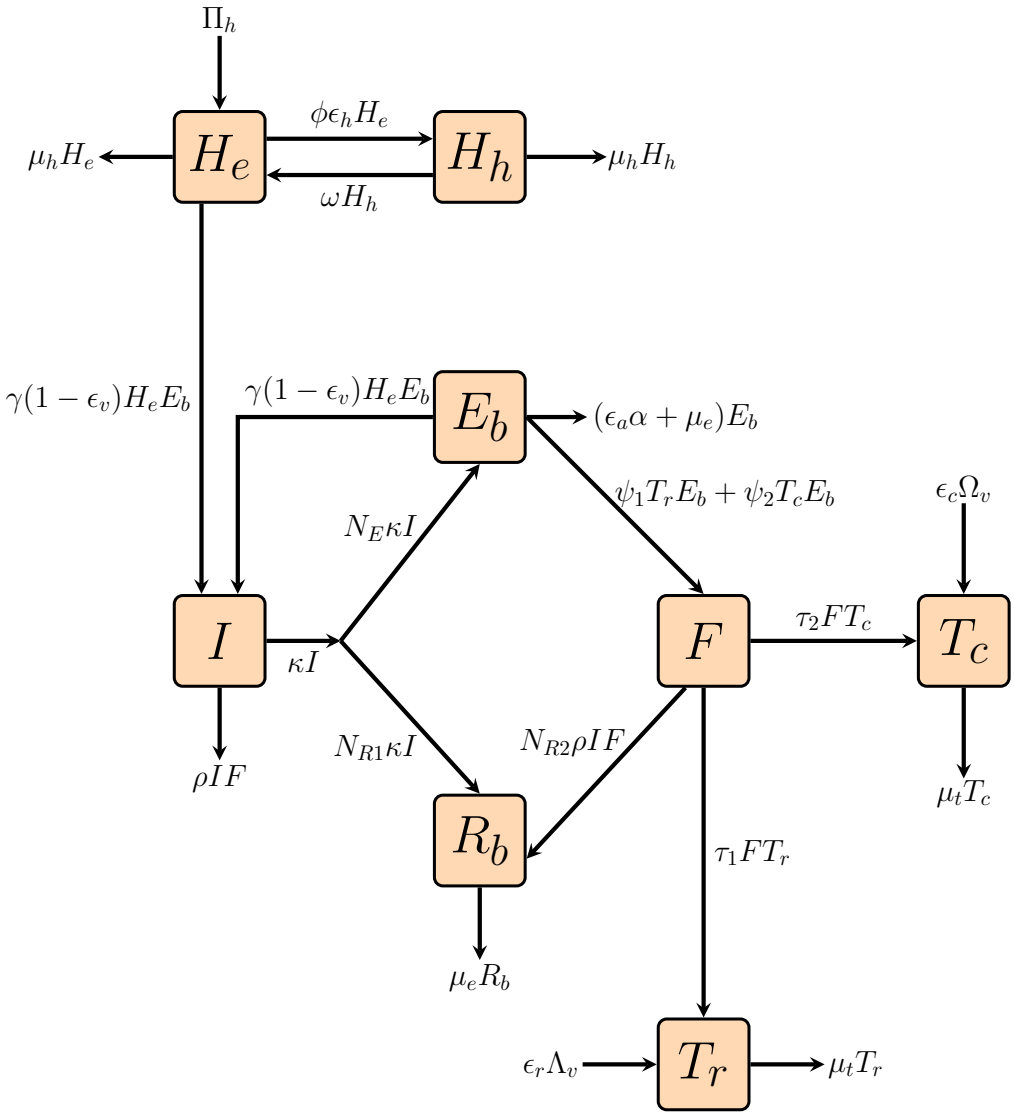


Figure 1: A schematic representation of model (1).

Parameters	Description
Π_h	Rate of replenishment of healthy epithelial cells
γ	Effective contact rate between healthy epithelial cells and EB forms
ϕ	Rate of protection of healthy epithelial cells by the humoral immune response
ω	Waning rate of the protection of healthy epithelial cells by humoral immune response
Λ_v	Production rate of resident memory T cells
Ω_v	Production rate of circulating memory T cells
κ	Rate at which infected cells lyse/burst
ρ	Rate at which IFN-γ lyse/destroy infected cells
α	Rate of macrophage engulfment of free extracellular EB forms
ε_v	Efficacy of vaccine
ε_h	Efficacy of the humoral response in protecting healthy epithelial cells
ε_r	Modification parameter accounting for vaccine-induced increase in the production of *Chlamydia*-specific T_{RM}
ε_c	Modification parameter accounting for vaccine-induced increase in the production of *Chlamydia*-specific T_{CM}
ε_a	Modification parameter accounting for vaccine-induced increase in the EB-engulfment rate of antibodies
τ_1	IFN-γ-induced proliferation rate of resident memory T cells
τ_2	IFN-γ-induced proliferation rate of circulating memory T cells
ψ_1	Vaccine-induced increase in production rate of IFN-γ by resident memory T cells
ψ_2	Vaccine-induced increase in production rate of IFN-γ by circulating memory T cells
μ_h	Natural mortality rate of host epithelial cells
μ_t	Natural mortality rate of resident memory T-cells
μ_c	Natural mortality rate of circulating memory T-cells
μ_e	Natural mortality rate of EB forms
μ_r	Natural mortality rate of RB forms
N_E	Number of EBs released on lysis of infected cells
N_{R1}	Number of RBs released on lysis of infected cells
N_{R2}	Number of RBs released on lysis of infected cells due to IFN-γ's inhibitory action on the CDC

Table 2: Description of the parameters of the model (1). T_{RM} (or T_{CM}) means resident (or circulating) memory cells.

2.1. Basic Properties

2.1.1. Positivity of solutions.

It is crucial to prove that all the state variables of the model (1) subjected to positive initial conditions remain non-negative for all time $t > 0$. This is to certify that the model system is biologically meaningful.

Theorem 2.1. *Given that the initial values of the state variables of the model (1) are non-negative, the model does not predict negative values for the state variables at any future time.*

Proof. Let $H_e(0) > 0$, $H_h(0) \geq 0$, $I(0) \geq 0$, $T_r(0) \geq 0$, $T_c(0) \geq 0$, $F(0) \geq 0$, $E_b(0) \geq 0$, and $R_b(0) \geq 0$.

Also let $\hat{t} = \sup\{t > 0 : H_e(t) > 0, H_h(t) > 0, I(t) > 0, T_r(t) > 0, T_c(t) > 0, F(t) > 0, E_b(t) > 0, R_b(t) > 0\} \in [0, t]$.

From the first equation of (1), we have

$$\frac{\mathrm{d}}{\mathrm{d}t}\left(H_e(t)\exp\left\{(\phi\varepsilon_h+\mu_h)t+\int_0^t(\gamma(1-\varepsilon_v)E_b(\theta)-\tau_1F(\theta))\mathrm{d}\theta\right\}\right)$$
$$=(\Pi_h+\omega H_h)\exp\left\{(\phi\varepsilon_h+\mu_h)t+\int_0^t(\gamma(1-\varepsilon_v)E_b(\theta)-\tau_1F(\theta))\mathrm{d}\theta\right\}.$$

This implies that

$$H_e(\hat{t})\exp\left\{(\phi\varepsilon_h+\mu_h)\hat{t}+\int_0^{\hat{t}}(\gamma(1-\varepsilon_v)E_b(\theta)-\tau_1F(\theta))\mathrm{d}\theta\right\}-H_e(0)$$
$$=\int_0^{\hat{t}}(\Pi_h+\omega H_h)\exp\left\{(\phi\varepsilon_h+\mu_h)\eta+\int_0^{\eta}(\gamma(1-\varepsilon_v)E_b(\theta)-\tau_1F(\theta))\mathrm{d}\theta\right\}\mathrm{d}\eta.$$

Thus,

$$H_e(\hat{t})=H_e(0)\exp\left\{-\left((\phi\varepsilon_h+\mu_h)\hat{t}+\int_0^{\hat{t}}(\gamma(1-\varepsilon_v)E_b(\theta)-\tau_1F(\theta))\mathrm{d}\theta\right)\right\}$$
$$+\exp\left\{-\left((\phi\varepsilon_h+\mu_h)\hat{t}+\int_0^{\hat{t}}(\gamma(1-\varepsilon_v)E_b(\theta)-\tau_1F(\theta))\mathrm{d}\theta\right)\right\}$$
$$\times\int_0^{\hat{t}}(\Pi_h+\omega H_h)\exp\left\{(\phi\varepsilon_h+\mu_h)\eta+\int_0^{\eta}(\gamma(1-\varepsilon_v)E_b(\theta)-\tau_1F(\theta))\mathrm{d}\theta\right\}\mathrm{d}\eta$$
$$>0.$$

Similarly, it can be shown that $H_h(t)\geq 0$, $I(t)\geq 0$, $T_r(t)\geq 0$, $T_c(t)\geq 0$, $F(t)\geq 0$, $E_b(t)\geq 0$, and $R_b(t)\geq 0$ for any time $t>0$. Hence, every solution of the model system (1) will always be positive for all non-negative initial conditions. □

2.1.2. Invariant regions.

The model (1) is analysed in an apposite biologically feasible region $\mathcal{D}_2$. The model system (1) is shown to be dissipative, that is, solutions of the model are uniformly bounded in a subset $\mathcal{D}_2$ of $\mathbb{R}^8_+$.

Lemma 2.2. *The region $\mathcal{D}_2$, defined by the compact set*
$\mathcal{D}_2=\{(H_e(t),H_h(t),I(t),T_r(t),T_c(t),F(t),E_b(t),R_b(t))\in\mathcal{D}_1 : H_e\leq H_e^*, H_h\leq H_h^*, I\geq 0, T_r\geq 0, T_c\geq 0, F\geq 0, E_b\geq 0, R_b\geq 0\}$,
where
$\mathcal{D}_1=\Big\{(H_e(t),H_h(t),I(t),T_r(t),T_c(t),F(t),E_b(t),R_b(t))\in\mathbb{R}^8_+ : P(t)\leq\frac{\Pi_h}{\mu_h}, T_r(t)\leq m_1, T_c(t)\leq m_2,$
$I(t)\geq 0, F(t)\geq 0, E_b(t)\geq 0, R_b(t)\geq 0\}$;
$m_1=T_r(t)\leq(T_r(0)+c_1)c_2$; $c_1=\int_0^t\varepsilon_r\Lambda_v\exp\left\{-\tau_1\int_0^{\eta}F(\theta)\,d\theta\right\}d\eta$; $c_2=\exp\left\{\tau_1\int_0^t F(\theta)\,d\theta\right\}$,
and
$m_2=T_c(t)\leq(T_c(0)+c_3)c_4=m_2$; $c_3=\int_0^t\varepsilon_c\Omega_v\exp\left\{-\tau_2\int_0^{\eta}E_b(\theta)\,d\theta\right\}d\eta$; $c_4=\exp\left\{\tau_2\int_0^t E_b(\theta)\,d\theta\right\}$,
is positively invariant and attracting for the model (1) with initial conditions in $\mathbb{R}^8_+$.

Proof. Let $P(t)=H_e(t)+H_h(t)$. Since all the parameters and state variables of model (1) are non-negative for all time $t\geq 0$, then from the first equation of model (1),

$$\frac{\mathrm{d}P(t)}{\mathrm{d}t}\leq\Pi_h-\mu_hP(t).$$

Using a standard comparison theorem by Lakshmikantham *et al.* [67], it can be shown that

$$P(t) \leq P(0)e^{-\mu_h t} + \frac{\Pi_h}{\mu_h}(1 - e^{-\mu_h t}).$$

Whenever $P(0) \leq \Pi_h/\mu_h$, solutions of model (1) are increasing monotonically and are bounded above by Π_h/μ_h. Conversely, whenever $P(0) > \Pi_h/\mu_h$, solutions of model (1) are monotone decreasing and bounded below by Π_h/μ_h. In both cases, at limiting equilibrium, $\lim_{t\to\infty} P(t) = \Pi_h/\mu_h$.

From the fourth equation of (1),

$$\frac{\mathrm{d}T_r}{\mathrm{d}t} \leq \varepsilon_r \Lambda_v + \tau_1 F T_r,$$

that is,

$$\frac{\mathrm{d}T_r}{\mathrm{d}t} - \tau_1 F T_r \leq \varepsilon_r \Lambda_v.$$

Thus,

$$T_r(t) \leq \left(T_r(0) + \int_0^t \varepsilon_r \Lambda_v \exp\left\{-\tau_1 \int_0^\eta F(\theta)\mathrm{d}\theta\right\} \mathrm{d}\eta\right) \exp\left\{\tau_1 \int_0^t F(\theta)\mathrm{d}\theta\right\}.$$

Since the integrands of the above relation are continuous functions on the compact set $[0, t]$, then by the boundedness theorem, they are bounded on the set. The integrands are thus Riemann integrable on $[0, t]$ and the integrals are finite. This implies that

$$T_r(t) \leq (T_r(0) + c_1)c_2 = m_1,$$

where $c_1 = \int_0^t \varepsilon_r \Lambda_v \exp\left\{-\tau_1 \int_0^\eta F(\theta)\mathrm{d}\theta\right\} \mathrm{d}\eta$ and $c_2 = \exp\left\{\tau_1 \int_0^t F(\theta)\mathrm{d}\theta\right\}$.

Similarly, from the fifth equation of (1),

$$T_c(t) \leq \left(T_c(0) + \int_0^t \varepsilon_c \Omega_v \exp\left\{-\tau_2 \int_0^\eta E_b(\theta)\mathrm{d}\theta\right\} \mathrm{d}\eta\right) \exp\left\{\tau_2 \int_0^t E_b(\theta)\mathrm{d}\theta\right\}.$$

Thus, $T_c(t) \leq (T_c(0)+c_3)c_4 = m_2$, where $c_3 = \int_0^t \varepsilon_c \Omega_v \exp\left\{-\tau_2 \int_0^\eta E_b(\theta)\mathrm{d}\theta\right\} \mathrm{d}\eta$ and $c_4 = \exp\left\{\tau_2 \int_0^t E_b(\theta)\mathrm{d}\theta\right\}$.

Hence, the region $\mathcal{D}_1$,

$$\mathcal{D}_1 = \left\{(H_e(t), H_h(t), I(t), T_r(t), T_c(t), F(t), E_b(t), R_b(t)) \in \mathbb{R}_+^8 : P(t) \leq \frac{\Pi_h}{\mu_h}, T_r(t) \leq m_1,\right.$$
$$\left. T_c(t) \leq m_2, I(t) \geq 0, F(t) \geq 0, E_b(t) \geq 0, R_b(t) \geq 0\right\}$$

is positively invariant and attracting for the model (1).

From the first equation of (1), using the fact that $P(t) = H_e(t) + H_h(t) \leq \frac{\Pi_h}{\mu_h}$, it follows that

$$\begin{aligned}
\frac{\mathrm{dH}_e}{\mathrm{dt}} &= \Pi_h + \omega H_h - \gamma(1-\varepsilon_v)H_e(t)E_b(t) - \phi\varepsilon_h H_e(t) - \mu_h H_e(t) \\
&\leq \Pi_h + \omega H_h(t) - (\phi\varepsilon_h + \mu_h)H_e(t) \\
&\leq \Pi_h + \omega\left(\frac{\Pi_h}{\mu_h} - H_e(t)\right) - (\phi\varepsilon_h + \mu_h)H_e(t) \\
&= \Pi_h\left(\frac{\mu_h + \omega}{\mu_h}\right) - (\omega + \mu_h + \phi\varepsilon_h)H_e(t) \\
&= (\omega + \mu_h + \phi\varepsilon_h)\left(\frac{\Pi_h(\omega + \mu_h)}{\mu_h(\omega + \mu_h + \phi\varepsilon_h)} - H_e(t)\right) \\
&= (\omega + \mu_h + \phi\varepsilon_h)(H_e^* - H_e(t)),
\end{aligned}$$

where $H_e^* = \dfrac{\Pi_h(\omega + \mu_h)}{\mu_h(\omega + \mu_h + \phi\varepsilon_h)}$.

Hence,

$$H_e(t) \leq H_e^* - (H_e^* - H_e(0))e^{-kt},$$

where $k = \omega + \mu_h + \phi\varepsilon_h$. From the above relation, $H_e(t)$ either approaches H_e^* asymptotically or there exists some finite time after which $H_e(t) \leq H_e^*$.

From the second equation of model (1),

$$\begin{aligned}
\frac{\mathrm{d}H_h}{\mathrm{d}t} &= \phi\varepsilon_h H_e - \omega H_h - \mu_h H_h \\
&\leq \phi\varepsilon_h[\frac{\Pi_h}{\mu_h} - H_h] - \bar{c}H_h \\
&= \frac{\phi\varepsilon_h(\Pi_h - \mu_h H_h)}{\mu_h} - \bar{c}H_h \\
&= \left[\left(\frac{\Pi_h\phi\varepsilon_h}{\phi\varepsilon_h\mu_h + \mu_h\bar{c}} - H_h\right)(\phi\varepsilon_h + \bar{c})\right] \\
&= (\bar{c} + \phi\varepsilon_h)(H_h^* - H_h),
\end{aligned}$$

where $H_h^* = \dfrac{\Pi_h\phi\varepsilon_h}{\mu_h(\bar{c} + \phi\varepsilon_h)}$ and $\bar{c} = \omega + \mu_h$.

Hence,

$$H_h(t) \leq H_h^* - (H_h^* - H_h(0))e^{-kt}.$$

This implies that $H_h(t)$ either approaches H_h^* asymptotically or there exists some finite time after which $H_h(t) \leq H_h^*$. Consequently, any solution $H_e(t)$, $H_h(t)$, $I(t)$, $T_r(t)$, $T_c(t)$, $F(t)$, $E_b(t)$, and $R_b(t)$ at $t \geq 0$, of model (1), that commences in the positive orthant $\mathbb{R}_+^8$, either remains confined in, enters, or asymptotically approaches the region $\mathcal{D}_2$, where

$\mathcal{D}_2 = \{(H_e(t), H_h(t), I(t), T_r(t), T_c(t), F(t), E_b(t), R_b(t)) \in \mathcal{D}_1 : H_e \leq H_e^*, H_h \leq H_h^*, I \geq 0, T_r \geq 0, T_c \geq 0, F \geq 0, E_b \geq 0, R_b \geq 0\}$.

□

2.2. Existence and stability of equilibria

2.2.1. Local stability of the CFE

The *Chlamydia*-free equilibrium (CFE) of the model system (1), which is obtained by setting the right hand side of the model system (1) to zero, and then choosing solutions where $E_b = R_b = I = 0$, is given by

$$\begin{aligned}
E_0 &= \{H_e^*, H_h^*, I^*, T_r^*, T_c^*, F^* E_b^*, R_b^*\}, \\
&= \left(\frac{\Pi_h(\omega + \mu_h)}{\mu_h(\omega + \mu_h + \phi\varepsilon_h)}, \frac{\Pi_h\phi\varepsilon_h}{\mu_h(\omega + \mu_h + \phi\varepsilon_h)}, 0, \frac{\varepsilon_v\Lambda_v}{\mu_t}, \frac{\varepsilon_c\Omega_v}{\mu_t}, 0, 0, 0\right).
\end{aligned} \tag{2}$$

Using the next generation operator method (as described by van den Driessche and Watmough [68]) on the model system (1), the linear stability of the equilibrium E_0 can be established. Using the notations in [68], the matrices $\mathbf{F}$ and $\boldsymbol{V}$, for the transmission (new infection) terms and transition terms of the infected subsystem (formed by the differential equations for compartments $I, E_b,$ and R_b in model equations (1)), respectively, are given by

$$\boldsymbol{F} = \begin{pmatrix} 0 & \gamma(1 - \varepsilon_v)H_e^* & 0 \\ 0 & 0 & 0 \\ 0 & 0 & 0 \end{pmatrix} \tag{3}$$

and

$$\boldsymbol{V} = \begin{pmatrix} \kappa & 0 & 0 \\ -\kappa N_E & \alpha\varepsilon_a + \mu_e + \gamma(1-\varepsilon_v)H_e^* & 0 \\ -\kappa N_{\mathrm{R1}} & 0 & \mu_e \end{pmatrix}. \tag{4}$$

Thus, the basic reproduction number $\mathcal{R}_0$ of the model (1), given by the spectral radius of the next generation matrix $\boldsymbol{F}\boldsymbol{V}^{-1}$, is

$$\mathcal{R}_0 = \frac{N_E\gamma\,(1-\varepsilon_v)\,H_e^*}{\gamma\,(1-\varepsilon_v)\,H_e^* + (\alpha\varepsilon_a + \mu_e)}. \tag{5}$$

The basic reproduction number $\mathcal{R}_0$ is written in such a way that one can track the contribution of the infected and infectious classes (infected epithelial cells and elementary bodies, respectively) to the epidemic. The $\mathcal{R}_0$ expression in (5) is simply the product of the infection rate of healthy epithelial cells by EBs ($\gamma(1-\varepsilon_v)H_e^*$), number of infectious *Chlamydia* (EB) released by a bursting infected epithelial cell (N_E), and the expected duration of infectiousness of EBs $\left(\dfrac{1}{\gamma\,(1-\varepsilon_v)\,H_e^* + (\alpha\varepsilon_a + \mu_e)}\right)$.

Implementing *Theorem 2* of van den Driessche and Watmough [68], the following result is established.

Lemma 2.3. *The Chlamydia-free equilibrium (CFE) E_0, of the model system (1), is locally stable whenever $\mathcal{R}_0 \leq 1$ and unstable if $\mathcal{R}_0 > 1$.*

2.2.2. Global stability of CFE

Theorem 2.4. *The CFE of the model system (1), given by Equation (2), is globally asymptotically stable (GAS) in $\mathcal{D}_2$ whenever $\mathcal{R}_0 \leq 1$ and unstable otherwise.*

Proof. Consider the candidate Lyapunov function

$$\mathbb{V} = N_E I(t) + E_b(t),$$

with Lyapunov derivative (where a dot represents differentiation with respect to t) given by

$$\begin{aligned}
\dot{\mathbb{V}} &= N_E\dot{I} + \dot{E}_b \\
&= N_E(\gamma(1-\varepsilon_v)H_eE_b - \kappa I - \rho IF) + (N_E\kappa I - \gamma(1-\varepsilon_v)H_eE_b - \varepsilon_a\alpha E_b - \mu_eE_b) \\
&= E_b(\gamma(1-\varepsilon_v)H_e(N_E-1)) - N_E\rho IF - \alpha\varepsilon_aE_b - \mu_eE_b \\
&\leq E_b(\gamma(1-\varepsilon_v)H_e(N_E-1) - \alpha\varepsilon_a - \mu_e) \\
&\leq E_b(\gamma(1-\varepsilon_v)H_e^*(N_E-1) - \alpha\varepsilon_a - \mu_e) \quad (\text{since } N_E - 1 > 0 \text{ and } H_e \leq H_e^* \text{ in } \mathcal{D}_2) \\
&= E_b(\gamma(1-\varepsilon_v)N_EH_e^* - \gamma(1-\varepsilon_v)H_e^* - \alpha\varepsilon_a - \mu_e] \\
&= E_b\left((\gamma(1-\varepsilon_v)H_e^* + \alpha\varepsilon_a + \mu_e)\left(\frac{\gamma(1-\varepsilon_v)N_EH_e^*}{\gamma(1-\varepsilon_v)H_e^* + \alpha\varepsilon_a + \mu_e} - 1\right)\right) \\
&= E_b(\gamma(1-\varepsilon_v)H_e^* + \alpha\varepsilon_a + \mu_e)\left(\frac{\gamma N_e\Pi_h\,(1-\varepsilon_v)\,(\mu_h+\omega)}{\gamma\Pi_h\,(1-\varepsilon_v)\,(\mu_h+\omega) + \mu_h\,(\alpha\varepsilon_a+\mu_e)\,(\mu_h+\phi\varepsilon_h+\omega)} - 1\right) \\
&= E_b(\gamma(1-\varepsilon_v)H_e^* + \alpha\varepsilon_a + \mu_e)(\mathcal{R}_0 - 1) \leq 0, \quad \text{when } \mathcal{R}_0 \leq 1.
\end{aligned}$$

Since all the model parameters and variables are non-negative, it follows that $\dot{\mathbb{V}} \leq 0$ for $\mathcal{R}_0 \leq 1$ and $\dot{\mathbb{V}} = 0$ if and only if $E_b = 0$. Hence, $\mathbb{V}$ is a Lyapunov function on $\mathcal{D}_2$. Furthermore, $\mathcal{D}_2$ is a compact and absorbing subset of $\mathbb{R}^8_+$, and the largest compact invariant set in $\{(H_e, H_h, T, T_r, T_c, F, E_b, R_b) \in \mathcal{D}_2 : \dot{\mathbb{V}} = 0\}$ is the singleton E_0. Thus, by Lasalle's invariance principle [69], $I \to 0$ and $E_b \to 0$ as $t \to \infty$. Substituting $I = E_b = R_b = 0$ into the model equations (1) shows that $H_e \to H_e^*$, $H_h \to H_h^*$, $T_r \to T_r^*$, $T_c \to T_c^*$, and $R_b \to 0$ as $t \to \infty$. Hence, every solution of the model system (1), with initial conditions in $\mathcal{D}_2$, approaches the CFE E_0 as $t \to \infty$ (that is, the CFE E_0 is GAS in $\mathcal{D}_2$) whenever $\mathcal{R}_0 \leq 1$.

□

2.2.3. Existence of CPE

In order to obtain the *Chlamydia*-present equilibrium (CPE) of model (1), we set the right hand sides of the model equations (1) to zero, and solve for all its state variables. We also express the state variables in terms of the force of infection

$$\lambda^* = \gamma E_b^{**}. \tag{6}$$

Thus, the CPE of model (1) is given by

$$E_1 = \{H_e^{**}, H_h^{**}, I^{**}, T_r^{**}, T_c^{**}, F^{**} E_b^{**}, R_b^{**}\}, \tag{7}$$

where

$$\begin{aligned}
H_e^{**} &= \frac{\pi_h\left(\omega+\mu_h\right)}{\lambda^*\left(1-\varepsilon_v\right)\left(\omega+\mu_h\right)+\mu_h\left(\phi\varepsilon_h+\omega+\mu_h\right)},\\
H_h^{**} &= \frac{\pi_h\phi\varepsilon_h}{\lambda^*\left(1-\varepsilon_v\right)\left(\omega+\mu_h\right)+\mu_h\left(\phi\varepsilon_h+\omega+\mu_h\right)},\\
I^{**} &= \frac{\lambda^*(1-\varepsilon_v)H_e^{**}}{\kappa+\rho F^{**}},\\
T_r^{**} &= \frac{\varepsilon_r\Lambda_v}{\mu_t-\tau_1 F^{**}},\\
T_c^{**} &= \frac{\varepsilon_c\Omega_v}{\mu_t-\tau_2 F^{**}},\\
F^{**} &= \frac{1}{6}\frac{Z^{2/3}+2C_2Z^{1/3}-12C_3C_1+4C_2^2}{C_1Z^{1/3}},\\
E_b^{**} &= \frac{N_E\kappa I^{**}-\lambda^*(1-\varepsilon_v)H_e^{**}}{\varepsilon_a\alpha+\mu_e},\\
R_B^{**} &= \frac{N_{R1}\kappa I^{**}+N_{R2}\rho I^{**}F^{**}}{\mu_e},
\end{aligned} \tag{8}$$

where $Z = 12\sqrt{3}\sqrt{27\,C_1{}^2C_4{}^2-18\,C_1C_2C_3C_4+4\,C_1C_3{}^3+4\,C_2{}^3C_4-C_2{}^2C_3{}^2}C_1+108\,C_4C_1{}^2-36\,C_3C_2C_1+8\,C_2{}^3$, $C_1 = k_6\tau_1\tau_2\rho$, $C_2 = \rho\,\Lambda_v\varepsilon_r\psi_1\tau_2+\rho\,\Omega_v\varepsilon_c\psi_2\tau_1+\rho\,\mu_t\tau_1+\rho\,\mu_t\tau_2$,
$C_3 = \rho\,\left(\alpha\,\varepsilon_a+\mu_e\right)\mu_t{}^2+\rho\,\mu_t\psi_1\varepsilon_r\Lambda_v+\rho\,\mu_t\psi_2\varepsilon_c\Omega_v+\tau_2\psi_1\varepsilon_r\Lambda_v\left(\kappa\,N_E-\kappa\right)+\tau_1\psi_2\varepsilon_c\Omega_v\left(\kappa\,N_E-\kappa\right)$
$k_1 = \mu_t\psi_1\varepsilon_r\Lambda_v$, $k_2 = \tau_2\psi_1\varepsilon_r\Lambda_v$, $k_3 = \mu_t\psi_2\varepsilon_c\Omega_v$, $k_4 = \tau_1\psi_2\varepsilon_c\Omega_v$, $k_5 = \kappa\,N_E-\kappa$, $k_6 = \alpha\,\varepsilon_a+\mu_e$.
Substituting for E_b^{**} in the relation (6), we obtain

$$\begin{aligned}
\lambda^* &= \frac{\gamma}{\varepsilon_a\alpha+\mu_e}\left[N_E\kappa I^{**}-\lambda^*(1-\varepsilon_v)H_e^{**}\right]\\
&= \frac{D_1}{D_2}\left(\frac{\kappa\mathcal{R}_0}{\kappa+\rho F^{**}}-1\right),
\end{aligned} \tag{9}$$

where $D_1 = \mu_h(\varepsilon_a\alpha+\mu_e)(\omega+\mu_h+\phi\varepsilon_h)+\gamma(1-\varepsilon_v)(\omega+\mu_h)\Pi_h$, and $D_2 = (\varepsilon_a\alpha+\mu_e)(\omega+\mu_h)(1-\varepsilon_v)$.
From Equations (8), F^{**} is a positive constant independent of λ^*. For λ^* to be biologically relevant, i.e. $\lambda^* > 0$, $\mathcal{R}_0 > 1+\dfrac{\rho F^{**}}{\kappa} > 1$. We define $\mathcal{R}_0^c = 1+\dfrac{\rho F^{**}}{\kappa}$ as the *critical* $\mathcal{R}_0$.

Lemma 2.5. *The model system* (1)

- *has no Chlamydia-present (endemic) equilibrium if* $\mathcal{R}_0 < \mathcal{R}_0^c < 1$, *where* $\mathcal{R}_0^c$ *is a threshold called the critical* $\mathcal{R}_0$.
- *has one positive Chlamydia-present equilibrium* E_1 *in* $\mathcal{D}_2$, *whenever* $\mathcal{R}_0 > \mathcal{R}_0^c > 1$.

- *has no positive equilibrium otherwise.*

Thus, the above mathematical analyses show that the model system (1) has a globally asymptotically stable *Chlamydia*-free equilibrium (CFE) whenever $\mathcal{R}_0 \leq 1$, and a unique *Chlamydia*-present equilibrium (CPE) when $\mathcal{R}_0 > \mathcal{R}_0^c > 1$. Simply put, the *C. trachomatis* infection will be cleared if $\mathcal{R}_0 \leq 1$, and would persist otherwise.

3. Numerical Simulations

3.1. Sensitivity analysis

Sensitivity analysis quantifies how variability (prediction imprecision) in predictors (input parameters) influence the value of outcome variables (responses) [70, 71]. In order to examine the sensitivity of $\mathcal{R}_0$ to variations in some parameters, we use Latin Hypercube Sampling (LHS) and partial rank correlation coefficient (PRCC) with 10000 Monte Carlo simulations per run. LHS is a stratified Monte Carlo sampling technique used for unbiased sampling of predictors in a multi-dimensional parameter space [70]. It is a very efficient sampling design which allows for the variation of predictor values simultaneously, in which each value is used only once in the analysis [71]. It is useful for executing an uncertainty analysis [70, 71]. Uncertainty analysis is useful for evaluating the prediction imprecision in a response due to the uncertainty in estimating the predictor. Sensitivity analysis extends uncertainty analysis by ranking the predictors in terms of their contribution (order of importance) to the variability of each of the responses [70, 71]. A sensitivity analysis is performed by the calculation of PRCCs for each predictor (sampled by the LHS scheme) and each model outcome variable [71].

The calculation of PRCCs is useful for classifying the importance of predictor-response correlations [70]. It enables the establishment of the statistical relationship between each predictor and each response(s), other predictors being held constant at their expected value [71]. A PRCC measures the degree of monotonicity between a specific predictor and a response. The sign (positive or negative) of the PRCC of a predictor indicates the qualitative (but not quantitative) affiliation (increase or decrease, respectively) it has with the response [70, 71]. The magnitude of the PRCC shows how important the uncertainty in the estimation of the predictor value is, to the prediction imprecision in the response value [71]. The relative importance of predictors can be determined via a comparison of the values of their PRCCs [71]. One can also access the monotonicity between a predictor and response by examining scatter plots of its PRCCs [71].

The sampling and sensitivity analysis methods used in SaSAT (Sampling and Sensitivity Analysis Tools [70]) were implemented in order to conduct uncertainty and sensitivity analysis on the model system (1). In the analysis, we use PRCCs, as described above, to distinguish and measure statistical influence, in particular, the monotonicity of the input variables on the response, which is the basic reproduction number $\mathcal{R}_0$. Tornado plots are used for illustrating the results of sensitivity analyses [70]. Input parameters with positive PRCCs are depicted by bar plots to the right, and with positive values on the horizontal axis, while input parameter with negative PRCCs are depicted by bar plots to the left, and with negative values on the horizontal axis.

In the implementation of each LHS sampling scheme, a uniform probability density function (pdf) is specified for each unknown parameter pdf (See Blower et al [71] for other PRCC methodology). Generally, model parameters with large PRCC values and corresponding small p-values (< 0.05) are considered most influential in PRCC analysis [72]. In order to assess if each PRCC value is significantly different from zero, we derive their p-values from Student's t-test as described on Page 242 of the LHS technique paper by Blower et al. [71].

Figure 2 is the tornado plot of the PRCC of all the predictors of our model system as described in Table 2. It shows that the predictors with the most positive influence on the *in vivo* propagation of a chlamydial infection, with their PRCCs and corresponding t-values (all p-values < 0.00001) bracketed as an ordered pair, are Π_h, the production rate of healthy epithelial cells (0.4184, 142.3424), γ, effective contact rate between epithelial cells and EB chlamydial forms (0.3592, 120.7180), N_E, the number of EBs released on lysis of an infected cell (0.1967, 65.1094), and ω, the waning rate of the protection of epithelial cells by the humoral immune response (HIR) (0.1588, 44.5970). On the other hand, the predictors with the most

negative influence on $\mathcal{R}_0$, with their PRCCs and corresponding t-values (all p-values < 0.00001) bracketed as an ordered pair, are ε_v, the efficacy of the mucosal chlamydial vaccine (-0.4929, -58.9374), ε_h, the efficacy of the HIR in protecting epithelial cells (-0.3069, -49.7342), ϕ, the rate of protection of epithelial cells by the HIR (-0.2966, -49.2320), and α, the macrophage engulfment rate of extracellular EBs (-0.1608, -34.2236). This result suggests that the two most important factors to be considered for achieving a disease-free state *in vivo* are ε_v, the efficacy of the mucosal chlamydial vaccine and ε_h, the efficacy of the HIR in protecting epithelial cells.

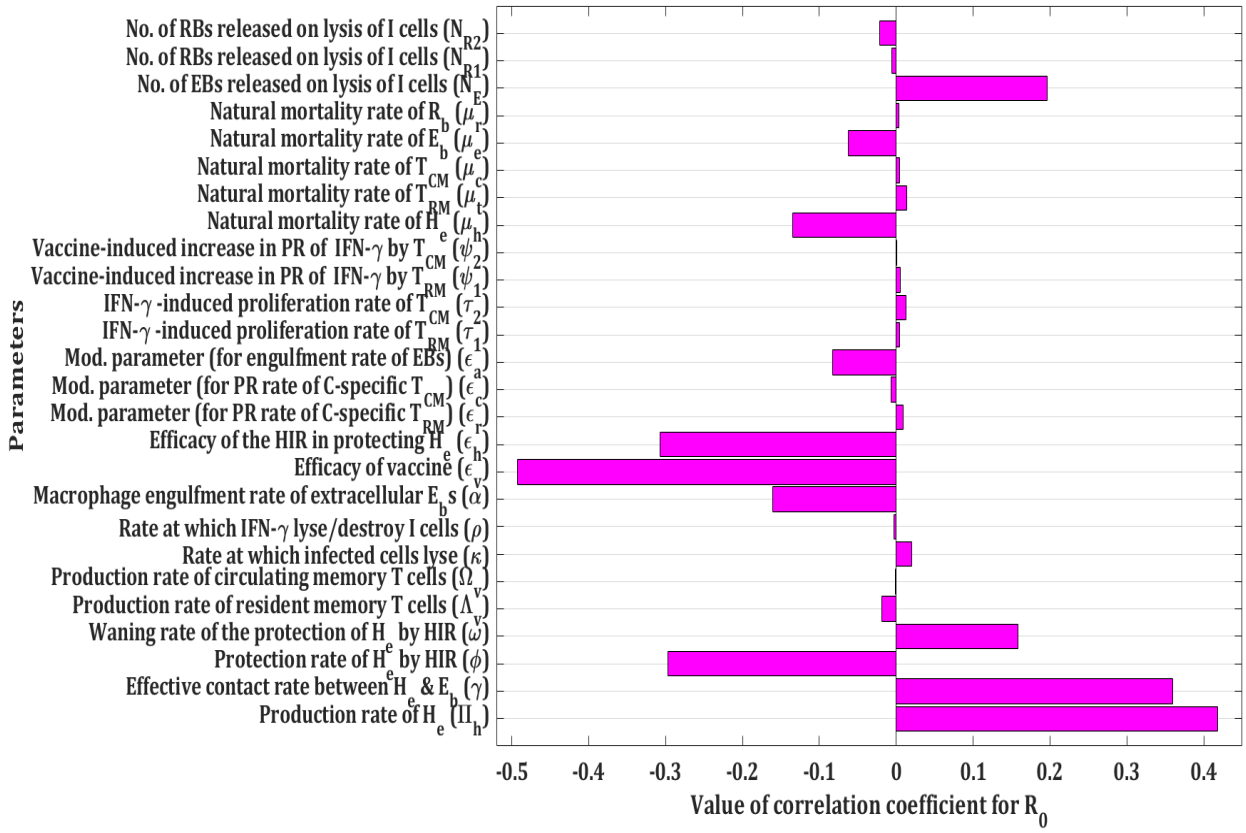


Figure 2: Tornado plot of the PRCC of $\mathcal{R}_0$ to the input parameters of model system (1), as described in Table 2, using $\mathcal{R}_0$ as the output. Mod. = Modification; HIR = Humoral immune response; No.= Number; T_{RM} = Tissue-resident memory T cells; T_{CM} = Circulating memory T cells.

In order to investigate the existence of any non-monotonicity between the basic reproduction number $\mathcal{R}_0$ and selected predictors/parameters (those with high absolute values of PRCC), we produce scatter plots of their PRCCs and examine them. The plots compare the $\mathcal{R}_0$ (log 10 scale) against each selected parameter. For this analysis, only predictors with fairly significant monotonic relationships with the $\mathcal{R}_0$ are displayed. Figures 3 and 4 display the monotonic relationship between the indicated parameters and the log scale of the basic reproduction number $\mathcal{R}_0$, simply put, they illustrate the variations in $\mathcal{R}_0$ against the input variables. Obviously, at $\log(\mathcal{R}_0) = 0$, $\mathcal{R}_0 = 1$. Figures 3(a), 3(b), and 4(c) show the monotonic relationship between Π_h, the rate of replenishment/production of healthy epithelial cells, γ, the effective contact rate between healthy epithelial cells and EB forms, and N_E, the number of EB forms released on lysis of infected cells, respectively, and the basic reproduction number $\mathcal{R}_0$. The figures indicate that a monotonic increase in the specified predictors produces a monotonic increase in the response $\mathcal{R}_0$. It can be seen on Figure 3(d) that

there exists some monotonic but weak relationship between ω, the waning rate of the protection of epithelial cells by the humoral immune response, and $\mathcal{R}_0 = 1$. The figure indicates that a monotonic increase in ω will produce a slight monotonic increase in $\mathcal{R}_0$.

Figures 3(c) and 4(a) show the monotonic relationship between ϕ, the rate of protection of healthy epithelial cells by the humoral immune response, α, the macrophage engulfment rate of extracellular EB forms, respectively, and the basic reproduction number $\mathcal{R}_0$. The figures indicate that a monotonic decrease in the specified predictors produces a monotonic decrease in the response $\mathcal{R}_0$. The predictor that shows the most significant correlation to the basic reproduction number $\mathcal{R}_0$ is ε_v, the efficacy of the *Chlamydia* vaccine. It can be seen that on Figure 4(b) that the higher the efficacy of the vaccine, the lower the $\mathcal{R}_0$. In particular, it can be seen that a vaccine of an efficacy of about 90% will facilitate the prevention of the progression of a *Chlamydia* infection, hence, eradicating the infection.

We carried out further numerical investigation (sensitivity analysis) by increasing the rates at which some biological processes occur in order to see how they affect $\mathcal{R}_0$, and thus the prognosis of the disease. When the highest range of only ϕ, the protection rate of healthy epithelial cells by the humoral immune response, was increased to 35 hr^{-1}, $\mathcal{R}_0$ could not be brought below unity. This suggests that the protection conferred upon epithelial cells by the humoral immune response is not enough to prevent chlamydial infection, even if it is higher than the known biological plausibility. However, when both ϕ and α (the macrophage engulfment rate of EBs) were increased to 20 hr^{-1}, it was seen that $\mathcal{R}_0$ was brought below unity. This would be good for the prognosis of chlamydial infection if those values were biologically plausible, but they are not currently plausible. These results are not graphically displayed here.

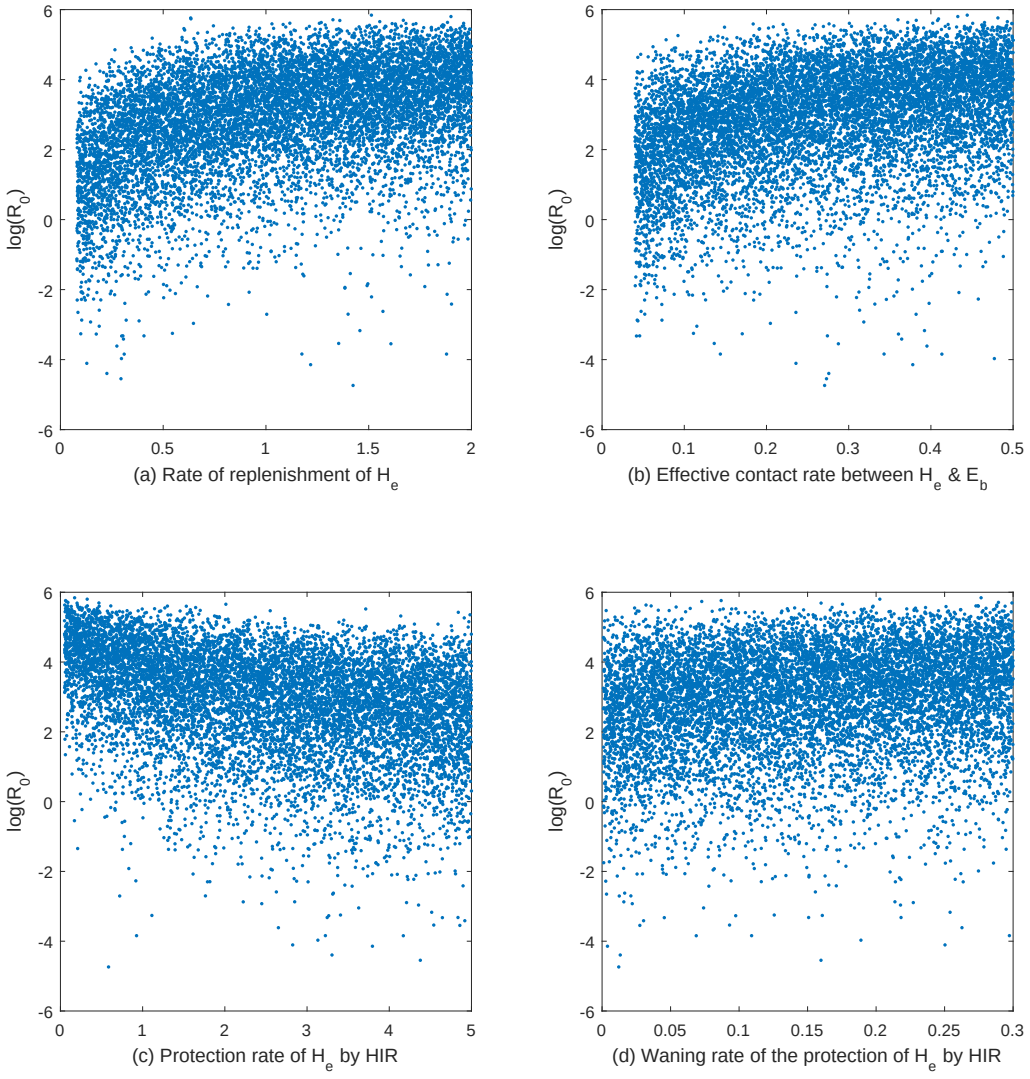


Figure 3: Scatter plots that compare the basic reproduction number $\mathcal{R}_0$ against selected parameters. HIR means the humoral immune response.

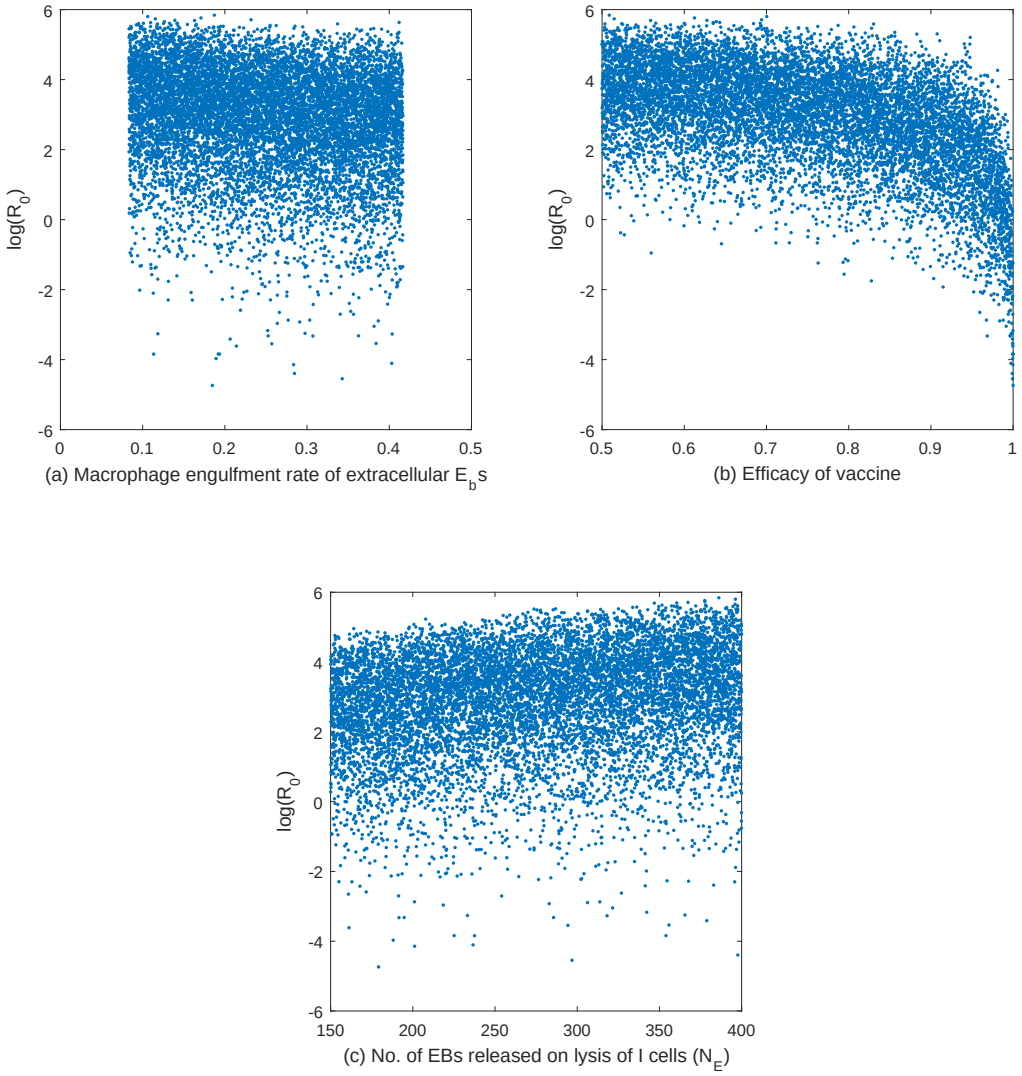


Figure 4: Scatter plots that compare the basic reproduction number $\mathcal{R}_0$ against selected parameters.

Parameters	Baseline Value	Range	Reference
Π_h	2.1	[1,2.1] hr^{-1}	[55, 46]
γ	0.000083	0.00008333 hr^{-1}	★
ϕ	3	[0.05,5] hr^{-1}	[55]
ω	0.1	0.1 hr^{-1}	[55]
Λ_v	0.009	[0.003,0.009] hr^{-1}	★
Ω_v	0.002	[0.002083,0.003333] hr^{-1}	[73]
κ	0.020833	0.0138-0.025 hr^{-1}	[46]
ρ	4	[0.05,5] hr^{-1}	[55]
α	0.3	[0.08333,0.41667] hr^{-1}	[46]
ε_v	Varies	$(0, 1]$	♣
ε_h	0.5	$(0, 1]$	♣
ε_r	1.2	$[1, 2]$	♠
ε_c	1.2	$(1, 2)$	♠
ε_a	1.2	$(1, 2)$	♠
τ_1	0.001	0.001 hr^{-1}	★
τ_2	0.001	0.001 hr^{-1}	★
ψ_1	0.00005	0.00005 hr^{-1}	★
ψ_2	0.00005	0.00005 hr^{-1}	★
μ_h	0.0008333	$[0, 0.02]$ hr^{-1}	[74]
μ_t	0.004	[0.0008333,0.00625] hr^{-1}	[73]
μ_c	0.004	[0.0008333,0.00625] hr^{-1}	[73]
μ_e	0.005	[0.00375,0.015] hr^{-1}	[51]
N_E	200	150-400	[55]
N_{R1}	10	10-50	★
N_{R2}	20	10-50	★

Table 3: Values and ranges of the parameters of the model (1). ♣: As described in Section 2, these parameters are bounded by 0 and 1. ♠: As described in Section 2, these modification parameters are strictly greater than 1, since in the absence of the chlamydial vaccine, there will still be the production of the *Chlamydia*-specific immune cells that they describe. It is supposed that the maximum increase/proliferation of these cells that the chlamydial vaccine can induce is double. ★: Estimated values.

3.2. Critical Vaccine Efficacy

High vaccine efficacies are often difficult to achieve in real world experiments. It has been established that all vaccines are partially efficacious, that is, vaccine efficacies are always less than a 100% [75]. Thus, we estimate the critical efficacy of an effective *Chlamydia* vaccine if it does not need to be up to 100%. The biological interpretation of our vaccine efficacy is the reduction of the probability of infection of a healthy epithelial cell per contact with an infectious *Chlamydia* (EB forms). We solve for the expression for the critical vaccine efficacy (ε_v^c), by setting $\mathcal{R}_0$ in Equation 5 to 1. This gives:

$$\varepsilon_v^c = 1 - \frac{\alpha\varepsilon_a + \mu_e}{\gamma H_e^*(N_E - 1)}, \tag{10}$$

where $H_e^* = \dfrac{\Pi_h(\omega + \mu_h)}{\mu_h\left(\omega + \mu_h + \phi\varepsilon_h\right)}$.

Lemma 3.1. *The basic reproduction number $\mathcal{R}_0 < 1$ whenever $\varepsilon_v > \varepsilon_v^c$.*

3.2.1. Investigating the relationship between the Critical Vaccine Efficacy and the efficacy of the humoral immune response in protecting healthy epithelial cells

It has been established that a mucosal chlamydial vaccines induce an enhanced protective immunity against subsequent infection [16, 12, 22, 2, 6]. Consequently, it is important that we quantify the influence of the critical vaccine efficacy on the efficacy of the humoral immune response in protecting healthy epithelial cells in the presence of the vaccine. Figure 5 depicts the relationship between the critical vaccine efficacy, ε_v^c, and the efficacy of the humoral immune response (HIR) in protecting healthy epithelial cells, ε_h. It can be seen that for all possible values of ε_h, $\varepsilon_h \in [0, 1]$, values of the critical vaccine efficacy ε_v^c is in the

range [0.73,1]. Since high vaccine efficacies are hard to achieve, and we require that the vaccine is able to induce significant protection of healthy epithelial cells, we suppose that the efficacy of the humoral immune response in protecting healthy epithelial cells is 0.5. Thus, we take this as the baseline value of ε_h as shown in Table 3.

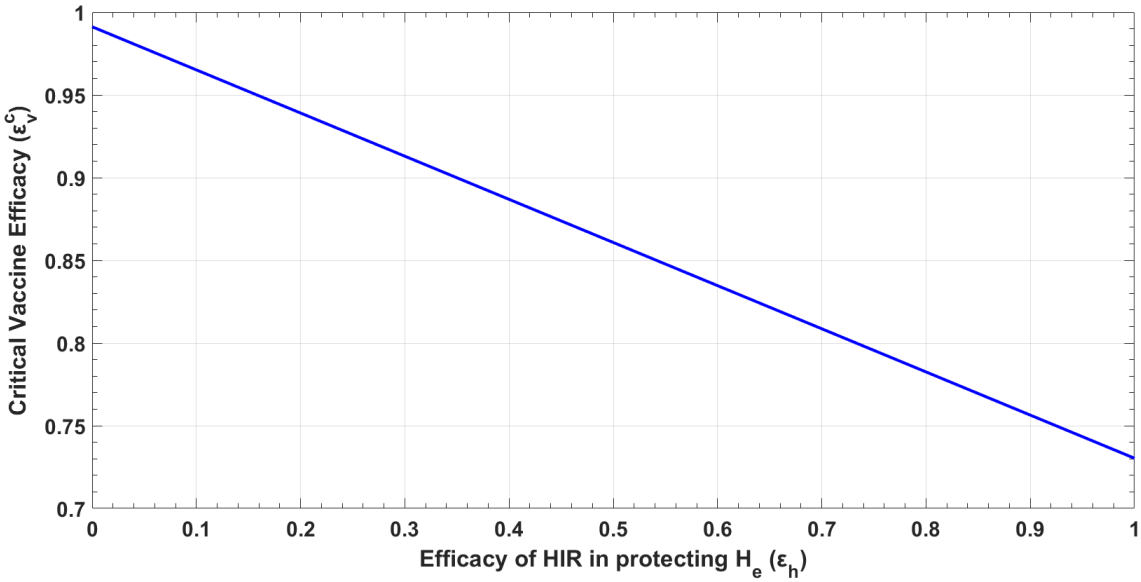
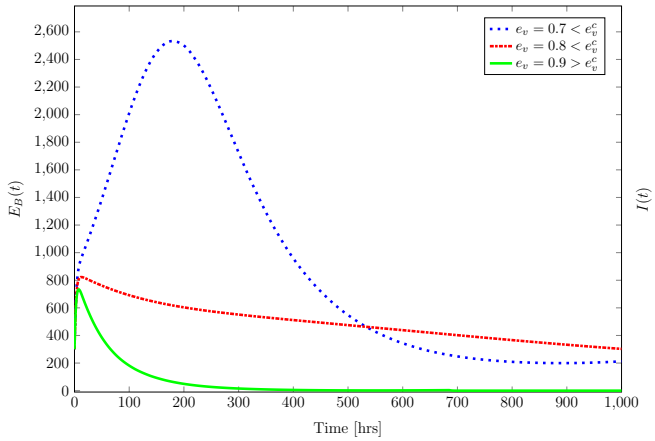
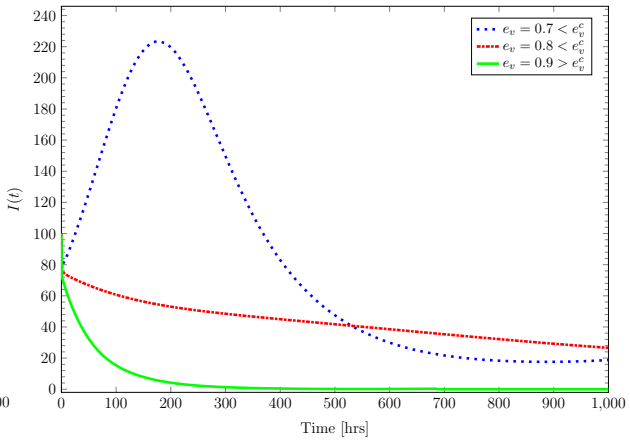


Figure 5: Graphical illustration of the relationship between the critical vaccine efficacy, ε_v^c, and the efficacy of the humoral immune response (HIR) in protecting healthy epithelial cells, ε_h.

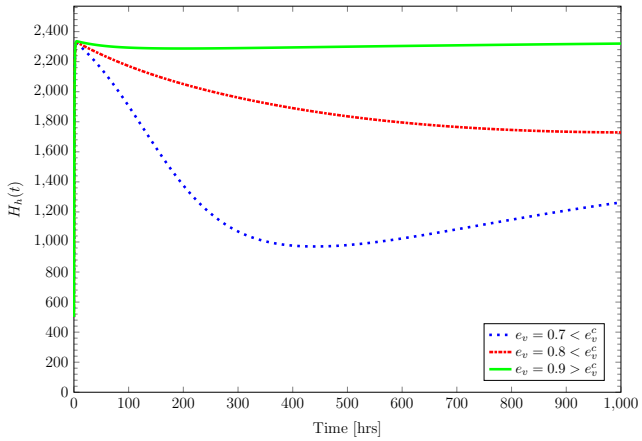
Whenever the vaccine efficacy ε_v is greater than the critical vaccine efficacy ε_v^c, *Chlamydia* is cleared from the host system, since $\mathcal{R}_0 < 1$, and Theorem 2.4 guarantees the *in vivo* clearance of the pathogen under this setting. Numerical results of Lemma 3.1 are shown on Figure 6, for three different values of ε_v. For the baseline values shown in Table 3, the critical vaccine efficacy $\varepsilon_v^c = 0.86$, which is also shown on Figure 5. Figures 6a and 6b show that for values of $\varepsilon_v < \varepsilon_v^c$, the infection may not be abated, even in the presence of a *Chlamydia* vaccine pre-infection. However, for any value $\varepsilon_v > \varepsilon_v^c$, the infection can be eliminated from the host system. Figure 6c also shows the impact of the vaccine efficacy on protected epithelial cells. It indicates that for values of $\varepsilon_v < \varepsilon_v^c$, the ability of the humoral immune response to protect healthy epithelial cells from *Chlamydia* infection may wane, while it would increase for values of $\varepsilon_v > \varepsilon_v^c$.



(a) Concentration of elementary bodies, $E_b(t)$



(b) Concentration of infected epithelial cells, $I(t)$



(c) Concentration of protected healthy epithelial cells, $H_h(t)$

Figure 6: Numerical simulation of the *Chlamydia* model (1), showing the time course plot of (a) $E_b(t)$, concentration of elementary body forms, (b) $I(t)$, concentration of infected epithelial cells, and (c) $H_h(t)$, concentration of protected healthy epithelial cells, respectively, all over 1000 hours post infection, and for $\varepsilon_v = 0.7 < \varepsilon_v^c$, $\varepsilon_v = 0.8 < \varepsilon_v^c$, and $\varepsilon_v = 0.9 > \varepsilon_v^c$.

Equation (10) shows that the numerical value of the critical vaccine efficacy (ε_v^c) depends on other model parameters, including the efficacy of the humoral immune response in protecting healthy epithelial cells (ε_h), reviewed above. Thus considering the implications of Lemma 3.1 and Theorem 2.4, we assess the impact of the *Chlamydia* vaccine by depicting contour plots of the basic reproduction number $\mathcal{R}_0$ as a function of ε_v, the vaccine efficacy, and ε_h, the efficacy of the humoral immune response in protecting healthy epithelial cells in Figure 7. In Figure 7, it can be seen that for values of $\mathcal{R}_0 \leq 1$, the use of an imperfect *Chlamydia* vaccine can lead to the elimination of the disease. For example, for relatively low values of $\mathcal{R}_0$, such as $\mathcal{R}_0 = 1.433$, the use of a *Chlamydia* vaccine with a modest efficacy of 80%, and with a 50% efficacy in the protection of healthy epithelial cells from *Chlamydia* infection, can bring about the elimination of chlamydial infection. A relatively low $\mathcal{R}_0$ value of 1.304 would also yield $\varepsilon_v = 75\%$ and $\varepsilon_h = 70\%$. Note that we have only used values of ε_v and ε_h between the interval $[0.3, 1]$ because we observed that values of the parameters between the interval $[0, 0.3)$ do not necessarily yield relatively low values of $\mathcal{R}_0$, for parameter values in Table 3.

We differentiate the basic reproduction number $\mathcal{R}_0$ with respect to ε_v. This gives:

$$\frac{\partial \mathcal{R}_0}{\partial \varepsilon_v} = -\frac{N_E \gamma (\alpha \varepsilon_a + \mu_e) H_e^*}{(\gamma (1 - \varepsilon_v) H_e^* + (\alpha \varepsilon_a + \mu_e))^2} < 0. \tag{11}$$

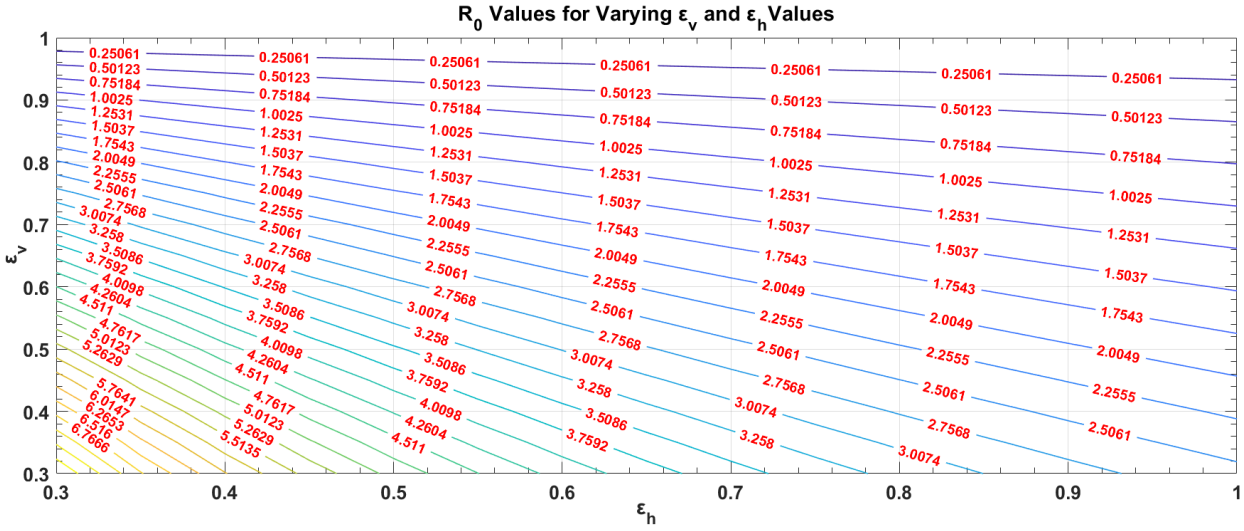


Figure 7: Simulation of the *Chlamydia* vaccine model (1), showing contour plots of $\mathcal{R}_0$ as a function of ε_v, the vaccine efficacy, and ε_h, the efficacy of the humoral immune response in protecting healthy epithelial cells.

Equation 11 shows that $\mathcal{R}_0$ is a decreasing function of ε_v, which implies that a vaccine with a good efficacy will reduce the *in vivo* concentration of *Chlamydia*.

We monitor the effect of efficacy of the humoral response in the protection of healthy epithelial cells from chlamydial infection, by differentiating $\mathcal{R}_0$ with respect to ε_h. This gives:

$$\frac{\partial \mathcal{R}_0}{\partial \varepsilon_h} = -\frac{\mu_h \phi \gamma \Pi_h N_E (1-\varepsilon_v)(\omega+\mu_h)(\alpha\varepsilon_a+\mu_e)}{(\mu_h(\alpha\varepsilon_a+\mu_e)(\omega+\mu_h+\phi\varepsilon_h)+\gamma\Pi_h(1-\varepsilon_v)(\omega+\mu_h))^2} < 0. \tag{12}$$

Equation 12 shows that $\mathcal{R}_0$ is a decreasing function of ε_h, which implies that if the vaccine brings about more protection of healthy epithelial cells from infection, then the reproduction number $\mathcal{R}_0$ decreases.

We also monitor the effect of the vaccine-induced increase in the engulfment rate of EB forms of *Chlamydia* by antibodies (macrophages) by differentiating $\mathcal{R}_0$ with respect to ε_a. This gives:

$$\frac{\partial \mathcal{R}_0}{\partial \varepsilon_a} = -\frac{N_E \alpha \gamma (1-\varepsilon_v) H_e^*}{(\gamma\,(1-\varepsilon_v)\,H_e^* + (\alpha\varepsilon_a+\mu_e))^2} < 0. \tag{13}$$

Equation 13 shows that $\mathcal{R}_0$ is a decreasing function of ε_a, which implies that if the vaccine increases the concentration of extracellular *Chlamydia* engulfed by antibodies, then the reproduction number $\mathcal{R}_0$ decreases.

3.3. More Numerical Results

We investigate the *Chlamydia* burden in an *in vivo Chlamydia* infection post-vaccination, with varying vaccine efficacy, by tracking the concentrations of elementary bodies, infected epithelial cells, and protected epithelial cells over about forty-two days (1000 hours) post-infection. Using the parameter values in Table 3, the model system (1) was simulated for varying values of ε_v, $0 \leq \varepsilon_v \leq 1$, the vaccine efficacy, and ε_h, $0 \leq \varepsilon_h \leq 1$, the efficacy of the humoral immune response in protecting healthy epithelial cells. The corresponding concentrations of the EB forms, infected epithelial cells, and of healthy epithelial cells protected from *Chlamydia* infection by the humoral immune response, are all graphically displayed on Figures 8, Figures 9, and Figures 10, respectively.

Numerical results, as shown graphically on both Figures 8 and 9, of the model system (1), show that for some combinations of the vaccine efficacy (ε_v) and the efficacy of the humoral immune response in protecting epithelial cells, (ε_h), the *Chlamydia* infection does not burden the host system in the presence of the *Chlamydia* vaccine. This is characterised by the clearance of the pathogen by the final time of the

simulation (that is, concentrations of EB forms and of infected epithelial cells are both zero). As shown on Figure 10, for such combinations of ε_v and ε_h, surges in the concentrations of healthy epithelial cells that are protected by the humoral immune response (H_h), in the presence of the vaccine, also indicate the anti-*Chlamydia* potency of the vaccine.

3.4. Exploring Conditions for Lower Vaccine Efficacies

We explore conditions under which a mucosal chlamydial vaccine may have efficacies lower than the critical vaccine efficacy described in Section 3.2. The efficacy of the vaccine, ε_v, and efficacy of the humoral response in protecting healthy epithelial cells, ε_h, are varied. This is to quantify the effects of different combinations of these efficacies on the disease prognosis, by evaluating the concentrations of chlamydial EB forms, infected epithelial cells, and epithelial cells protected by the humoral immune response at the end of the simulation.

Based on the numerical simulation results, as shown on Figures 8, 9, and 10, an efficacy of 45% is sufficient for a *Chlamydia* vaccine. However, important characteristics of the vaccine is that its presence in the host system should have the ability to (1) boost the natural humoral immune response which protects healthy epithelial cells from infection to at least 90% of its biologically plausible potency (see Figure 8); (2) boost the production of resident and circulating memory T cells each by 20% ($\varepsilon_r = \varepsilon_c = 1.2$; see Table 3); and (3) increase the rate at which macrophages engulf extracellular *Chlamydia* (EB forms) by 20% ($\varepsilon_a = 1.2$; this may mediate antibody-dependent cell-mediated cytotoxicity (ADCC)). An effective *Chlamydia* vaccine may also sufficiently have an efficacy of 65%, but it should have the ability to boost the natural humoral immune response by at least 85% of its biologically plausible potency (see Figure 8), other rates being equal.

These results show that if the mucosal *Chlamydia* vaccine has the right properties, a lower vaccine efficacy may suffice.

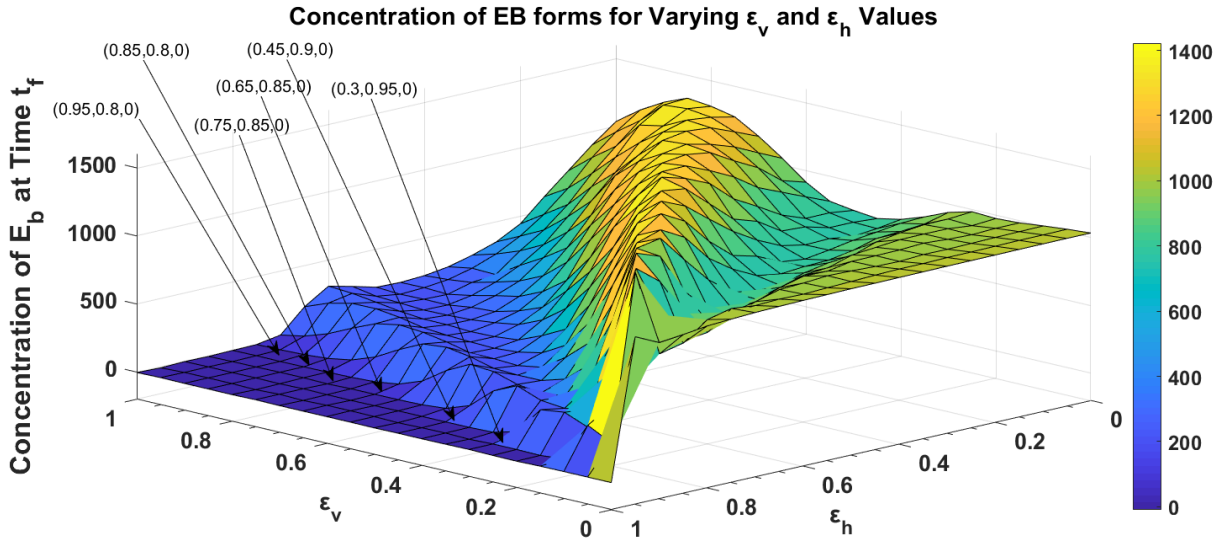


Figure 8: Numerical simulation of the *Chlamydia* model (1), showing a 3D representation of the concentration of elementary body forms (*Chlamydia* burden) at the end of the simulation (time $t_f = 1000$ hours post-infection), for varying values of ε_v, the vaccine efficacy, and ε_h, the efficacy of the humoral immune response in protecting healthy epithelial cells.

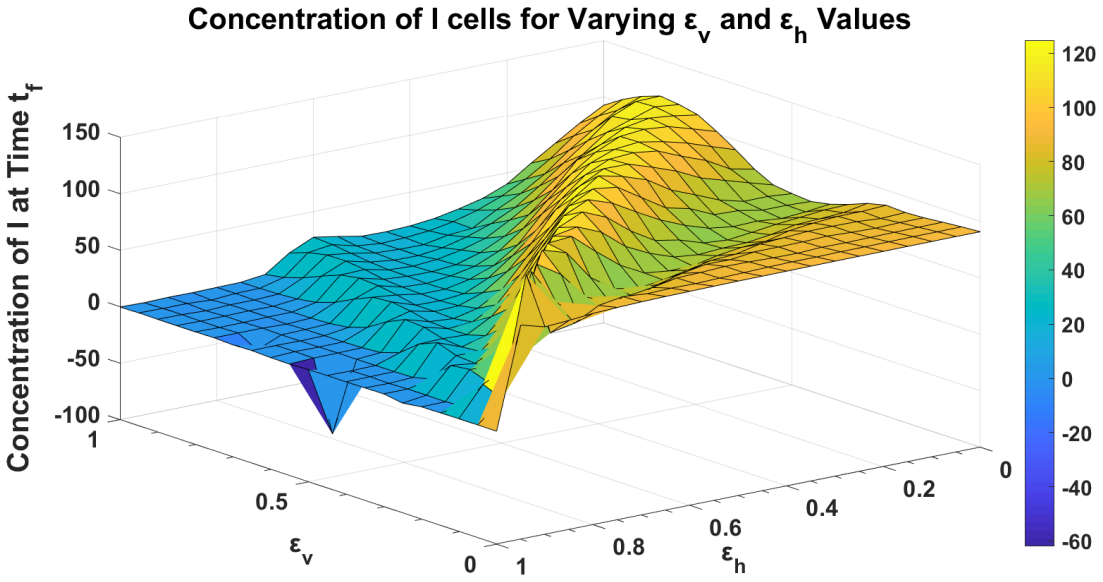


Figure 9: Numerical simulation of the *Chlamydia* model (1), showing a 3D representation of the concentration of infected epithelial cells at the end of the simulation (time $t_f = 1000$ hours post-infection), for varying values of ε_v, the vaccine efficacy, and ε_h, the efficacy of the humoral immune response in protecting healthy epithelial cells.

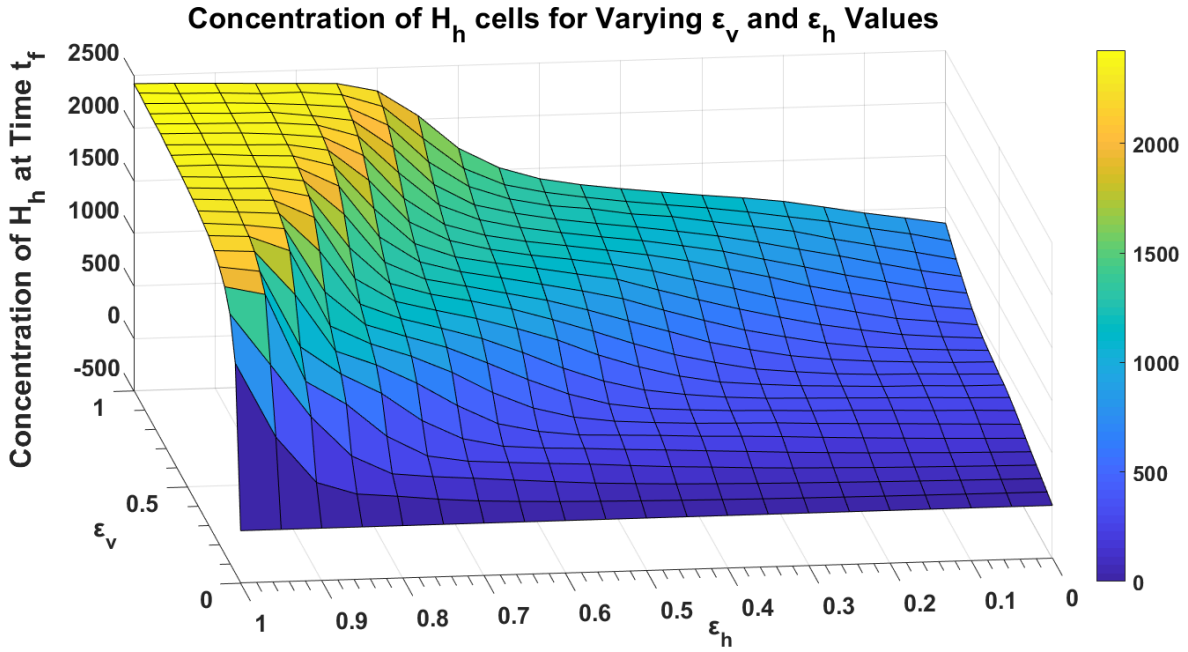


Figure 10: Numerical simulation of the *Chlamydia* model (1), showing a 3D representation of the concentration of protected healthy epithelial cells at the end of the simulation (time $t_f = 1000$ hours post-infection), for varying values of ε_v, the vaccine efficacy, and ε_h, the efficacy of the humoral immune response in protecting healthy epithelial cells.

4. Discussion

In this paper, the mathematical modelling literature has been extended by the theoretical assessment of the potential role of an imperfect anti-*Chlamydia* vaccine on the within-host dynamics of *C. trachomatis*.

The impacts of a potentially effective mucosal *Chlamydia* vaccine on the within-host dynamics and prognosis of a genital chlamydial infection were investigated. The study aims at estimating the requisite efficacy of an efficacious mucosal vaccine that could promote a stable disease-free state in vivo by (1) eliciting protective immunity against chlamydial challenge, and (2) efficiently clearing a chlamydial infection upon a re-challenge with *Chlamydia*. The developed model uses a prototype vaccine that induces similar protective immunity like that described by the study of Stary *et al.* [2].

In addition to the incorporation of the effects of a potentially efficacious mucosal *Chlamydia* vaccine, the model specifically extends the work by Sharomi and Gumel [55] by (i) adding a new compartment for the dynamics of *Chlamydia*-specific resident memory T cells; (ii) adding a new compartment for the dynamics of *Chlamydia*-specific circulating memory T cells; (iii) allowing for the proliferation of resident memory T cells and circulating memory T cells; (iv) allowing for a vaccine-induced (additional) protective immunity against infection of healthy epithelial cells by *Chlamydia* via antibody blocking; and (v) adding a compartment for the inhibitory action of IFN-γ. In order to gain qualitative insights into the model, mathematical analyses of the model were carried out. The model has a globally asymptotically stable *Chlamydia*-free equilibrium when its associated basic reproduction number $\mathcal{R}_0 \leq 1$. The model also has a unique *Chlamydia*-present equilibrium when the basic reproduction number $\mathcal{R}_0 > \mathcal{R}_0^c > 1$, where $\mathcal{R}_0^c$ is a threshold called the *critical* $\mathcal{R}_0$.

The quantification of how the variability in model parameters affect the value of the basic reproduction number $\mathcal{R}_0$ was investigated by carrying out sensitivity analysis of $\mathcal{R}_0$ to input parameters. In particular, the monotonic relationship between the model's basic basic reproduction number $\mathcal{R}_0$ and some predictor variables, in particular Π_h, the rate of replenishment/production of healthy epithelial cells, γ, the effective contact rate between healthy epithelial cells and EB forms, and N_E, the number of EB forms released on lysis of infected cells, were investigated. Simulation results indicate that a monotonic increase in the specified predictors produces a monotonic increase in the response $\mathcal{R}_0$. In addition, an investigation of the monotonic relationship between ϕ, the rate of protection of healthy epithelial cells by the humoral immune response, α, the macrophage engulfment rate of extracellular EB forms, and the basic reproduction number $\mathcal{R}_0$, indicates that a monotonic decrease in the specified predictor variables produces a monotonic decrease in the response $\mathcal{R}_0$. The predictor variable that shows the most significant inverse correlation to the $\mathcal{R}_0$ was ε_v, the efficacy of the mucosal *Chlamydia* vaccine. Summarily, the sensitivity analysis results suggest that a *Chlamydia* vaccine of an efficacy of about 90% may facilitate the prevention of the progression of a *Chlamydia* infection, hence, eradicating the infection.

The *Chlamydia* burden in an *in vivo Chlamydia* infection post-vaccination, for varying values of ε_v, $0 \leq \varepsilon_v \leq 1$, the vaccine efficacy, and ε_h, $0 \leq \varepsilon_h \leq 1$, the efficacy of the humoral immune response in protecting healthy epithelial cells, by tracking the concentrations of elementary bodies, infected epithelial cells, and protected epithelial cells over 1000 hours post-infection, was investigated. The vaccine was shown to be able to possess sufficiently low efficacies, however, it must also satisfy some other immune-related conditions if it must be effective. The mathematical expression for, and the values of, the *critical vaccine efficacy*, using parameter values presented in Table 3 was also obtained.

Results of the numerical simulations of the model show that a vaccine with a minimum (critical) efficacy of 86% may be required for the *in vivo* control of *Chlamydia* burden, which is characterised by the concentrations of infectious *Chlamydia* (EB forms) and of infected epithelial cells. Conditions under which lower vaccine efficacies may suffice are also explored. We also assessed the impact of the described *Chlamydia* vaccine by depicting contour plots of $\mathcal{R}_0$ as a function of ε_v, the vaccine efficacy, and ε_h, the efficacy of the humoral immune response in protecting healthy epithelial cells. Simulation results indicate that for values of $\mathcal{R}_0 \leq 1$, the use of an imperfect *Chlamydia* vaccine may lead to the clearance of the pathogen.

In conclusion, model results suggest that a *Chlamydia* vaccine that (1) decreases the concentration of newly infected epithelial cells; (2) increases the concentration of extracellular *Chlamydia* engulfed by antibodies; (3) boosts the protection of healthy epithelial cells from infection; may reduce the concentration of *Chlamydia in vivo*, thereby elimination the chlamydial infection overall. Candidate vaccines that possess similar properties as described by our model results may be efficacious in the *in vivo* control of *Chlamydia trachomatis* genital infection. It should be noted that results of this model should be viewed in the light of its associated parameters and assumptions.

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