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Superinfections can induce evolutionarily stable coexistence of pathogens

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Abstract Parasites reproduce and are subject to natural selection at several different, but intertwined, levels. In the recent paper, Gilchrist and Coombs (Theor. Popul. Biol. 69:145–153, 2006) relate the between-host transmission in the context of an SI model to the dynamics within a host. They demonstrate that within-host selection may lead to an outcome that differs from the outcome of selection at the host population level. In this paper we combine the two levels of reproduction by considering the possibility of superinfection and study the evolution of the pathogen's within-host reproduction rate p. We introduce a superinfection function $\phi = \phi(p, q)$, giving the probability with which pathogens with trait q, upon transmission to a host that is already infected by pathogens with trait p, "take over" the host. We consider three cases according to whether the function $q \to \phi(p, q)$ (i) has a discontinuity, (ii) is continuous, but not differentiable, or (iii) is differentiable in $q = p$. We find that in case (i) the within-host selection dominates in the sense that the outcome of evolution at the host population level coincides with the outcome of evolution in a single infected host. In case (iii), it is the transmission to susceptible hosts that dominates the evolution to the extent that the singular strategies are the same as when the possibility of superinfections is ignored. In the biologically most relevant case (ii), both forms of reproduction contribute to the value of a singular trait. We show that when ϕ is derived from a branching process variant of the submodel for the within-host interaction of pathogens and target cells, the superinfection functions fall under case (ii). We furthermore demonstrate that the superinfection model allows for steady coexistence of pathogen traits at the host population level, both on the ecological, as well as on the evolutionary time scale.

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1 Introduction

Evolution of virulence is an intriguing topic that has been on the minds of scientists for many decades. It was believed for a long time that all pathogens would eventually evolve to be benign to their hosts. The words of R. Dubos (1965) reflect this, in that time widely accepted, idea: "Given enough time, a state of peaceful coexistence eventually becomes established between any host and parasite."

This belief, sometimes termed *conventional wisdom*, was rejected only very recently (the first ideas and models can be found in [3,13,14,26–28,30]), when it was realised that it was based on the misconception that natural selection favours what is best for a species as a whole.

If, on the other hand, we take as a starting point that natural selection acts on finer levels, such as for example the parasite's reproductiveness, then some ambiguity arises in the case of microparasites (bacteria, viruses, protozoa and fungi) that reproduce within a host, but need to be transmitted at some stage to another host to keep the reproduction cycle going. These parasites reproduce at two different levels and are subject to natural selection at each of these.

For instance, imagine a simple within-host scenario in which pathogens compete for only one resource (i.e., one particular kind of cells that we shall call "target" cells), and where the pathogen's reproduction rate is the only trait subject to natural selection. In such a case, the pessimization principle [10,36] applies: optimal or, more precisely, continuously stable values of the pathogen's reproduction rate are those that minimize the availability of uninfected target cells within the single host that is being considered.

Imagine, on the other hand, that the spread of an infectious agent in a population of susceptible hosts is described by a simple SI model,

$$\begin{aligned} \frac{\mathrm{d}S}{\mathrm{d}t} &= b - \beta SI - \delta S, \\ \frac{\mathrm{d}I}{\mathrm{d}t} &= \beta SI - (\alpha + \delta)I, \end{aligned} \tag{1}$$

where S and I denote, respectively, the number of susceptible and infected (and also infectious) individuals, b stands for the population birth rate, β is the transmission rate and the parameters α and δ denote, respectively, the disease induced mortality and the death rate related to causes other than the disease. The disease induced death rate α is what a majority of the literature refers to as *virulence* [4,9,10,14,19,34,37,40,41].

If we now consider the virulence α as the only evolving trait and assume that infection with pathogens of one trait provides individuals with complete cross-immunity

to infections with different traits, then the pessimization principle implies that, at the host population level, the evolutionary winner is the trait that (locally) minimizes the steady state value of the number of susceptible hosts, $\hat{S}$. Since $\hat{S} = \frac{b}{\delta \mathscr{R}_0}$, we can equivalently say that natural selection tends to (locally) maximize the pathogen's basic reproduction ratio $\mathscr{R}_0$ (i.e., the expected number of new infections, caused by one infected individual that is introduced into a disease free environment), which is in this case given by

$$\mathscr{R}_0 = \frac{b\beta}{\delta(\alpha + \delta)}.$$

If b, β and δ are kept constant, then the basic reproduction ratio increases when α decreases and so the pathogens indeed evolve to be avirulent, just as the conventional wisdom predicts (we exclude the possibility of mutualism, i.e., α can not take negative values). This seems, however, a very unlikely scenario: virulence as well as transmissibility are in reality likely to be related to within-host characteristics, such as the rate at which pathogens reproduce inside a host or the pathogen load. But while an increased reproduction within a host may enhance transmissibility β, it may also harm the host by increasing α. In other words, there is a trade-off between pathogen production and transmissibility on the one side and virulence on the other.

In the last years several papers and books [1,2,5,6,9,14,15,19,25,28,34,37,38,40,41] appeared in which the evolution of virulence was studied, while taking into account such trade-offs. Most of them (with the exception of [1,2,19]), however, kept the within-host characteristics implicit and only assumed that the transmission parameter β is somehow directly related to virulence. In other words, it is assumed that $\beta = \beta(\alpha)$.

In [19], the very paper that inspired this work, both the within- and the between-host dynamics were made explicit. The authors considered the rate at which pathogens are produced inside a host as the only trait subject to natural selection and determined the continuously stable strategies (CSSs) for selection at the within-host, as well as for selection at the host population level. However, even though the between-host model was related to the dynamics within a host, the authors used a single infection model to describe the between-host dynamics. According to this model, no host can harbour more than one pathogen trait, which directly excludes reinfection induced evolution at the within-host level. The two levels at which natural selection works thus remain separated and, as a consequence, natural selection at these two levels may appear to be in conflict, i.e., the evolutionarily stable trait on one level may differ from the evolutionarily stable trait on the other.

In reality, natural selection does not act exclusively at any of these two levels. When random mutations of the pathogen evoke variation and, subsequently, selection within one individual, this also influences the evolutionary dynamics at the host population level (say, by influencing transmission) which, in turn, may have an effect on the within-host dynamics. In order to study the evolution of infectious diseases in a realistic manner, we must therefore consider the two levels as being coupled: we must relate the between-host dynamics to within-host characteristics and furthermore take into

account that hosts may harbour more than one pathogen trait, either at one and the same time or consecutively.

Multiple infections, which may result either from additional transmissions or from random mutations of the pathogens inside a host, can be modeled in different ways, depending also on the within-host model used. The particular within-host model we use in this paper does not allow for steady coexistence of different traits. In other words, there may be a period in which several traits are present within a host, but one of them will eventually outcompete the others. *Coinfection* models take the period of coexistence explicitly into account. *Superinfection* models, on the other hand, assume that, if the new trait takes over, it does so immediately. Both ways of modeling the additional infections have advantages as well as downsides. One can rightfully argue that superinfection models are less realistic compared to coinfection models since there will always be a period of time, however short, in which several traits will be present inside a host. But the added realism in coinfection models entails specification of the pathogen level immediately after transmission took place and there is, in general, hardly any biological information that can be translated into such a specification in the context of caricatural models of within-host dynamics.

In this paper we model multiple infections as superinfections. We take the SI model underlying (1) as the basis for the description of the spread of an infectious agent in a population of hosts. We relate the transmissibility β and the disease induced death rate α to within-host dynamics. By assuming that the dynamics within a host is fast compared to the dynamics at the host population level, β and α will actually depend on the steady state values of the within-host variables. Moreover, this difference in time scales of within- and between- host processes implies that when more than one trait is present inside a host, the best within-host competitor immediately eliminates all other traits. Multiple infections thus indeed manifest themselves as superinfections.

We assume throughout that mutations are rare on the time scale of transmission and demography. We thus adopt the Adaptive Dynamics' point of view and use some standard terminology of this field throughout the paper.

The main part of the paper is structured as follows. In Sect. 2 we present the two basic ingredients of the main superinfection model in Sect. 3, i.e., the within-host and the single infection between-host model. We single out the pathogen's within-host reproduction rate p as the only evolving trait and characterize the continuously stable values of p for the evolution at each of the two isolated levels. Even though natural selection in reality does not act solely at any of these two levels, we show in Sect. 3 that the singular strategies obtained at the two isolated levels can be seen as two extreme ends of the possible evolutionary outcomes of the combined selection. The precise value of a singular strategy, however, will depend on the way superinfections are incorporated into the model. Following [34], we introduce a *superinfection function* $\phi(p, q)$, describing the ability of pathogens with trait q to "take over" a host that is already infected by pathogens with trait p, and show that when the function $q \mapsto \phi(p, q)$

(i) has a jump discontinuity in $q = p$, then the CSSs coincide with the CSSs at the within-host level.

(ii) is continuous, but not differentiable, in $q = p$, then both levels contribute to the value of a singular strategy. In particular, when the CSSs at the two isolated levels are unique, the convergence stable strategy lies in-between the within-host CSS and the CSS of the single infection model.
(iii) is differentiable in $q = p$, then the singular strategies are the same as the ones given by the single infection model.

For the ease of formulation we sometimes use the terms *jump*, *mechanistic*, and *smooth* to describe, respectively, case (i), (ii) and (iii). This terminology is justified in Sect. 5, where we model in more detail the initial stages of an introduction of a mutant trait in a single infected host. Since the mutant trait is then likely to be present only in small quantities, we model the invasion as a stochastic birth-and-death process. We find that the smoothness of the superinfection function relates to the ability of the mutant trait to survive the initial phase of low abundance in an already infected host, which itself depends on the starting dose of the mutant.

In Sect. 4 we observe that singular strategies can no longer be characterized by way of an optimization principle when superinfections are taken into account. Among other things, this implies that the superinfection model allows for coexistence of pathogen traits at the host population level. While epidemiological coexistence (namely, coexistence on the time scale of transmission and demography) comes as no surprise (in fact, many previous papers in which superinfection models were studied [23,34,37,40,41,44] have also encountered such coexistence), the fact that coexistence can be maintained on the evolutionary time scale is a bit surprising (cf. [41] and Sect. 6) and has, to our knowledge, in the context of a superinfection model only been found in [37]. In Sect. 4 we furthermore demonstrate that the existence of branching points (which can lead to evolutionary coexistence of different strains), is promoted by a high transmission dose. The branching points themselves are of an unusual, asymmetric type.

Even though the models we use are very caricatural, the results of this paper highlight the following important point: in order to understand the evolution of infectious diseases one needs to form a sound understanding of how the host mediates interactions between slightly different strains that infect the host, either simultaneously or consecutively. Indeed, the way the host handles multiple infections is reflected in the (non)smoothness of the superinfection function, which, as we show, has a huge impact on the outcome of evolution.

Some concluding remarks are collected in Sect. 6.

Admittedly, we have chosen a very particular setting, namely, an SI model which assumes that individuals retain the infection until death, but that, perhaps due to reinfections or random mutations that take place inside the host, the pathogens' trait may change in the course of the infection. Although this shortens significantly the list of infectious diseases that are covered, we believe that this setting is worth studying for two reasons. Firstly, the assumptions made here provide an acceptable caricature for many relevant pathogens such as HIV, Hepatitis B and Hepatitis C virus. Secondly, the ideas and the framework presented in this paper can be extended to study evolutionary dynamics in other settings that include different dynamics at the within-host, as well as at the host population level (SEI, SIR, etc. models, different incidence rates, etc.).

2 Preliminaries

The aim of this section is to introduce two models, the within-host and the single infection between-host model, that will constitute the building blocks of the main model in the next section.

2.1 The within-host dynamics

To describe the dynamics within one host we use the following system of ODEs,

$$\frac{\mathrm{d}T}{\mathrm{d}t} = \lambda - kVT - dT, \tag{2a}$$

$$\frac{\mathrm{d}T^*}{\mathrm{d}t} = kVT - (\mu(p) + d)T^*, \tag{2b}$$

$$\frac{\mathrm{d}V}{\mathrm{d}t} = pT^* - kVT - cV. \tag{2c}$$

The three variables in (2), T, T^* and V represent, respectively, the number of uninfected and infected target cells and the number of free pathogens. The system (2) corresponds to the following scenario.

In the absence of the infectious agent, target cells are produced at a constant rate λ and die at a constant per capita rate d. When the host is infected, free pathogens inside a host die at per capita rate c. The mass action term kVT is used to model the process of infection within a host: it says that the rate at which pathogens find uninfected target cells, successfully bind to the surface of the cell and/or enter the target cell, is proportional to the product of the numbers of uninfected target cells and free pathogens. Upon infection, the uninfected target cell and the pathogen that infected it, form an infected cell. Hence, the term kVT is substracted from (2a) and (2c) and added in (2b). Infected target cells produce free pathogens at a rate p. This production comes at a cost, namely, it increases the death rate of infected target cells by $\mu(p)$. We shall assume that $\mu \in \mathscr{C}^2(\mathbb{R})$ and that it is a nonnegative, increasing function of the production rate p.

Remark 1 According to (2), a free pathogen infects individual cells of the host. The model is thus certainly a meaningful description for viruses (that, after binding to a cell, enter, or at least inject their genetic material, into the host cell [38]), but not so for bacteria and fungi that live in the interstitial fluid. Our formulation seems to focus on non-lytic viruses, since we suggest that free viruses are produced during the lifetime of an infected target cell. However, if viruses are produced within the cell and are released only when the cell dies, exactly the same mathematical formulation applies (the number of virus particles in the infected cell that dies is on average $\frac{p}{\mu(p)+d}$ and if we multiply the rate $(\mu(p) + d)T^*$ by this quantity we obtain pT^*). The model is thus equally appropriate when the virus is released via lysis.

Remark 2 System (2) is one of the models commonly used in the literature to describe the virus dynamics within a host [7,39]. According to (2), a free pathogen disappears

as a free particle at the moment it infects a target cell. A variation of this model, which is also used quite often [19,38], neglects the term $-kVT$ in (2c). The reasoning behind this omission is that the number of pathogens is much higher than the number of target cells and while the term $-kVT$ is relevant in (2a), it is negligible (compared to other terms) in (2c). While this simplified model was shown to yield a very good fit for, for instance, some stages of HIV progression, it may not do so in the very first stages of an infection, when the pathogens are rare. Since we are in this paper mainly interested in the ability of mutant traits to invade the resident pathogen population (and so we focus precisely on the initial stages of the introduction of a mutant trait) we shall work with the full model (2). Note, however, that omission of the term $-kVT$ in (2c) has one important modeling consequence. Namely, if we leave out the term $-kVT$ in (2c), the free pathogen is not "lost" upon infecting a target cell and can therefore infect more than one cell. This observation will be of great importance in Sect. 5, where we shall describe the initial stages of a reinfection as a stochastic birth-and-death process.

System (2) has two equilibria: the infection free steady state in which there are no pathogens and no infected target cells,

$$\hat{V} = \hat{T}^* = 0 \quad \text{and} \quad \hat{T} = \frac{\lambda}{d} =: T_0 \tag{3}$$

and a nontrivial equilibrium given by

$$\hat{T} = \frac{c}{k(\mathscr{B}_0(p) - 1)}, \tag{4a}$$

$$\hat{T}^* = \frac{\mathscr{B}_0(p)}{p}\left(\lambda - \frac{cd}{k(\mathscr{B}_0(p) - 1)}\right), \tag{4b}$$

$$\hat{V} = \frac{\lambda}{c}\left(\mathscr{B}_0(p) - 1\right) - \frac{d}{k}. \tag{4c}$$

Here, $\mathscr{B}_0$ stands for the so called *burst size*, i.e., the expected number of pathogens produced by one infected target cell. If the pathogen production rate of the infected target cell equals p, then

$$\mathscr{B}_0(p) = \frac{p}{\mu(p) + d}.$$

The nontrivial steady state given by (4) is biologically meaningful only when all three components in (4) are positive. The first, $\hat{T}$, is positive when the burst size exceeds one. If we then rewrite (4b) (and (4c)), we find that the other two components are strictly positive only when the pathogen's within-host reproduction ratio is larger than one. The within-host basic reproduction ratio of a pathogen, $\mathscr{R}_0^w$ (the superscript w serves to distinguish it from the pathogen's basic reproduction ratio at the host population level), is defined as the expected number of new pathogens produced by a single pathogen introduced into a virgin cell environment. Since free pathogens need to enter uninfected target cells in order to reproduce and since the probability with which the pathogen enters a target cell in a virgin environment equals $\frac{k\lambda}{k\lambda + dc}$, the within-host

basic reproduction ratio of a pathogen with trait p equals

$$\mathscr{R}_0^w(p) = \frac{k\lambda}{k\lambda + dc}\mathscr{B}_0(p).$$

The nontrivial equilibrium is thus biologically meaningful only when $\mathscr{R}_0^w(p) > 1$. When it exists, it is also locally asymptotically stable, while the infection free steady state is unstable in that case (see [7] for a global stability result and also [35] for local stability of variants of (2)).

We shall throughout the paper consider the rate of pathogen production p as the only trait subject to natural selection. All the other parameters in the within-host model will be kept constant throughout. Furthermore, we shall sometimes omit the word "pathogen" and simply describe hosts or cells as being "infected by a certain trait".

We assume for simplicity that, if a target cell is infected with one trait, it is protected from further infections. In other words, we do not consider superinfections or coinfections at the cell level. Since this assumption implies that the pathogens compete within a host for only one resource, viz. uninfected target cells, the evolutionary dynamics at the within-host level is very simple. Namely, when a mutant trait, say q, is introduced into a host where the trait p is resident, the mutant is successful (according to the deterministic model) if and only if it exploits the resource better than the resident, i.e., when $\hat{T}(q) < \hat{T}(p)$. This is sometimes called the *pessimization principle*: when the environment the pathogens experience is one dimensional and an invasion results in competitive exclusion of one of the traits, then natural selection necessarily leads to the worst possible environment [10,16,36]. Note, incidentally, that minimization of $\hat{T}$ is equivalent to maximization of $\mathscr{R}_0^w$ and also to maximization of $\mathscr{B}_0$.

The precise conclusions, however, will depend on the trade-off function $\mu(p)$. In the following two examples we present the two most commonly used trade-off relations.

Example 1 Let μ be a concave function of the form $\mu(p) = \frac{ap}{p+b}$. This makes $\hat{T}(p)$ a strictly decreasing function. The requirement $\mathscr{R}_0^w > 1$, which can be rewritten as $\hat{T}(p) < T_0$ (see (3) and (4a)), hence gives a lower bound for the admissible values of p, denoted by $p_{\min}$, but there exists no upper bound for p (see Fig. 1). In this case the minimum of $\hat{T}(p)$ is not reached for finite p. In practice, however, it is likely that there are physiological constraints on the pathogen production rate p. In such a case then, natural selection will drive p towards the physiological maximum, $p_{\max}$.

Example 2 Let us now take $\mu(p) + d = de^{ap}$. Requiring that $0 < \hat{T}(p) < T_0$, we find a lower, as well as an upper bound, for feasible values of the pathogen production rate (see Fig. 2). The minimal value of $\hat{T}(p)$ is now obtained for some intermediate value $p^* \in (p_{\min}, p_{\max})$.

In general, of course, the trade-off function μ may be more complicated and there may be more than one CSS for the within-host selection. We shall in any case assume that

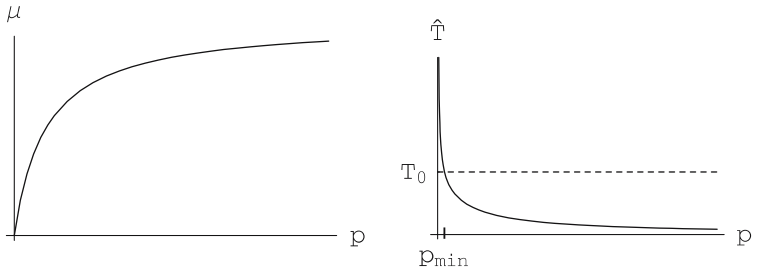

Fig. 1 An example of a concave trade-off function $\mu(p)$ and the corresponding $\hat{T}(p)$

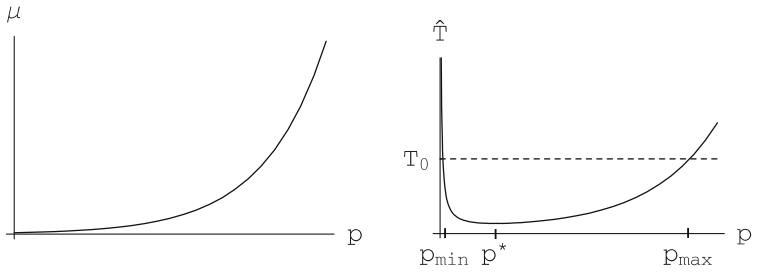

Fig. 2 An example of a convex trade-off function $\mu(p)$ and the corresponding $\hat{T}(p)$

- there exists a finite interval $[p_{\min}, p_{\max}]$ of feasible values of p,
- the function $\hat{T}(p)$ is nowhere locally constant and
- there are no accumulation points of the extrema of $\hat{T}(p)$.

If we denote the set of all within-host CSSs by $\mathscr{C}^*$, then $p^* \in \mathscr{C}^*$ is either one of the boundary points, $p^* \in \{p_{\min}, p_{\max}\}$, or a point in $(p_{\min}, p_{\max})$ that, in a generic case, satisfies

$$\left.\frac{\mathrm{d}\hat{T}(q)}{\mathrm{d}q}\right|_{q=p^*} = 0 \quad \text{and} \quad \left.\frac{\mathrm{d}^2\hat{T}(q)}{\mathrm{d}q^2}\right|_{q=p^*} > 0.$$

Note that we have assumed that $\hat{T}(p)$ is differentiable twice and so we can indeed compute and characterize singular points for the within-host evolution by way of differentiation.

2.2 The single infection between-host model

When studying the spread of an infectious agent at the host population level, one can make several different assumptions concerning the way in which traits interact within one host. The *single infection model* [19], for instance, assumes complete cross-immunity between different traits. That is, hosts infected by some trait p are completely protected from further infections by other traits. In our opinion, this assumption is inconsistent with acknowledging that within-host selection may take place. Indeed, if

an internally produced mutant may take over, then why would that be impossible for a competitor introduced from outside?

We shall nevertheless now formulate this single infection model as we shall need it (or, more precisely, the singular traits it admits) in the next section.

We make the following assumptions regarding the dynamics at the host population level. In a disease free environment, individuals are born at a rate b and die at per capita rate δ. Individuals become infectious at the moment they are infected and retain the infection until death, which now occurs at an increased per capita rate $\alpha + \delta$. Transmission occurs according to mass action with proportionality constant β, which incorporates both the rate at which a susceptible individual comes into contact with an infectious host and the probability with which a contact results in a transmission.

We shall assume that the parameters b and δ, which capture the population dynamics in the absence of the disease, are constant. The disease induced mortality α and the transmission parameter β, however, will depend on the infection status of the host. For instance, one can imagine that hosts that carry more pathogens may be more infectious than the ones in which the amount of pathogens is lower. Similarly, α may depend on the parasite abundance in the host but perhaps also on the amount of uninfected target cells (which seems likely at least in the case of HIV, where the virus attacks the very cells that should serve to protect the host). We shall therefore assume that α and β depend on the production rate p, but that this dependence is expressed mechanistically in terms of the within-host variables, T, T^* and V. Since we have furthermore assumed that the dynamics within a host is fast compared to the dynamics at the population level, we assume that α and β are in fact functions of the steady state values $\hat{T}$, $\hat{T}^*$ and $\hat{V}$, say,

$$\alpha(p) = A(\hat{T}(p), \hat{T}^*(p), \hat{V}(p)), \quad \beta(p) = B(\hat{T}(p), \hat{T}^*(p), \hat{V}(p))$$

and assume that A, $B \in \mathscr{C}^2(\mathbb{R}^3, \mathbb{R})$.

If we now assume that two traits, say, p and q, circulate in the population, then the above assumptions give rise to the following system

$$\begin{aligned}
\frac{\mathrm{d}S}{\mathrm{d}t} &= b - \beta(p)SI_p - \beta(q)SI_q - \delta S,\\
\frac{\mathrm{d}I_p}{\mathrm{d}t} &= \beta(p)SI_p - (\alpha(p) + \delta)I_p,\\
\frac{\mathrm{d}I_q}{\mathrm{d}t} &= \beta(q)SI_q - (\alpha(q) + \delta)I_q,
\end{aligned}$$

where S denotes the number of susceptible individuals and I_p, I_q denote the number of individuals infected by, respectively, trait p and trait q. Note that there are, due to complete cross-immunity between the traits, no doubly infected individuals.

In the absence of trait q, the system admits two equilibria: the disease free steady state $\hat{S}(p) = \frac{b}{\delta} =: S_0$, $\hat{I}_p(p) = 0$, and the nontrivial equilibrium, which is biologically

meaningful only when

$$\mathscr{R}_0(p) = \frac{b}{\delta} \cdot \frac{\beta(p)}{\alpha(p) + \delta} > 1.$$

When the endemic steady state exists, it is also a global attractor (cf. [12], Exercise 3.11).

Since selection at the population level occurs in this case solely through the subpopulation of susceptible hosts, we already know that the pessimization principle will apply and that the basic reproduction ratio $\mathscr{R}_0(p)$ will be (locally) maximized in the course of evolution. However, with our minds already on the following section, we now reformulate the CSS conditions in terms of the *invasion exponent*, $s_p(q)$, which denotes the growth rate of the subpopulation of hosts infected by trait q, when introduced into a steady resident population in which (only) trait p is present. In this case

$$s_p(q) = \beta(q)\hat{S}(p) - (\alpha(q) + \delta), \tag{5}$$

with

$$\hat{S}(p) = \frac{\alpha(p) + \delta}{\beta(p)}. \tag{6}$$

The number of local maxima of $\mathscr{R}_0$ will depend on the choice of trade-off functions α and β and the value of δ. These choices will, along with the condition $0 < \hat{S}(p) < S_0$, pose restrictions on the values of p that guarantee persistence at the host population level. Taking these, along with similar restrictions at the within-host level (see the previous subsection) into account, we obtain the set of feasible values of the pathogen production rate p.

If we denote by $\mathscr{C}^\bullet$ the set of CSS values for selection at the between-host level (in the context of the single infection model), then $p^\bullet \in \mathscr{C}^\bullet$ either lies on the boundary of the domain, or is a point in the interior, which in a generic case satisfies

$$\frac{\partial s_{p^\bullet}(q)}{\partial q}\Big|_{q=p^\bullet} = 0 \quad \text{and} \quad \frac{\partial^2 s_{p^\bullet}(q)}{\partial q^2}\Big|_{q=p^\bullet} < 0.$$

3 The superinfection model

We now include the possibility that infected individuals are reinfected. We assume that the within-host dynamics is fast compared to the dynamics at the host population level. Since the within-host model we use does not allow for steady coexistence of different traits, this means that when a mutant trait is introduced (in one host) into a monomorphic pathogen population, the invasion is either unsuccessful, or it results in an immediate trait substitution within the host. The single infection model is therefore replaced by the following, so called *superinfection model*,

$$\begin{aligned}\frac{\mathrm{d}S}{\mathrm{d}t} &= b - \beta(p)SI_p - \beta(q)SI_q - \delta S,\\ \frac{\mathrm{d}I_p}{\mathrm{d}t} &= \beta(p)SI_p - (\alpha(p)+\delta)I_p + \Phi(q,p)I_pI_q,\\ \frac{\mathrm{d}I_q}{\mathrm{d}t} &= \beta(q)SI_q - (\alpha(q)+\delta)I_q + \Phi(p,q)I_pI_q,\end{aligned} \tag{7}$$

where

$$\Phi(p,q) = \beta(q)\phi(p,q) - \beta(p)\phi(q,p) \tag{8}$$

and where the *superinfection function* $\phi(p,q)$ describes the ability of the pathogens with trait q to "take over" a host that is already infected by trait p. More precisely, we define

$$\phi(p,q) := \textit{ the probability with which the trait } q\textit{, upon transmission to a host already infected by trait } p\textit{, eliminates } p \textit{ and thus takes over the host.}$$

Since the ability of the invading trait q to grow in a host that is already infected by trait p is completely determined by $\hat{T}(p)$ and $\hat{T}(q)$, we shall actually deal only with functions $\phi(p,q)$ that can be written as functions of $\hat{T}(p)$ and $\hat{T}(q)$, say,

$$\phi(p,q) = \psi(\hat{T}(p), \hat{T}(q)). \tag{9}$$

Suppose now that the newly introduced trait q does worse at the within-host level than the resident trait p, i.e., $\hat{T}(p) < \hat{T}(q)$. Then q is actually unable to infect a host that is already infected by trait p. And since also the traits that reduce $\hat{T}$ to exactly the same level as the resident cannot grow (cf. Sect. 5), we shall assume that ψ is a nonnegative function that satisfies

H$_1$**.** $\psi(x,y) = 0$ whenever $y \geq x$.

To determine whether a trait q will grow when introduced into a population of hosts with resident trait p, we compute the invasion exponent, which we shall now denote by $r_p(q)$, and which in this case equals

$$\begin{aligned} r_p(q) &= \beta(q)\hat{S}(p) - (\alpha(q)+\delta) + \Phi(p,q)\hat{I}(p)\\ &= s_p(q) + \hat{I}(p)\left(\beta(q)\psi(\hat{T}(p),\hat{T}(q)) - \beta(p)\psi(\hat{T}(q),\hat{T}(p))\right), \end{aligned} \tag{10}$$

where s denotes the invasion exponent for the single infection model (given in (5) and (6)) and

$$\hat{I}(p) = \frac{b}{\alpha(p)+\delta} - \frac{\delta}{\beta(p)} = \frac{b}{\beta(p)\hat{S}(p)} - \frac{\delta}{\beta(p)}$$

denotes the equilibrium value of I in the absence of trait q.

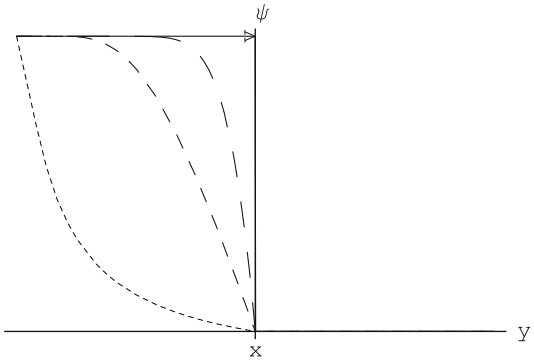

Fig. 3 For every function ψ we have $\psi(x, y) = 0$ whenever $y \geq x$. For $y < x$, the *solid line* represents the "jump" case, the *two dashed lines* in the middle represent the "mechanistic" case and the *bottom dashed line* represents the "smooth" case

The evolutionary dynamics of p and, in particular, the corresponding singular strategies, depend heavily on the behaviour of $\psi(x, y)$ when $y \uparrow x$. We shall consider the following three possibilities for ψ as a function of y: (i) the "*jump*" case, i.e., when $y \mapsto \psi(x, y)$ has a jump discontinuity in the point $y = x$, (ii) the "*mechanistic*" case, i.e., when $y \mapsto \psi(x, y)$ is continuous, but not differentiable, in $y = x$ and (iii) the "*smooth*" case when $y \mapsto \psi(x, y)$ is differentiable in $y = x$ (cf. Fig. 3).

The remaining part of this section is devoted to showing that these three classes of superinfection functions determine three very different ways in which natural selection can work out. We shall not, however, be concerned at this point with details on how the behaviour of $\psi(x, y)$ for $y \uparrow x$ reflects the way in which interactions of different traits within one host are modeled. This will be our task in Sect. 5. We furthermore emphasize that the analysis of this section is local analysis, based on the assumption that mutations are not only rare on the time scale of transmission and demography, but also "sufficiently" small. In other words, we assume that the mutant trait q is "sufficiently" close to the resident trait. Some observations regarding the global evolutionary dynamics are collected in Sect. 4.

So let us denote by $\mathscr{C}^{\bullet *}$ the set of continuously stable strategies of the superinfection model and begin with case (i).

3.1 The "jump" case

As an example of a discontinuous superinfection function, one may have the following in mind.

Example 3 Let

$$\phi(p, q) = \begin{cases} 1, & \hat{T}(q) < \hat{T}(p), \\ 0, & \text{otherwise.} \end{cases} \tag{11}$$

In this case, superinfections occur in accordance with the deterministic description: the invading trait q is successful if and only if it is able to win the internal competition with the resident trait p. In particular, ϕ does not distinguish between winning

strategies: traits that are only slightly better at the within-host level have the same advantage over the resident than significantly better within-host strategies.

So let us assume that ψ is a nonnegative function that satisfies $\mathbf{H}_1$ and has a discontinuity in (x, x). Because of this discontinuity, we cannot find and characterize singular strategies by way of differentiation. Instead, a simple observation of orders of magnitude will show that (i) singular strategies coincide with the ones given by the within-host model and (ii) their "character" is the same as in the context of the within-host model. In particular, all convergence stable singular traits are uninvadable. In symbols, $\mathscr{C}^{\bullet *} = \mathscr{C}^*$.

To see that this is indeed the case, we first consider $p^* \in \mathscr{C}^*$. Since the within-host CSS p^* (locally) minimizes $\hat{T}(p)$, we can find a neighbourhood of p^*, say U, such that for $q \in U \setminus \{p^*\}$ we have $\hat{T}(q) > \hat{T}(p^*)$. So if $q \in U$ then

$$r_{p^*}(q) = s_{p^*}(q) - \beta(p^*)\hat{I}(p^*)\phi(q, p^*).$$

Since $s_p(q)$ is differentiable as a function of q in the point $q = p$ and $s_p(p) = 0$, we have $s_{p^*}(q) = \mathscr{O}(\varepsilon)$ for $q = p^* + \mathscr{O}(\varepsilon)$. The term $\beta(p^*)\hat{I}(p^*)\phi(q, p^*)$, on the other hand, is $\mathscr{O}(1)$ and so $r_{p^*}(q) < 0$ when $q = p^* + \mathscr{O}(\varepsilon) \in U$. That is, p^* is an ESS.

To see that p^* is also an evolutionary attractor we first take $\overline{p} < p^*$ such that $\hat{T}'(q) \leq 0$ for $q \in [\overline{p}, p^*]$. For $p \in (\overline{p}, p^*)$ and $\varepsilon > 0$ small enough so that $\overline{p} < q_- = p - \varepsilon$ we have $\hat{T}(q_-) > \hat{T}(p)$. Similarly, for $\varepsilon > 0$ such that $q_+ = p + \varepsilon < p^*$ we have $\hat{T}(p) > \hat{T}(q_+)$. Hence, traits that move closer to p^* do better at the within-host level than the residents, while the traits that move away from p^* do worse. We therefore have

$$\begin{aligned} r_p(q_+) &= s_p(q_+) + \beta(q_+)\hat{I}(p)\phi(p, q_+), \\ r_p(q_-) &= s_p(q_-) - \beta(p)\hat{I}(p)\phi(q_-, p) \end{aligned}$$

and with the same comparison of orders of magnitude of the two terms on the RHS we find that p^* is locally attracting from the left.

In a similar fashion we find that p^* is (locally) attracting from the right and so p^* is indeed a CSS. Furthermore, these last arguments also show that (i) there can be no evolutionarily stable strategies but the elements of $\mathscr{C}^*$ (if $p \notin \mathscr{C}^*$ then $r_p(q)$ will be positive, for ε small enough, on at least one of the intervals $(p - \varepsilon, p)$ or $(p, p + \varepsilon)$) and (ii) there are no other singular strategies but the extrema of $\hat{T}$. Moreover, in the case when $\mathscr{C}^{\bullet *}$ contains more than one point, it is the traits that (locally) maximize $\hat{T}$ that separate the domains of attraction, exactly as is the case for within-host evolution.

We conclude this subsection with two examples.

Example 4 Let us take $\lambda = 1, k = 10, c = 0.1, d = 2$ in the within-host model and $\delta = 0.1, b = 1$ for the between-host model. We furthermore choose $\mu(p) = \frac{p}{p+1}$ and

$$\begin{aligned} \alpha(p) &= \frac{k}{\lambda}\hat{V}(p), \\ \beta(p) &= \hat{V}(p) + \hat{T}^*(p). \end{aligned}$$

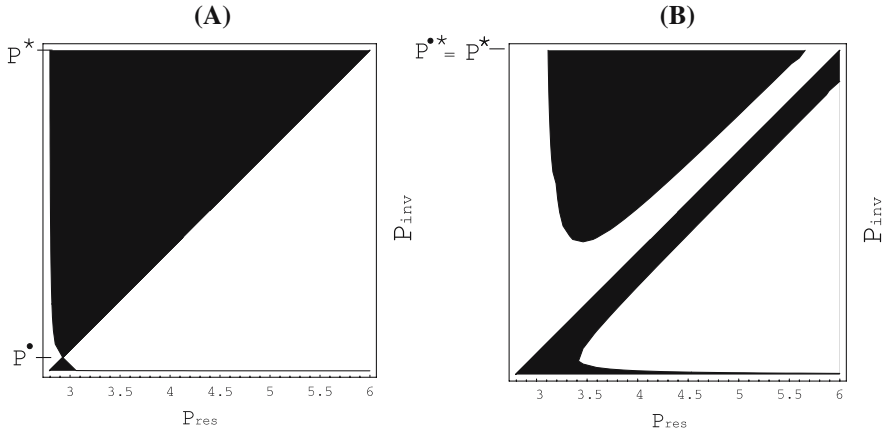

Fig. 4 **a** PIP for the single infection model, along with the within-host CSS, $p^* = p_{\max}$. **b** PIP for the superinfection model with ϕ as in (11). The *white* and the *black areas* correspond, respectively, to the regions in $(p_{\text{res}}, p_{\text{inv}})$ space that yield positive and negative values of the invasion exponent

In this case, $\hat{T}(p)$ is a strictly decreasing function. Taking into account the restriction $0 < \hat{T}(p) < T_0$, we therefore find a lower bound, $p_{\min}$, for the admissible trait values at the within-host level, but the model gives no upper bound on p (see also Example 1). We assume, however, that there is a physiological restriction $p_{\max} = 6$ on the values of the pathogen production rate. The condition $0 < \hat{S}(p) < S_0$ yields a lower bound $p'_{\min}$ but no upper bound. Since in this case $p_{\min} < p'_{\min} < p_{\max}$, we plot the pairwise invasibility plots on $[p'_{\min}, p_{\max}] \times [p'_{\min}, p_{\max}]$.

Figure 4 shows two pairwise invasibility plots: in (A) we see the PIP for the single infection model, while (B) shows the PIP for the superinfection model, where the superinfection function is the discontinuous function given in (11). The white and the black areas correspond, respectively, to the regions in (p, q) space that yield positive and negative values of the invasion exponent. The single infection model in this case yields a unique CSS, $p^\bullet \in (p'_{\min}, p_{\max})$, while the evolution at the within-host level drives p towards the physiological maximum, $p^* = p_{\max}$. We see that, when superinfections are modeled with a discontinuous superinfection function, p^* is indeed the only CSS at the host population level, i.e., $p^{\bullet *} = p^*$.

Example 5 We now take the same parameter values and trade-off functions α and β as in the previous example, only now $\mu(p) + d = d\mathrm{e}^{0.2p}$. Taking into account the restrictions $0 < \hat{T}(p) < T_0$ and $0 < \hat{S}(p) < S_0$, we first find the intervals of feasible values of p, $[p_{\min}, p_{\max}]$ and $[p'_{\min}, p'_{\max}]$, respectively. For the chosen trade-off functions μ, α and β and the chosen parameter values we find that $p_{\min} < p'_{\min}$ and $p'_{\max} < p_{\max}$ and so we plot the pairwise invasibility plots on $[p'_{\min}, p'_{\max}] \times [p'_{\min}, p'_{\max}]$.

In this case, p^* is unique (and will hence be a global attractor for the within-host evolution) and takes some intermediate value. The single infection model, however, admits two CSSs. The pairwise invasibility plots for this example are shown in Fig. 5. Figure 5a shows the PIP for the single infection model, along with p^*. With a

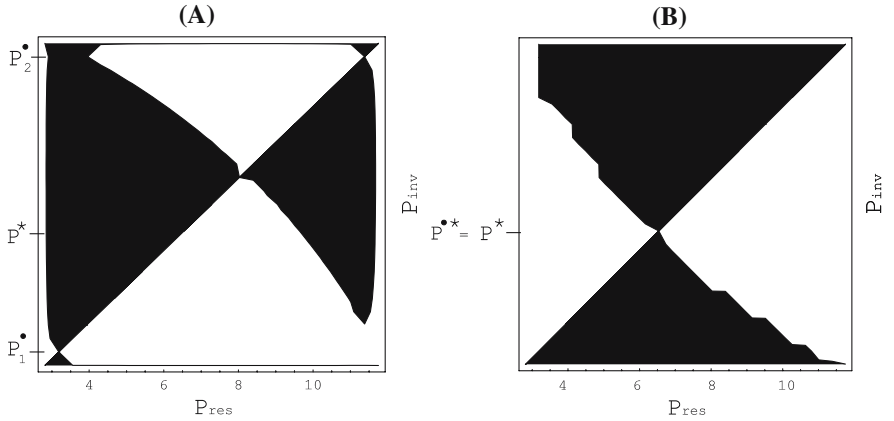

Fig. 5 Pairwise invasibility plots for **a** the single infection model (with p^* also shown) and **b** the superinfection model with ϕ as in (11). The *white* and the *black areas* correspond, respectively, to the regions in $(p_{\text{res}}, p_{\text{inv}})$ space that yield positive and negative values of the invasion exponent

discontinuous superinfection function, p^* becomes the only CSS also at the host population level, as is demonstrated in Fig. 5b (the scalloped curve in Fig. 5b is most likely a numerical artefact).

3.2 The "mechanistic" case

Suppose now that $\psi(x, y)$ is a nonnegative function that satisfies $\mathbf{H}_1$ and is furthermore continuous as a function of y in the point $y = x$.

Since $\psi(x, y) = 0$ whenever $x \leq y$, the function $y \mapsto \psi(x, y)$ is differentiable from the right in $y = x$. In fact,

$$\lim_{y \downarrow x} \frac{\psi(x, y)}{y - x} = 0.$$

We shall now furthermore assume that

$\mathbf{H}_2$. The limits

$$\lim_{y \uparrow x} \frac{\psi(x, y)}{y - x} \quad \text{and} \quad -\lim_{y \downarrow x} \frac{\psi(y, x)}{y - x}$$

exist, are either negative or zero, and are for any feasible x equal to each other. We denote them by $D(x)$.

Note that, when $D(x) = 0$ for some x, the function $y \mapsto \psi(x, y)$ is differentiable in the point $y = x$. In this subsection we consider the case when $D(x)$ is not identically zero, leaving the case $D = 0$ for the next subsection.

The reason for posing the additional assumption $\mathbf{H}_2$ on ψ is twofold. Firstly, we believe that this condition poses no real restriction, as we shall see in Sect. 5 that this

assumption is fulfilled for the most natural choices of superinfection functions, i.e., the superinfection functions obtained when modeling the initial stages of a superinfection as a stochastic birth-and-death process. Secondly, we shall now prove that with this additional assumption, the function $\Phi(p, q)$ in (8) is differentiable as a function of q in the point $q = p$, which means that we can, at least in principle, determine the singular strategies analytically.

Let us mention, however, that we have also performed a series of numerical experiments (not shown here) with continuous superinfection functions that do not satisfy assumption $\mathbf{H}_2$, and found that all the results that we shall present below (apart from, of course, analytical characterization of singular points) hold also for such functions.

Lemma 1 *Let $y \mapsto \psi(x, y)$ be nonnegative and continuous in $y = x$. Furthermore, assume that ψ satisfies $\mathbf{H}_1$ and $\mathbf{H}_2$ and let*

$$D(x) = \lim_{y \uparrow x} \frac{\psi(x, y)}{y - x} = -\lim_{y \downarrow x} \frac{\psi(y, x)}{y - x}. \tag{12}$$

The function

$$\Phi(p, q) = \beta(q)\psi(\hat{T}(p), \hat{T}(q)) - \beta(p)\psi(\hat{T}(q), \hat{T}(p))$$

is differentiable as a function of q in the point $q = p$.

Proof Let us verify that the limits

$$\lim_{\varepsilon \downarrow 0} \frac{\Phi(p, p - \varepsilon)}{-\varepsilon} \quad \text{and} \quad \lim_{\varepsilon \downarrow 0} \frac{\Phi(p, p + \varepsilon)}{\varepsilon}$$

exist and are equal.

We shall only consider the case when $\hat{T}'(p) > 0$, i.e., in a neighbourhood of p, potentially successful invaders are precisely the ones that lie to the left of p. The other cases are considered in a similar manner.

In this case, there exists an interval, say (p, p'), so that for every $\varepsilon > 0$ for which $p < p + \varepsilon < p'$ we have $\hat{T}(p) < \hat{T}(p + \varepsilon) < \hat{T}(p')$ and so

$$\begin{aligned}\Phi(p, p + \varepsilon) &= \beta(p + \varepsilon)\psi(\hat{T}(p), \hat{T}(p + \varepsilon)) - \beta(p)\psi(\hat{T}(p + \varepsilon), \hat{T}(p)) \\ &= -\beta(p)\psi(\hat{T}(p + \varepsilon), \hat{T}(p)).\end{aligned}$$

Hence

$$\lim_{\varepsilon \downarrow 0} \frac{\Phi(p, p + \varepsilon)}{\varepsilon} = \beta(p) D(\hat{T}(p)) \frac{\mathrm{d}\hat{T}}{\mathrm{d}q}\Big|_{q=p}.$$

Furthermore, there exists an interval (p'', p) so that for every $\varepsilon > 0$ such that $p'' < p - \varepsilon < p$ we have $\hat{T}(p'') < \hat{T}(p - \varepsilon) < \hat{T}(p)$, and so

$$\begin{aligned}\Phi(p, p - \varepsilon) &= \beta(p - \varepsilon)\psi(\hat{T}(p), \hat{T}(p - \varepsilon)) - \beta(p)\psi(\hat{T}(p - \varepsilon), \hat{T}(p)) \\ &= \beta(p - \varepsilon)\psi(\hat{T}(p), \hat{T}(p - \varepsilon)).\end{aligned}$$

Hence

$$\lim_{\varepsilon\downarrow 0}\frac{\Phi(p,\,p-\varepsilon)}{-\varepsilon}=\beta(p)D(\hat{T}(p))\frac{\mathrm{d}\hat{T}}{\mathrm{d}q}\Big|_{q=p}.$$

□

We can therefore compute the selection gradient and find that

$$\begin{aligned}\frac{\partial r_p(q)}{\partial q}\Big|_{q=p}&=\frac{\partial s_p(q)}{\partial q}\Big|_{q=p}+\beta(p)D(\hat{T}(p))\hat{I}(p)\frac{\mathrm{d}\hat{T}}{\mathrm{d}q}\Big|_{q=p}\\&=-\beta(p)\left[\frac{\mathrm{d}\hat{S}}{\mathrm{d}q}\Big|_{q=p}-D(\hat{T}(p))\hat{I}(p)\frac{\mathrm{d}\hat{T}}{\mathrm{d}q}\Big|_{q=p}\right],\end{aligned}\tag{13}$$

where we have used in the second line that

$$\frac{\partial s_p(q)}{\partial p}+\frac{\partial s_p(q)}{\partial q}=0$$

holds on the line $q=p$.

Singular points are obtained by putting $\frac{\partial r_p(q)}{\partial q}\big|_{q=p}=0$.

If $D(\hat{T}(p))=0$ for some feasible trait value p, then p is a singular strategy if and only if it is a singular strategy for the selection at the between-host level in the context of the single infection model. If $D(\hat{T}(p))\neq 0$, then p can only be a singular strategy when (i) it is both a singular strategy for selection at the between-host level (as given by the single infection model) and for the within-host selection or (ii) selection at the within-host level works in a different direction than selection at the between-host level (in the context of the single infection model). Indeed, if selection tends to increase p at both levels then $\hat{S}'(p)<0$ and $\hat{T}'(p)<0$, which gives a positive selection gradient in $q=p$. Similary, there can be no singular points in areas where both selection pressures tend to decrease p. In particular (13) implies that when p^* and $p^\bullet$ are unique, the singular strategy lies in-between the within-host CSS and the CSS obtained with the single infection model (cf. Example 6).

Remark 3 Note that when $\frac{\mathrm{d}\hat{S}}{\mathrm{d}q}\Big|_{q=p}=0$, then

$$\operatorname{sign}\frac{\partial r_p(q)}{\partial q}\Big|_{q=p}=-\operatorname{sign}\frac{\mathrm{d}\hat{T}}{\mathrm{d}q}\Big|_{q=p},$$

which can be interpreted as meaning that superinfection leads to an increased virulence: indeed, if $\hat{S}$ has a unique minimum, $p=p^\bullet$, then evolution ends in $p^\bullet$ if superinfections are ignored, but when we take superinfections into account, we move from $p^\bullet$ in the direction of decreasing $\hat{T}$ and, for reasonable choices of how α depends on the internal state $(\hat{T},\hat{T}^*,\hat{V})$, this means increasing α. The increased CSS value

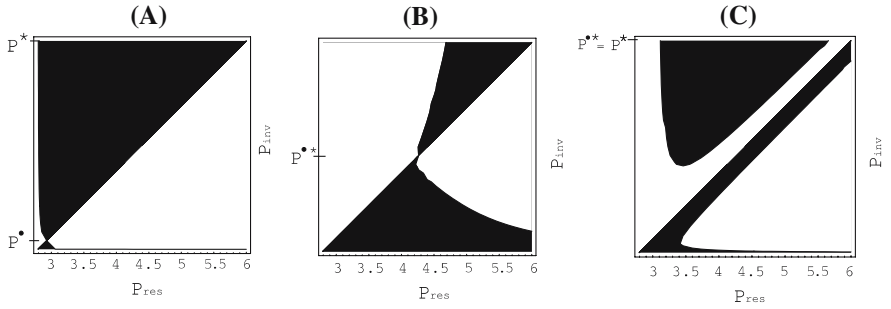

Fig. 6 Pairwise invasibility plots for **a** single infection model **b** the "mechanistic" case and **c** the "jump" case. The CSS for the mechanistic case, $p^{\bullet *}$, lies inbetween $p^{\bullet}$ and p^*

of virulence as a consequence of superinfections has also been observed by others [34,37,40,41].

In contrast with the single infection case and the "jump" case, however, we may encounter convergence stable singular traits that are invadable, viz. branching points (cf. Fig. 8). Such traits turn out to be very interesting for the course of evolution since they evoke evolutionary branching and potentially lead to evolutionary coexistence of pathogen traits [18,31]. We shall return to this important point in Sect. 4.

Let us now present two examples. In both of the examples we take superinfection functions with

$$\psi_n(x, y) = \begin{cases} 1 - \left(1 - \dfrac{kx}{c + kx} + \dfrac{ky}{c + ky}\right)^n, & y < x, \\ 0, & \text{otherwise} \end{cases} \tag{14}$$

for some $n \in \mathbb{N}$. As we shall see in Sect. 5, these are the functions obtained when we describe the initial stages of a superinfection as a stochastic birth-and-death process.

Example 6 In this example we basically repeat the experiment from Example 4, only this time with a continuous superinfection function. In Fig. 6 we plot three PIPs: for comparison, (a) and (c) show, respectively, the PIP for the single infection model and the superinfection model with a discontinuous superinfection function (i.e., the "jump" case), while (b) shows the PIP when the superinfection function is constructed with ψ_n in (14), taking $n = 100$. In this case, the convergence stable singular strategy $p^{\bullet *}$ is also an ESS, hence a CSS. Note however, that the uninvadability of $p^{\bullet *}$ is indeed local, with the interval of neighbouring traits that cannot invade $p^{\bullet *}$ being very small.

Example 7 We now take the same parameter values and trade-off functions as in Example 5. In Fig. 7 we plot six pairwise invasibility plots. For comparison, we include the PIP for the single infection model (Fig. 7a) and the PIP for the "jump" case (Fig. 7f). In Fig. 7b–e we construct PIPs by taking the superinfection functions ψ_n given by (14) with, respectively, $n = 10$, 50, 100 and 1,000. While in Fig. 7b and c the convergence stable singular traits are also ESSs, this is no longer the case in Fig. 7d and e (but the interval on which the convergence stable singular trait is invadable is again very small).

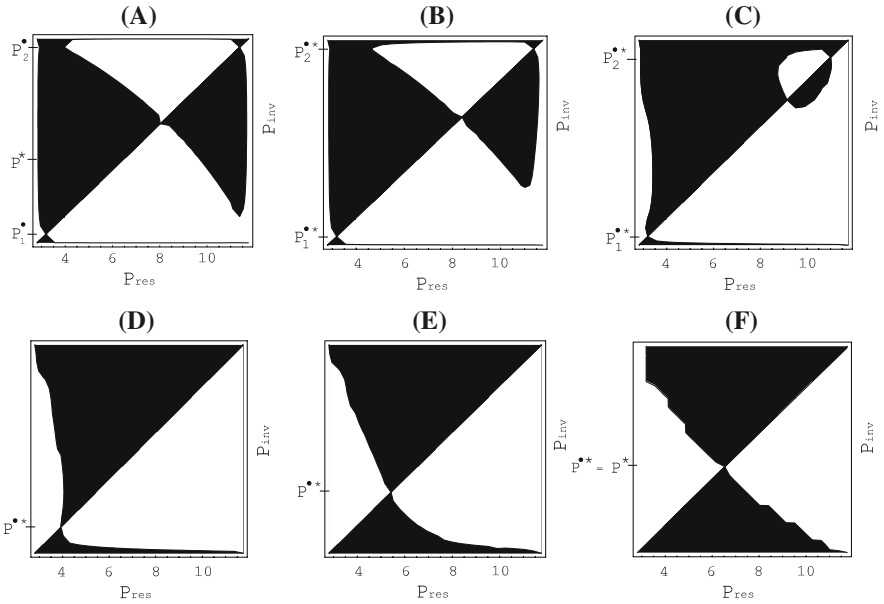

Fig. 7 Pairwise invasibility plots for **a** single infection model **b–e** superinfection model with continuous superinfection functions derived from (14) with, respectively, $n = 10, 50, 100$ and $1,000$, **f** the "jump" case

3.3 The "smooth" case

We have seen that, in the "jump" case, a slightly better within-host competitor has a large advantage at the host population level and, as a consequence, singular strategies at the host population level coincide with the singular strategies of the within-host model.

By contrast, when $D(x)$ in (12) is equal to zero for every feasible x, we find that

$$\frac{\partial r_p(q)}{\partial q}\Big|_{q=p} = \frac{\partial s_p(q)}{\partial q}\Big|_{q=p}, \tag{15}$$

or, in words, the selection gradient is exactly the same as when the possibility of superinfections is ignored.

Equality (15) then implies the following: (i) singular strategies of the superinfection model coincide with the ones given by the single infection model, (ii) singular strategies that are (local) evolutionary attractors in the context of the single infection model are also attractors for the monomorphic dynamics in the context of the superinfection model and (iii) singular traits that are (local) repellors in the context of the single infection model remain repellors also when the superinfections are taken into account.

In those points in which the selection gradient vanishes (15) offers no insight into the course of evolution. And while the convergence stable strategies are necessarily ESSs in the context of the single infection model, this may no longer be the case for the superinfection model. We may, in fact, encounter evolutionary branching points, which

is impossible in the context of the single infection model. Note, however, that when $y \mapsto \phi(x, y)$ and $y \mapsto \phi(y, x)$ are differentiable twice in $y = x$, these derivatives are necessarily zero and hence the singular traits have precisely the same character as in the context of the single infection model.

4 The (non)existence of an optimization principle and coexistence of pathogen traits

For selection at the two isolated levels, the pessimization principle implies that the convergence stable strategies are necessarily uninvadable, while evolutionary repellors are invadable. In Sect. 3.1 we showed that this is also the case if the superinfection function is discontinuous, i.e., when a slightly better within-host competitor has a large advantage over the resident. One might thus be tempted to believe that an optimization (or pessimization) principle exists, at least in the "jump" case, also for the superinfection model. This is, in fact, not so.

If infected hosts can be reinfected, the environment the pathogens experience is no longer one dimensional as the pathogens can, conditionally on their capacity to win the within-host competition, also superinfect an already infected host. When the dimension of the environment is larger than one we can, in general, no longer expect that we will find optimization in the course of evolution [32], nor can we be sure that the pathogen population remains monomorphic at the host population level.

The (non)existence of an optimization principle can easily be recognized from a pairwise invasibility plot. Namely, if such a principle exists, the PIPs must be skew symmetric [8] (i.e., they must be invariant under reflection across the main diagonal and simultaneous change of the two signs/colors, in our case black and white). The examples of Sect. 3 (in particular, Figs. 6b, c, 7b–f) thus clearly show that the outcome of evolution can not be determined by an optimization principle when superinfections are possible (these examples in fact only show that there is no optimization in the "jump" and the "mechanistic" case. Skew symmetry is lost also in the "smooth" case, but since this case is biologically not as relevant as the other two, we do not include the PIPs).

The lack of skew symmetry implies that we encounter pairs (p, q) for which either $r_p(q) > 0$ and $r_q(p) > 0$ or $r_p(q) < 0$ and $r_q(p) < 0$. In the former case, the traits are said to be *mutually invadable*, while they are *mutually uninvadable* in the latter.

If p and q are mutually invadable, trait p is able to invade q when q is rare, but is not able to outcompete it since q can also invade p when p is rare. In such a case, a so called *protected dimorphism* arises. The evolutionary significance of dimorphisms (or, more generally, polymorphisms) depends on whether these dimorphisms that are found on the epidemiological time scale can be maintained also on the evolutionary time scale. In principle, two questions need to be answered. First, if the pathogen population is monomorphic to begin with, can polymorphisms be created and maintained in the course of evolution? And second, if the pathogen population level is polymorphic to begin with, can polymorphisms stand the test of evolutionary persistence?

Retaining the assumption of Sect. 3 that mutations are small (and rare), we begin with the first question. We shall in fact only deal with the "mechanistic" and the "jump" case, since these two cases have clear biological interpretation. Evolutionary coexistence was found by way of numerical experiments also in the "smooth" case (results not shown).

4.1 Evolutionary branching points

When the trait of an initially monomorphic pathogen population evolves, by a series of trait substitutions, into a neighbourhood of a convergence stable singular trait around which coexistence is possible, the population becomes dimorphic. If the convergence stable singular trait is also uninvadable (and hence a CSS), dimorphisms occur only as transients in the evolution towards a monomorphic pathogen population and hence have no evolutionary significance. Such dimorphisms are also called *converging* dimorphisms [18,31].

If, on the other hand, the population becomes dimorphic in a neighbourhood of a *branching point*, i.e., a singular trait that is convergence stable and invadable, the dimorphisms can not be dismissed: the two traits become more and more distinct in the course of evolution (at least for a while, that is) and can lead to coexistence of two (or more, since further branching may occur) pathogen traits on the evolutionary time scale. One also speaks of *diverging dimorphisms* [18,31].

In the "jump" case, the singular strategies are the same, and furthermore have the same character, as the ones obtained with the within-host model. And since the within-host model does not allow for branching points, neither does the superinfection model in that case. Hence, if the pathogen population is initially monomorphic, the outcome of evolution at the host population level will be the same as the outcome in a single infected host, only that perhaps the CSS is at the host population level approached also via polymorphisms, and not only through trait substitutions, as is the case within one host. However, as we will see later on, existing dimorphisms can be maintained in the course of evolution provided that some additional conditions are satisfied.

By contrast, the (biologically most relevant) mechanistic superinfection submodel, can induce branching points. In a generic situation, branching points are convergence stable singular traits $p^{\bullet *}$ for which $q \mapsto r_{p^{\bullet *}}(q)$ has a strict local minimum in $q = p^{\bullet *}$. That is, the left as well as the right second derivative of $q \mapsto r_{p^{\bullet *}}(q)$ in $q = p^{\bullet *}$ will be strictly positive. But in the present "mechanistic" situation, it is not unusual for the left and the right second derivative to have different signs (see also Appendix B for a more detailed elaboration in the context of a caricatural example)!

That this indeed does happen is demonstrated in Fig. 8c. In such a case, evolution does not stop when a sequence of trait substitutions brings the trait into the vicinity of $p^{\bullet *}$. Just as in the case of a minimum of $q \mapsto r_{p^{\bullet *}}(q)$, mutual invasibility may lead to coexistence, but now (cf. Fig. 9) there is asymmetry, with very little scope for the lower trait when compared to the scope of the upper trait.

When a population undergoes branching, the invasion exponent $r_p(q)$ is no longer useful as it presupposes a monomorphic population. Instead, we replace it by $r_{p_1,p_2}(q)$, which denotes the growth rate of the population with trait q, when introduced into a

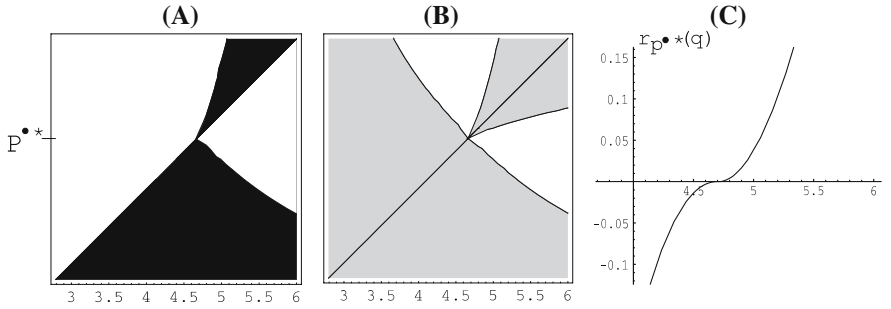

Fig. 8 **a** The PIP corresponding to the superinfection model with $\psi = \psi_{200}$ in (14). **b** *White areas* correspond to the regions in (p, q) space in which p and q are mutually invadable. *Grey regions* are the areas where $r_p(q)$ and $r_q(p)$ have opposite signs. **c** The graph of $r_{p^{\bullet *}}(q)$

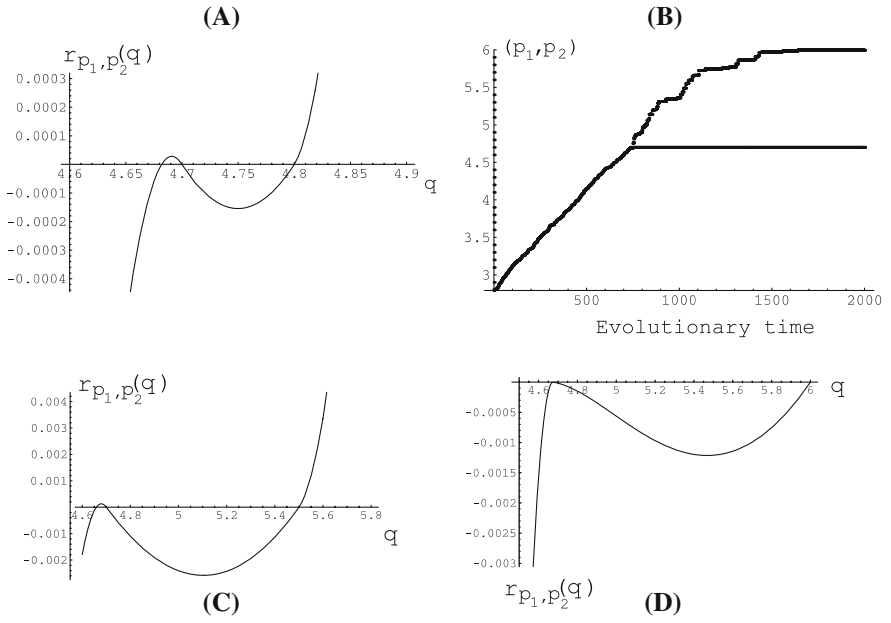

Fig. 9 **a** The graph of $r_{p_1,p_2}(q)$ with the initial dimorphism $(p_1, p_2) = (4.7, 4.8)$ shows that (p_1, p_2) can be invaded by a small mutation when $q < p_1$ or $q > p_2$. **b** The simulated evolutionary tree shows evolutionary branching leading to two distinct pathogen subpopulations. **c** The graph of $r_{4.7,5.5}(q)$. **d** The graph of $r_{4.675,6}(q)$

steady host population where traits p_1 and p_2 are resident. Note that this formalism needs to be extended if the population undergoes further branching. It is known that the dimension of the environment sets an upper bound on the number of pathogen traits that can coexist in a steady state [11]. When superinfections are taken into account, the environment is, in principle, infinite dimensional and given by $\{\hat{S}, \hat{I}_p\}$ with p in the domain of feasible trait values. Moreover, there may exist other attractors and not only steady states. In systems with multiple attractors, it is in general

not possible to predict an outcome of an invasion on the basis of the invasion criteria. However, the Tube Theorem in [17] guarantees that when the mutations are sufficiently small, the population will stay on the same attractor during the course of evolution. We shall thus not go any further into the attractors of (7) and their stability: the aim of this section is merely to demonstrate that evolutionarily stable coexistence of two pathogen traits is possible in the context of the superinfection model.

We therefore focus on the case with two resident traits and compute the invasion exponent $r_{p_1,p_2}(q)$, which takes the form

$$r_{p_1,p_2}(q) = \beta(q)\hat{S}(p_1, p_2) - (\alpha(q) + \delta) + \Phi(p_1, q)\hat{I}_{p_1}(p_1, p_2) + \Phi(p_2, q)\hat{I}_{p_2}(p_1, p_2) \tag{16}$$

where $\hat{S}(p_1, p_2)$ denotes the steady state value of the susceptible population when traits p_1, p_2 are present in the population and $\hat{I}_{p_1}, \hat{I}_{p_2}$ stand for, respectively, the nontrivial equilibrium values of the number of hosts infected by p_1 and p_2.

Using (7) we find that

$$\hat{S}(p_1, p_2) = \frac{b\Phi(p_1, p_2)}{\delta\Phi(p_1, p_2) + \beta(p_1)\beta(p_2)(\hat{S}(p_2) - \hat{S}(p_1))}, \tag{17a}$$

$$\hat{I}_{p_1}(p_1, p_2) = \frac{\beta(p_2)}{\Phi(p_1, p_2)}\left(\hat{S}(p_2) - \hat{S}(p_1, p_2)\right), \tag{17b}$$

$$\hat{I}_{p_2}(p_1, p_2) = \frac{\beta(p_1)}{\Phi(p_1, p_2)}\left(\hat{S}(p_1, p_2) - \hat{S}(p_1)\right), \tag{17c}$$

where $\hat{S}(p) = \frac{\alpha(p)+\delta}{\beta(p)}$.

Without any loss of generality we can assume that $\hat{T}(p_1) > \hat{T}(p_2)$. Hence $\Phi(p_1, p_2) = \beta(p_2)\phi(p_1, p_2)$ and (17) specifies a feasible (i.e., nonnegative and nontrivial) steady state if and only if

$$\hat{S}(p_1) < \hat{S}(p_1, p_2) < \hat{S}(p_2). \tag{18}$$

Recalling that the boundary steady states are given by

$$\hat{S}(p) = \frac{\alpha(p) + \delta}{\beta(p)} \quad \text{and} \quad \hat{I}(p) = \frac{1}{\beta(p)}\left(\frac{b}{\hat{S}(p)} - \delta\right),$$

we find that p_1 can invade the p_2-only steady state when $\hat{S}(p_1, p_2) < \hat{S}(p_2)$, while vice versa, p_2 can invade the p_1-only steady state when $\hat{S}(p_1) < \hat{S}(p_1, p_2)$. In other words, steady coexistence of two traits is possible if and only if the traits are mutually invadable. A necessary condition for this is that $\hat{S}(p_1) < \hat{S}(p_2)$ (that is, given that the trait p_2 wins the internal competition, p_1 must win the single infection between-host competition), while a sufficient condition is given in (18).

In a similar manner as finding the singular points for the monomorphic dynamics, we can determine the (internal) singular coalitions by putting

$$\frac{\mathrm{d}}{\mathrm{d}q} r_{p_1,p_2}(q)\Big|_{q=p_1} = \frac{\mathrm{d}}{\mathrm{d}q} r_{p_1,p_2}(q)\Big|_{q=p_2} = 0.$$

Note that we can depict such coalitions from a pairwise invasibility plot. Namely, each of the conditions $\frac{\mathrm{d}}{\mathrm{d}q} r_{p_1,p_2}(q)|_{q=p_1} = 0$ and $\frac{\mathrm{d}}{\mathrm{d}q} r_{p_1,p_2}(q)|_{q=p_2} = 0$ represents a curve (called isocline) in a (p, q) plane. If the isoclines intersect in the white area, where coexistence is possible, we obtain a feasible singular coexistence. The (un)invadability of a singular coalition can be (relatively) easily verified. Namely, a singular coalition is evolutionarily stable if and only if the traits that form it are uninvadable. Convergence stability, however, is not so straightforward, but the reader can find some results in [29]. We shall refrain from analytic treatment of singular coalitions and proceed by way of numerical examples.

Example 8 We take the parameter values and the trade-off functions as in Example 4 and investigate numerically whether evolutionary branching points exist if the superinfection function is given by (25) for some $n \in \mathbb{N}$. In this case, the function $p \mapsto \hat{T}(p)$ is a strictly decreasing function, which implies that the within-host CSS will take the value of the physiological restriction on trait values, in this case taken to be $p_{\max} = p^* = 6$.

We found that for low values of n (that is, when the host is reinfected by a relatively low dose) the convergence stable singular strategy will also be an ESS and will hence give rise only to converging dimorphisms. When n becomes larger, the convergence stable steady state becomes invadable, which can give rise to evolutionary coexistence of two distinct values of the pathogen production rate. This is demonstrated in Fig. 8, where $n = 200$. The convergence stable strategy for the monomorphic dynamics was found numerically and takes the value $p^{\bullet *} = 4.69345$. Note that $p^{\bullet *}$ in Fig. 8 is a branching point, however, of an unusual type as it is invadable from above, but not from below (see also Appendix B).

The simulated evolutionary tree in Fig. 9b shows monomorphic evolution towards $p^{\bullet *}$, after which branching occurs. In general, branching does not guarantee that we will find coexistence on the evolutionary time scale since one branch may go extinct and the other may evolve towards a monomorphic ESS. However, extinction after branching was not encountered in this case. We found that the within-host CSS, p^* (which is in this case at the physiological maximum, cf. Example 4), coexists at the host population level with trait $p = 4.675$.

Figure 9a and c shows the graph of $q \mapsto r_{p_1,p_2}(q)$ for two choices of resident coalitions (p_1, p_2): Fig. 9a shows the initial dimorphism and the invasion ability of neighbouring traits, while Fig. 9c is taken later on in the evolutionary time with $(p_1, p_2) = (4.7, 5.5)$. Note that the scope for successful invaders around the lower trait is indeed very small, compared to the scope for successful invaders around the upper trait. Figure 9b shows the result of a single simulation. Note also that the lower branch appears to be constant but is fact not so since $p^{\bullet *} = 4.69345$, but the lower trait in the evolutionarily stable coexistence takes the value $p = 4.675$.

We have performed a large number of simulations, which suggest that the coalition (4.675, 6) is convergence stable as well as uninvadable (the uninvadability is also seen in Fig. 9d).

In the near future we intend to study the adaptive dynamics for the case of a bounded β, e.g.,

$$\beta(p) = \frac{k\hat{V}(p)}{1+\hat{V}(p)}$$

and with no upper bound imposed on p at the within-host level (see Appendices A and B for motivation).

4.2 Maintenance of dimorphisms through the evolutionary time

We have demonstrated that the mechanistic case allows for evolutionary branching as well as evolutionary coexistence. We have furthermore shown that the "jump" case does not induce branching points; natural selection will in this case drive p towards a local minimum of $\hat{T}(p)$, only that perhaps convergence occurs also via dimorphisms and not only through trait substitutions.

Branching points are neither necessary nor sufficient for maintenance of polymorphisms. If one allows for mutations of arbitrary size, or starts off with a polymorphic population, evolutionary coexistence is possible also in the "jump" case. The only candidates for stable coalitions, however, are the minima of $\hat{T}$ that are mutually invadable (a necessary condition for such coexistence is therefore that $q \mapsto \hat{T}(p)$ has more than one local minimum). This can be seen if we use (17b) to rewrite (16) as

$$r_{p_1,p_2}(q) = s_{p_2}(q) - \left(\frac{\beta(q)}{\beta(p_2)}\Phi(p_1,p_2) - \Phi(p_1,q)\right)\hat{I}_{p_1}(p_1,p_2) + \Phi(p_2,q)\hat{I}_{p_2}(p_1,p_2) \tag{19a}$$

and (17c) to write

$$r_{p_1,p_2}(q) = s_{p_1}(q) + \left(\frac{\beta(q)}{\beta(p_1)}\Phi(p_1,p_2) + \Phi(p_2,q)\right)\hat{I}_{p_2}(p_1,p_2) + \Phi(p_1,q)\hat{I}_{p_1}(p_1,p_2). \tag{19b}$$

With the same reasoning as in Sect. 3.1 we can then show that the only convergence stable coalitions are the ones consisting of two local minima of $\hat{T}(p)$: if the mutations are small, i.e., lie in $(p_1-\varepsilon, p_1+\varepsilon)\cup(p_2-\varepsilon, p_2+\varepsilon)$ for some $\varepsilon > 0$, then each successful mutation will bring either p_1 or p_2 closer to a local minimum. Note, however, that in a generic case we will have either $\hat{T}(p_1) < \hat{T}(p_2)$ or $\hat{T}(p_2) < \hat{T}(p_1)$, but the better of the two at the within-host level may not outcompete the other at the

host population level, since the terms $s_{p_1}(q)$ and $s_{p_2}(q)$ can only be neglected in small enough neighbourhoods.

Finally, we observe that Nowak and May showed in [37] that in the "jump" case a continuum of traits can coexist on the epidemiological, as well as evolutionary, time scale. In [37], Nowak and May assume that mutations are uniformly distributed. In the present paper, where the mutations are assumed to be small, we find that the number of local minima of $\hat{T}$ sets an upper bound for the number of traits that can coexist on the evolutionary time scale. In Appendix A we give a simple example that clarifies how the size of mutations is indeed crucial for these conclusions.

5 The dynamics in the initial stages of a superinfection

In the initial stages of a superinfection, the invading trait q is likely to be present only in small quantities. Hence, even when $\hat{T}(q) < \hat{T}(p)$ (and so the newly introduced trait has the potential to outcompete the resident trait), trait q may go extinct due to demographic stochasticity in the initial stages of a superinfection, when it is still rare.

In this section we supplement the deterministic model by describing the initial stages of an invasion as a stochastic birth-and-death process. This will lead us to continuous (but not differentiable) superinfection functions, hence the word "mechanistic" for this case.

So let us begin by assuming that only one free pathogen with trait q is introduced into a host already infected by trait p. If we assume that the trait p resides at a stable equilibrium, then the new trait q is introduced into an environment given by the steady state value of $\hat{T}(p)$,

$$\hat{T}(p) = \frac{c}{k(\mathscr{B}_0(p) - 1)}. \tag{20}$$

The probability with which the clan of this initially introduced pathogen survives in an already infected host, is given as the smallest fixed point of a generating function [22]. In order to compute it, we must first derive the probabilities π_n with which one free pathogen with trait q will produce n new pathogens.

Now, in order to reproduce, a pathogen must bind to an uninfected target cell. This happens with probability

$$\frac{k\hat{T}(p)}{k\hat{T}(p) + c}.$$

When the pathogen enters a target cell, its survival relies on the survival of the target cell that hosts it. The life span of a target cell infected with trait q is exponentially distributed with parameter $(\mu(q)+d)$. The infected target cell produces free pathogens according to a Poisson process with parameter q. So the probability density that an infected target cell lives t units of time and in that time produces n offspring equals

$$(\mu(q) + d)\mathrm{e}^{-(\mu(q)+d)t}\mathrm{e}^{-qt}\frac{q^n t^n}{n!}.$$

Accounting for all possible times t, we arrive at the following expression for π_n,

$$\pi_n = \frac{k\hat{T}(p)}{k\hat{T}(p)+c}\int_0^\infty (\mu(q)+d)\mathrm{e}^{-(\mu(q)+d)t}\mathrm{e}^{-qt}\frac{q^n t^n}{n!}\mathrm{d}t, \tag{21}$$

which is valid for $n \geq 1$. The probability of having no offspring at all is, however, not given by π_0, as the pathogen may never reproduce simply because it never enters an uninfected target cell. Since the probability with which the pathogen dies before it binds to a cell equals $\frac{c}{k\hat{T}(p)+c}$, we thus obtain the following generating function $G(z)$,

$$G(z) = \frac{c}{k\hat{T}(p)+c} + \sum_{n=0}^{\infty}\pi_n z^n,$$

which, by using (21) and interchanging the order of summation and integration, can be written as

$$G(z) = \frac{c}{k\hat{T}(p)+c} + \frac{k\hat{T}(p)}{k\hat{T}(p)+c}\cdot\frac{1}{1+\mathscr{B}_0(q)(1-z)}. \tag{22}$$

The probability with which the clan of the invading pathogen goes extinct is given as the smallest solution of $G(z) = z$ (see [22] for details). Whether this solution lies in [0, 1] (and hence gives a meaningful value for a probability), depends on the value of $G'(1)$, which equals the invaders within-host reproduction ratio in the environment set by the resident,

$$\mathscr{R}_0^w(q)(\hat{T}(p)) = \frac{k\hat{T}(p)}{c+k\hat{T}(p)}\mathscr{B}_0(q).$$

If $\mathscr{R}_0^w(q)(\hat{T}(p)) \leq 1$, the clan will go extinct with certainty. If, on the other hand, $\mathscr{R}_0^w(q)(\hat{T}(p)) > 1$, the invasion will be successful with nonzero probability.

Let $P_1(p, q)$ denote the probability of extinction of trait q, following an introduction of a single free pathogen into an environment set by the resident trait p. Using (22) and (20) we obtain

$$P_1(p,q) = \min\left\{1, \frac{c}{c+k\hat{T}(p)} + \frac{1}{\mathscr{B}_0(q)}\right\} = \min\left\{1, 1-\frac{1}{\mathscr{B}_0(p)}+\frac{1}{\mathscr{B}_0(q)}\right\}.$$

Note that, having rewritten $P_1(p, q)$ in terms of burst sizes only, we can now also say that the invading trait has a nonzero probability of success only when its burst size exceeds the burst size of the resident trait p. We also observe that (i) $P_1(p, p) = 1$, as it should be since the resident trait resides at a stable equilibrium, and (ii) when $\mathscr{B}_0(q) \to \infty$, the invading trait will survive with certainty, provided that the pathogen initially introduced makes it to an uninfected target cell. The probability of extinction

must therefore equal the probability with which the pathogen dies before it enters a target cell. And indeed we find that

$$\lim_{\mathscr{B}_0(q)\to\infty} P_1(p,q) = \frac{c}{c+k\hat{T}(p)}.$$

Let now $\phi_1(p,q)$ denote the complementary probability that the clan of one free pathogen with trait q survives in the environment set by the resident trait. This is then given by

$$\phi_1(p,q) = \begin{cases} \dfrac{1}{\mathscr{B}_0(p)} - \dfrac{1}{\mathscr{B}_0(q)}, & \mathscr{B}_0(p) < \mathscr{B}_0(q), \\ 0, & \text{otherwise.} \end{cases} \tag{23}$$

When n particles are introduced, therefore, the probability of survival equals

$$\phi_n(p,q) = \begin{cases} 1 - P_1^n(p,q), & \mathscr{B}_0(p) < \mathscr{B}_0(q), \\ 0, & \text{otherwise} \end{cases} \tag{24}$$

or, to continue the thread of previous sections, rewritten in terms of $\hat{T}(p)$ and $\hat{T}(q)$,

$$\phi_n(p,q) = \begin{cases} 1 - \left(1 - \dfrac{k\hat{T}(p)}{c+k\hat{T}(p)} + \dfrac{k\hat{T}(q)}{c+k\hat{T}(q)}\right)^n, & \hat{T}(q) < \hat{T}(p), \\ 0, & \text{otherwise.} \end{cases} \tag{25}$$

In the limit when the number of initially introduced pathogens goes to infinity we have

$$\lim_{n\to\infty} \phi_n(p,q) = \begin{cases} 1, & \hat{T}(q) < \hat{T}(p), \\ 0, & \text{otherwise.} \end{cases}$$

That is, when a large number of pathogens with trait q is introduced, the deterministic description gives the full story: if the newly introduced trait goes extinct, it is because it loses the competition within the host and not due to bad luck while still rare.

Note that this description makes the superinfection functions $\phi = \phi_n(p,q)$ continuous (but not differentiable) as functions of q in the point $q = p$. The fact that they are increasing as functions of $\mathscr{B}_0(q)$, implies that the traits that significantly increase the burst size (i.e., the ones that significantly reduce the steady state level of uninfected target cells within the host) also have a better chance of surviving in the host than the traits which are only slightly better within-host competitors than the resident. When the number of initially introduced mutants goes to infinity, we obtain a superinfection function with a jump discontinuity in $q = p$, which furthermore does not discriminate among the winning strategies: every trait that reduces the steady state level of target cells to a lower level than the resident trait succeeds with probability one.

6 Concluding remarks

The individual host is a cul-de-sac for parasites that reproduce within it and hence parasite persistence relies on host-to-host transmission. When the risk that competitors enter the same host is negligible, this need for transmission sets an optimal level for within-host prudence. But if the same host can be re-infected by another strain, the parasites are confronted with a version of the milker-killer dilemma [45].

In order to evaluate the balance of the various evolutionary forces one needs, to begin with, a somewhat detailed description of the status of an infected host and how this status changes in the course of time. In principle, the status should incorporate all relevant information about parasite burden, immune response and harm done. Pragmatic first steps are usually based on a very caricatural description in terms of just a few variables, with dynamics generated by very simple deterministic rules. The link to the host population level is then made by specifying how the mortality of the host and the transmissibility of the parasite (upon contact of the host with another, susceptible, host) depend on the status. Various authors use terms like "nested" or "embedded" models to indicate that a submodel for within-host dynamics is used as a building block for an epidemic model at the host population level (see [1] for a survey and references).

In order to incorporate the effects of reinfection by another strain, one needs to address a number of rather subtle issues, having to do with the "dose" of parasites that enters the host at transmission. A first point is that demographic stochasticity may play a major role right away, simply because the dose is small. A second point is that there is no clue as to the choice of an initial value for the deterministic variable representing the new strain in the host status.

In the present paper, building on the work of Gilchrist and Coombs [19], we adopted a consistent series of simplifying assumptions to obtain a tractable model. First of all, we assumed that mutations are so rare at the epidemiological time scale that we can use the Adaptive Dynamics description of evolution as a sequence of trait substitutions and, possibly, branching. We consider an SI model, i.e., we ignore an immune response that clears the infection. This allows us to assume that within-host dynamics is so fast compared to (the time scale of) the host contact process and host demography, that we can neglect transients and focus on (steady state) attractors. Since the within-host model exhibits competitive exclusion, an added bonus is that, in the deterministic description, we have superinfection rather than coinfection (i.e., a newly arriving strain fails to have any effect if it is a weaker competitor, while outcompeting the resident strain instantaneously if it is a better competitor). This no-yes dichotomy is shaded down by demographic stochasticity, in the sense that "no" remains "no" but "yes" becomes "possibly", with the probability depending on the degree of competitive superiority. We use the branching process corresponding to the linearized within-host model to compute how this probability of successful take-over depends on both the difference in competitive ability and the dose (Mosquera and Adler [34] introduced this idea in the present context in their Appendix B, but, curiously, did not elaborate the link between the branching process and the mechanistic within-host submodel in their Appendix A; also, note that Pugliese [40,41] questions the absoluteness of the "no" and chooses to work with a smooth superinfection function).

Clearly all of these assumptions are open for debate (see [6,42] for inspiration). They are made in order to make progress, not as a terminus. The main conclusions that we derived from these assumptions are

(i) superinfection leads to increased virulence and
(ii) superinfection may lead to evolutionarily stable coexistence of different strains.

While the first is exactly what one would expect, the second is a bit surprising, given the fact that Pugliese [40,41] found converging rather than diverging dimorphisms. It appears that the mechanistic within-host model leads to a richer repertoire of adaptive dynamics than a phenomenological trade-off between transmissibility and host mortality. We are thus inclined to answer the rhetoric question in the title of the stimulating paper [15] with "yes", as did the authors!

Various attempts of devising meaningful, yet tractable, within-host submodels have been made [1,5,9,15,19,24,33,38,43] and it has been clarified how superinfection can be seen as a limiting case of coinfection [9,34]. In our view, it is now a great challenge to incorporate immunity. If the infectious period has a finite length due to the fact that the parasites are ultimately eliminated by the immune system, it becomes much more difficult, if not impossible, to postulate time scale differences that simplify the analysis. In addition, immunity has a "long term memory" effect [20,21]. Will it be possible to determine the influence of all these factors on the evolution of infectious diseases?

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Appendix A: The influence of mutation size on the number of traits that can coexist on the evolutionary time scale

The aim of this appendix is to relate our observations made in Sect. 4.2 regarding the number of traits that can coexist in the "jump" case to the ones obtained by Nowak and May [37]. We follow [37] by making the additional assumption that the total (host) population size is constant and normalized to 1. We thus replace (1) with the single equation

$$\frac{\mathrm{d}I}{\mathrm{d}t} = \beta I(1 - I) - (\alpha + \delta)I$$

and furthermore assume that β is constant. We also forget about the within-host processes and take α as the relevant trait that may change on the evolutionary time scale as a result of mutation and selection. For two co-circulating traits α_1 and α_2, the analogue of (7) is given by

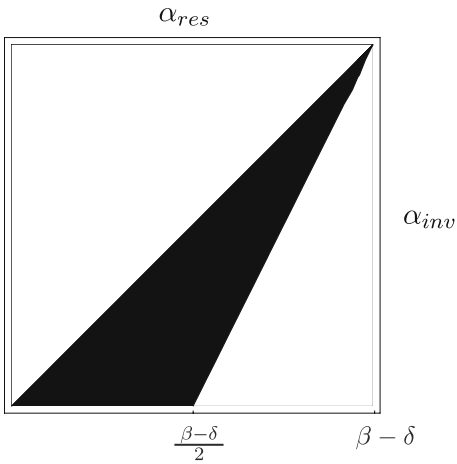

Fig. 10 Pairwise invasibility plot corresponding to (27), where the superinfection function is as in (28). *Black* and *white regions* correspond to the areas where the invasion exponent is, respectively, negative and positive

$$\begin{aligned}\frac{\mathrm{d}I_1}{\mathrm{d}t} &= \beta I_1(1-I_1-I_2)-(\alpha_1+\delta)I_1+\Phi(\alpha_2,\alpha_1)I_1I_2,\\ \frac{\mathrm{d}I_2}{\mathrm{d}t} &= \beta I_2(1-I_1-I_2)-(\alpha_2+\delta)I_2+\Phi(\alpha_1,\alpha_2)I_1I_2\end{aligned} \tag{26}$$

with $\Phi(\alpha_1,\alpha_2)=\beta\phi(\alpha_1,\alpha_2)-\beta\phi(\alpha_2,\alpha_1)$.

If α_1 is the resident trait, then the ability of α_2 to invade the environment set by α_1 is given by the invasion exponent

$$r_{\alpha_1}(\alpha_2)=\beta(1-\hat{I}_1)-(\alpha_2+\delta)+\Phi(\alpha_1,\alpha_2)\hat{I}_1, \tag{27}$$

where $\hat{I}_1=1-\frac{\alpha_1+\delta}{\beta}$.

To compare our results with [37] we shall now consider the "jump" case when

$$\phi(\alpha_1,\alpha_2)=\begin{cases}1, & \alpha_2>\alpha_1,\\ 0, & \text{otherwise.}\end{cases} \tag{28}$$

Since according to this particular superinfection function, a trait α_2 does not "feel" the presence of a less virulent resident α_1, every trait α_2 that is larger than α_1 is able to invade the resident. On the other hand, if $\alpha_2<\alpha_1$, then α_2 can invade if

$$\alpha_2<2\alpha_1+\delta-\beta.$$

These observations can also be read off from the PIP in Fig. 10 (note that, since $\mathscr{R}_0=\frac{\beta}{\alpha+\delta}$, relevant traits are restricted to the interval $0\le\alpha\le\beta-\delta$, where we, of course, assume that $\beta-\delta>0$).

This provides an example of *evolutionary suicide*: by adaptive dynamics, the trait evolves towards a value such that the population can not exist. Assumptions concerning the distribution of mutations are now crucially important. If mutants are uniformly

distributed over the interval $[0, \beta - \delta]$, a balance may arise between the adaptive shift towards the "deadly" (for the parasite) virulence $\beta - \delta$ and the inflow of mutants at lower virulence levels. As Nowak and May showed in [37], this balance may result in the coexistence of a continuum of traits.

Appendix B: A caricatural example of non-smooth branching

Following Pugliese [40] we now consider (26) with $\Phi(\alpha_1, \alpha_2) = \beta\phi(\alpha_2 - \alpha_1) - \beta\phi(\alpha_1 - \alpha_2)$ and assume that (i) $\phi \geq 0$, (ii) $\phi(x) = 0$ for $x \leq 0$ and (iii) ϕ is differentiable twice in zero from the right, with the right derivatives $\phi'(0) > 0$ and $\phi''(0)$.

In this "mechanistic" case, the invasion exponent is given by

$$r_{\alpha_1}(\alpha_2) = \begin{cases} \alpha_1 - \alpha_2 + (\beta - \alpha_1 - \delta)\phi(\alpha_2 - \alpha_1), & \alpha_2 > \alpha_1 \\ \alpha_1 - \alpha_2 - (\beta - \alpha_1 - \delta)\phi(\alpha_1 - \alpha_2), & \alpha_2 < \alpha_1 \end{cases} \tag{29}$$

and the selection gradient by

$$\frac{\partial r}{\partial \alpha_2}\Big|_{\alpha_2=\alpha_1} = -1 + (\beta - \alpha_1 - \delta)\phi'(0). \tag{30}$$

For $\phi'(0) > 0$, there exists a singular point α^*,

$$\alpha^* = \beta - \delta - \frac{1}{\phi'(0)}. \tag{31}$$

Let us now investigate whether the equation

$$r_{\alpha_1}(\alpha_2) = 0 \tag{32}$$

has solutions that, when viewed as curves in the $(\alpha_1, \alpha_2)-$ plane, emanate from the singular point on the diagonal. We define

$$\xi(x) = \begin{cases} \dfrac{\phi(x)}{x}, & x > 0 \\ \phi'(0), & x = 0. \end{cases} \tag{33}$$

Note that ξ is locally monotone whenever $\phi''(0) \neq 0$. When ξ is invertible we can, with r defined by (29), solve (32) to obtain

$$\alpha_2 = \begin{cases} \alpha_1 + \xi^{-1}\left(\dfrac{1}{\beta - \alpha_1 - \delta}\right), & \alpha_2 > \alpha_1, \\ \alpha_1 - \xi^{-1}\left(\dfrac{1}{\beta - \alpha_1 - \delta}\right), & \alpha_2 < \alpha_1. \end{cases} \tag{34}$$

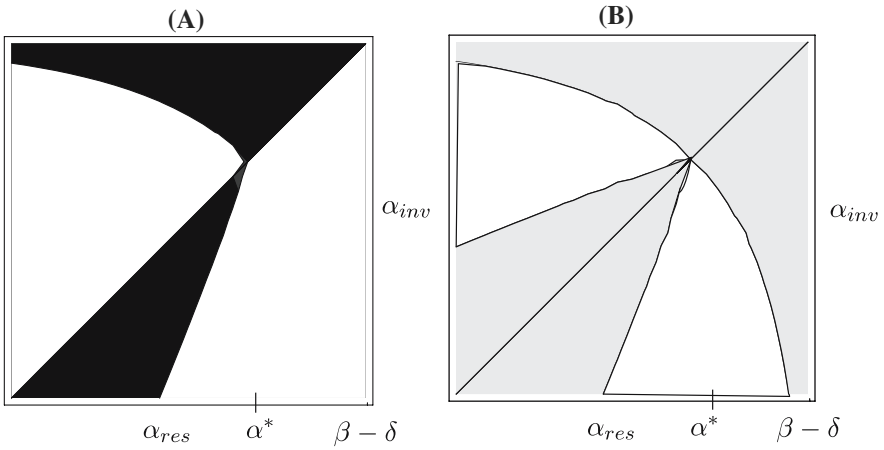

Fig. 11 **a** PIP constructed with the superinfection function $\phi(x) = 1 - \mathrm{e}^{-\theta x}$ for $x \geq 0$ and $\phi(x) = 0$ for $x < 0$. **b** *White areas* correspond to the regions in $(\alpha_{\mathrm{res}}, \alpha_{\mathrm{inv}})$–plane in which α_{res} and α_{inv} are mutually invadable. *Grey regions* are the areas where $r_{\alpha_{\mathrm{res}}}(\alpha_{\mathrm{inv}})$ and $r_{\alpha_{\mathrm{inv}}}(\alpha_{\mathrm{res}})$ have opposite signs

The expression (34) indeed defines a curve through the point (α^*, α^*). In this point, the curve has a corner,

$$\left.\frac{d\alpha_2}{d\alpha_1}\right|_{\alpha_1=\alpha^*} = \begin{cases} 1 + 2\dfrac{\phi'(0)^2}{\phi''(0)}, & \alpha_2 > \alpha_1, \\ 1 - 2\dfrac{\phi'(0)^2}{\phi''(0)}, & \alpha_2 < \alpha_1. \end{cases} \tag{35}$$

Under the reflection across the main diagonal, a straight line with slope $1+c$ transforms into a straight line with slope $(1+c)^{-1}$. For $c \neq 0$ the line with slope $(1+c)^{-1}$ does not coincide with the line with slope $1-c$. Elementary geometrical considerations reveal that, when the selection gradient changes from positive to negative when increasing the trait through the singular value (see (30)), there necessarily exists a region of mutual invasibility near the singular point (see Figs. 11, 12).

We find that the dimorphisms with $\alpha_2 > \alpha_1$ are explicitly given by

$$\begin{pmatrix} I_1 \\ I_2 \end{pmatrix} = \frac{1}{\beta\phi(\alpha_2 - \alpha_1)} \begin{pmatrix} \frac{1}{\phi(\alpha_2-\alpha_1)} - (\beta - \alpha_2 - \delta) \\ \frac{-1}{\phi(\alpha_2-\alpha_1)} + (\beta - \alpha_1 - \delta) \end{pmatrix} \tag{36}$$

from which it follows right away that mutual invasibility is both a sufficient and a necessary condition for coexistence. But will these dimorphisms be converging or diverging?

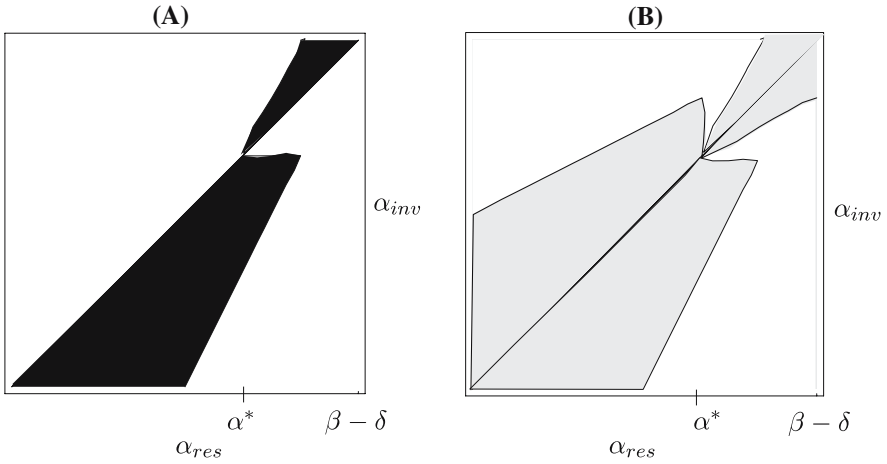

Fig. 12 **a** PIP constructed with the superinfection function defined in (38). **b** *White areas* correspond to the regions in $(\alpha_{\text{res}}, \alpha_{\text{inv}})$-plane in which α_{res} and α_{inv} and mutually invadable. *Grey regions* are the areas where $r_{\alpha_{\text{res}}}(\alpha_{\text{inv}})$ and $r_{\alpha_{\text{inv}}}(\alpha_{\text{res}})$ have opposite signs

From (27) it follows that

$$\frac{\partial^2 r}{\partial \alpha_2^2}\Big|_{\alpha_2=\alpha_1=\alpha^*} = \begin{cases} \dfrac{\phi''(0)}{\phi'(0)}, & \alpha_2 > \alpha_1, \\ -\dfrac{\phi''(0)}{\phi'(0)}, & \alpha_2 < \alpha_1, \end{cases} \tag{37}$$

so the invasion exponent changes sign in the singular point, exactly as in Fig. 8c. By continuity the invasion exponent $r_{\alpha_1,\alpha_2}(\alpha_3)$, the analogue of (16), must have, as a function of α_3, an odd number of zeros. Since $r_{\alpha_1,\alpha_2}(\alpha_1) = r_{\alpha_1,\alpha_2}(\alpha_2) = 0$, the generic number of zeros will be three and the graph is expected to look as the one in Fig. 9a or its mirror image. So the behaviour depicted in Fig. 9, in particular the relative immobility of one of the two traits, is a generic phenomenon.

Whether the upper or the lower of the two branches is the one that "moves", is determined by the sign of $\phi''(0)$. For the function ϕ, defined as $\phi(x) = 1 - \mathrm{e}^{-\theta x}$ for $x \geq 0$ and with $\theta > 0$, for instance, we have $\phi''(0) = -\theta^2 < 0$ and it will be the lower branch that will move (cf. Fig. 11).

For the function

$$\phi(x) = \begin{cases} x + \theta x^2, & 0 \leq x \leq \dfrac{\sqrt{1+4\theta}-1}{2\theta} \\ 1, & x > \dfrac{\sqrt{1+4\theta}-1}{2\theta}, \end{cases} \tag{38}$$

with $\theta > 0$, on the other hand, we have $\phi''(0) = 2\theta$ and it is the upper branch that will move (see Fig. 12).

In view of the evolutionary abyss at $\alpha = \beta - \delta$ (recall Appendix A), it is an intriguing question what will happen to the dimorphism in the long evolutionary run. We intend to take up this question in the near future. Note that letting $\theta \to \infty$ we converge to the "jump" case, but with the singular point being fixed at $\beta - \delta - 1$. Alternatively, one may parametrize ϕ so that $\phi'(0) \to \infty$ and then the singular trait moves to the evolutionary abyss. Pugliese [40] has observed that local features of the PIP are rather sensitive to details in ϕ while global features are more robust. He also notes that the exact shape of ϕ is probably beyond empirical investigation and that the model is, at best, a simplified description of a more complex process and concludes that a study of the exact dynamics near the singular point is mainy of mathematical interest. Based on the results presented in this paper we are slightly more optimistic: we hope that it will be possible to reveal at least some aspects of the complicated relationship between mechanisms and phenomena by deducing the superinfection function from a mechanistic within-host model, as in Sect. 5.

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