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ON THE EVOLUTION OF SEXUAL REPRODUCTION IN HOSTS COEVOLVING WITH MULTIPLE PARASITES

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Host–parasite coevolution has been studied extensively in the context of the evolution of sex. Although hosts typically coevolve with several parasites, most studies considered one-host/one-parasite interactions. Here, we study population-genetic models in which hosts interact with two parasites. We find that host/multiple-parasite models differ nontrivially from host/single-parasite models. Selection for sex resulting from interactions with a single parasite is often outweighed by detrimental effects due to the interaction between parasites if coinfection affects the host more severely than expected based on single infections, and/or if double infections are more common than expected based on single infections. The resulting selection against sex is caused by strong linkage-disequilibria of constant sign that arise between host loci interacting with different parasites. In contrast, if coinfection affects hosts less severely than expected and double infections are less common than expected, selection for sex due to interactions with individual parasites can now be reinforced by additional rapid linkage-disequilibrium oscillations with changing sign. Thus, our findings indicate that the presence of an additional parasite can strongly affect the evolution of sex in ways that cannot be predicted from single-parasite models, and that thus host/multiparasite models are an important extension of the Red Queen Hypothesis.

KEY WORDS: **Evolution of sex, high complementarity behavior, host–parasite, multiple infections, Red Queen.**

Sexual reproduction, here broadly defined as the production of offspring by recombining random sets of genes from the parents, remains one of the main conundrums of evolutionary biology. Although sexual reproduction is widespread in nature, its evolutionary benefit still remains debated. For a long time, the reasoning has been that it enhances genetic variability, and hence allows evolution to proceed faster than in a nonrecombining population. Although this argument is intuitive, it can be demonstrated that recombination does not always promote variability and variability in turn need not be beneficial (Otto and Lenormand 2002). Moreover, sex involves fitness costs, such as the twofold cost of sex in anisogamous species, implying that asexually reproducing organisms possess an immediate advantage over the sexually reproducing ones (Maynard Smith 1971; Lloyd 1980).

In response to these problems, a number of theories have been proposed in the past decades. One of the most prominent theories is the Red Queen Hypothesis (RQH) (Jaenike 1978; Bell 1982; Hamilton et al. 1990). It proposes that recombination may be advantageous when interacting species, such as hosts and their parasites, evolve in an evolutionary arms race. In such situations,

mixing of parental genes may provide an evolutionary advantage to the offspring due to the production of novel genotypes that escape the coevolving parasite species. In host–parasite systems, the probability of a host becoming infected by a certain parasite depends to a large extent on the genotype of the two species. Host–parasite systems are thought to have an inherent tendency to lead to coevolutionary dynamics characterized by fluctuating selection in both species (Jaenike 1978; Hamilton 1980; Nee 1989). A parasite genotype in a population of mostly susceptible host genotypes will quickly increase in frequency. After some time, novel genotypes in the host species may occur that are resistant, and thus disproportionately fit. This will lead to an increase of the novel host genotypes until the cycle begins anew, with the evolution of a parasite genotype able to attack the novel host genotype. Such dynamics may in principle continue for many generations, leading to a constant battle between the two species.

Population genetic models have shown that the coevolution between species may produce fluctuations in linkage disequilibria (LD), which in turn can cause fluctuations in epistasis by favoring different genotypes at different times. As a consequence of this process, the oscillating epistasis and LD can be of opposite signs resulting in a possible advantage to recombination (the so-called short-term effect, see e.g., Hamilton 1980; Barton 1995; Peters and Lively 1999). At the same time, recombination may also increase phenotypic variance in fitness and be selected for because it increases the selective response to directional selection (the so-called long-term effect). Which of these effects predominates in the RQH is currently debated (Peters and Lively 2007; Salathé et al. 2008a, 2009).

In recent years, the RQH has gained a substantial amount of support. Experimental studies have focused on the verification of the above assumptions in natural populations and tests of the RQH in general (Lively 1987; Dybdahl and Lively 1998; Carius et al. 2001; Fischer and Schmid-Hempel 2005), while theoretical approaches attempted to specify the precise conditions under which the RQH works. In particular, it has been argued that LD and epistasis should change sign every few generations for recombination to be beneficial (Barton 1995; Peters and Lively 1999; Gandon and Otto 2007). Salathé et al. (2008b) demonstrated that selection imposed by parasites on the host population need not be strong as long as the fitness costs for parasites that fail to infect their host are severe. Furthermore, the importance of the precise type of interactions has been the focus of attention of many studies (Agrawal and Lively 2002; Otto and Nuismer 2004; Kouyos et al. 2007), which showed that epistatic interactions between loci of coevolving species are essential for the RQH to work.

With few exceptions (Hamilton et al. 1990; Otto and Nuismer 2004; Kouyos et al. 2009), essentially the entire literature on the RQH focuses on the coevolution of host populations with a single parasite, and the role of coevolution with multiple parasites simultaneously has been largely ignored. Moreover, the studies that have addressed this problem reach contradictory conclusions about whether sexual reproduction is beneficial in multiparasite systems. The study by Hamilton et al. (1990) suggests that host/multiple-parasite systems tend to select for sex, although it does not allow to assess where the beneficial effect comes from because the role of individual factors cannot be disentangled. It is therefore unclear whether the observed selection for sex is a consequence of the interactions with additional parasite species per se or whether it is due to something else. Otto and Nuismer (2004) argue that in host/multiparasite models the change in frequency of a modifier allele is the sum of changes induced by each of the n independently acting parasites. However, the result is based on a number of assumptions (quasi-linkage equilibrium approximation assuming, for example, weak selection and epistasis), and it remains unclear whether their result can be extrapolated to the coevolutionary systems with epistatic interactions and strong selection. Finally, a recent study by Kouyos et al. (2009) shows that host/multiple-parasite systems can yield parameter areas of strong selection against sex caused by strong LD of constant sign. Importantly, the model proposed by Kouyos et al. considers only interactions between the two parasites and ignores all within-parasite interactions. However, according to some previous work on the RQH, such within interactions are likely, under certain conditions, to lead to selection for sex (Peters and Lively 2007; Salathé et al. 2008b) and it is not clear if/how such interactions could affect the result. In addition, all studies mentioned above have assumed that the entire host population is either simultaneously infected by both parasites or not infected at all, an assumption that in reality might not always be justified. As a result, the theoretical host/multiple-parasite literature offers contradictory predictions as to whether sexual reproduction is beneficial in such systems or not.

Nevertheless, the biological relevance of coevolution with multiple parasites is in our view indisputable (for reviews see Petney and Andrews 1998; Cox 2001; Woolhouse et al. 2002). Many studies can be found that document coinfection by many parasites being common in plants, animals, and humans (Baird 1951; Stirnadel and Ebert 1997; Petney and Andrews 1998; Sorensen and Minchella 1998; Brogden and Guthmiller 2002; Hood 2003). Moreover, hosts may be simultaneously coinfected by multiple strains of one parasite species (Read and Taylor 2001).

In this study, we aim to examine the impact of a host's exposure to two parasite species on the evolution of sexual reproduction in the host. To do this, we extend the standard population genetic host–parasite models by allowing hosts to interact with another parasite. In this way, we attempt to reconcile two apparently conflicting observations. On the one hand, if selection for/against sex by multiple parasites is expected to be additive under

certain conditions (Otto and Nuismer 2004), then, in a parameter range for which sex is beneficial in host/single-parasite systems (Salathé et al. 2008b) one might expect twice the selection for sex if two parasites are present. On the other hand, interactions between parasites might lead to other population-genetic effects that can disfavor increased recombination rates (Kouyos et al. 2009). To address this issue, we examine the impact of these factors on the evolution of sex across a wide range of parameter space, and dissect factors selecting for and against sex. We find that due to interactions between the two parasites, both on the genetic and the population level, the host/multiparasite models differ nontrivially from the single parasite models, and that the effect of coevolution with two parasites generally cannot be predicted solely on the basis of the single parasite effects.

The Model

GENERAL CONSTRUCTION

To investigate the role of sex in host/multiple-parasite systems, we extend the standard host–parasite models used in the literature (Otto and Nuismer 2004; Kouyos et al. 2007; Salathé et al. 2008b) to allow for two parasite populations interacting with the host population, such that the host can, with a given probability, be infected by either only one of the two parasites or simultaneously by both. For the sake of simplicity, we consider a deterministic model in which selection acts in the haploid phase. Hosts reproduce sexually with a random partner, whereas pathogens reproduce clonally. The model assumes that the host genome has five biallelic loci arranged in the following order: PPMQQ (variations of such order of loci are considered in Fig. S5). The two P loci determine the genetic interaction between the host and the parasite species p, and the two Q loci determine the genetic interaction between the host and the parasite species q. The locus M is a recombination modifier locus, which determines the host recombination rate and is selectively neutral. Assuming two modifier alleles m and M, there will be three possible combinations of these alleles during the diploid phase of reproduction, resulting in three recombination rates per genome: r_{MM}, r_{Mm}, and r_{mm}. In general we assume that

$$r_{MM} > r_{Mm} \geqslant r_{mm}. \tag{1}$$

To account for the fact that parasites typically have considerably shorter generation time than their hosts, we introduce a parameter n_{pg}, which denotes number of parasite generations per host generation. We also assume that a fraction f_p of the host population is infected with only parasite species p, a fraction f_q is infected with only parasite species q, and a fraction f_{pq} is infected with both parasite species p and q. We further denote the genotype frequencies for the host of genotype a by f_a^h, and frequencies of the parasite of species p or q and genotype b by $f_b^{p_i}$, where $p_i = \{p, q\}$. The host and each of the parasite populations is initiated in linkage equilibrium with random allele frequencies, except that all hosts have initially allele m, and thus recombine at the rate r_{mm}. After a burnin phase of 1000 host-generations, a modifier-phase begins and a mutant allele M is introduced randomly into 50% of the host population; coevolution continues for another 1000 host-generations after which the frequency of the M allele, f_M^{final}, is recorded. The increase (decrease) in this final frequency with respect to the initial frequency of 50% is used as the measure of selection for (against) sex. For all simulations shown, unless stated otherwise, we set the recombination rates $r_{MM} = 0.10$, and $r_{Mm} = r_{mm} = 0$. For this choice of recombination rates, there is no genetic exchange between the genomes carrying allele M and m. This parameter setting describes the competition between the sexual and asexual clones, or more precisely, the type of linkage modifier and the selected loci that occurs when sexual and asexual clones compete; see also Salathé et al. (2008a). (The results for higher recombination rates are shown in Fig. S2) Finally, we assume the mutation rate to be $\mu = 10^{-5}$ per locus per generation, the population size to be infinite, and the host–parasite generation ratio to be $n_{pg} = 2$ (unless stated otherwise).

Each reproduction cycle in the host consists of three steps: recombination $\mathcal{R}$, selection $\mathcal{S}^h$, and mutation $\mathcal{M}^h$. In the parasites, a reproduction cycle consists only of selection $\mathcal{S}^{Pi}$ and mutation $\mathcal{M}^{Pi}$ as we assume that parasites do not recombine. The recombination step assumes a single cross-over event at a random location, and the mutation step assumes that the probability of a forward mutation is the same as that of a backward mutation (for a more detailed description of these two steps see Kouyos et al. 2006). The selection step takes into account all interactions between the host and parasite populations. The effect of these interactions on the host is defined in three arrays: $w_{ij}^{h|p}$, which represents interactions with a single parasite p, $w_{ik}^{h|q}$, which represents interactions with a single parasite q, and $w_{ijk}^{h|pq}$, which represents interactions with both parasites p and q, respectively. The arrays give the fitness of the host of genotype i when attacked by genotype j of pathogen species p and/or by genotype k of pathogen species q. The fitness of the host genotype i is then

$$w_i^h = (1 - f_p - f_q + f_{pq}) + (f_p - f_{pq}) \sum_{j=1}^{4} w_{ij}^{h|p} f_j^p + (f_q - f_{pq}) \sum_{k=1}^{4} w_{ik}^{h|q} f_k^q + f_{pq} \sum_{j,k=1}^{4} w_{ijk}^{h|pq} f_j^p f_k^q. \tag{2}$$

The fitness effect of the host on each parasite population is taken into account by the use of the fitness interaction matrices $w_{ji}^{p_i}$, which specify the fitness value of the parasite $p_i = \{p, q\}$ of genotype j when infecting the host of genotype i. In analogy to the previous case, these arrays should be summed over the

frequencies of all the species that affect the calculated fitness. Thus, the fitness of the two parasite species is obtained by

$$w_j^{p_i} = \sum_{i=1}^{32} w_{ji}^{p_i} f_i^h. \tag{3}$$

Finally, the frequencies after selection are given by

$$\mathcal{S}^h\left[f_i^h\right] = \frac{w_i^h}{\bar{w}^h} f_i^h, \qquad \mathcal{S}^{p_i}\left[f_j^{p_i}\right] = \frac{w_j^{p_i}}{\bar{w}^{p_i}} f_j^{p_i}, \tag{4}$$

where the bar indicates the mean fitness, and again $p_i = \{p, q\}$.

The host and parasite populations are updated simultaneously. In particular, at every parasite generation, a fraction $f_p + f_q - f_{pq}$ of hosts and all parasites undergo viability selection. During that time, a fraction of $1/n_{pg}$ hosts reproduces (mutation and recombination) as well as all parasites (mutation only). Thus, our model captures the following essential features of host–parasite systems: (1) that parasites reduce the viability of their host, (2) that typically not all hosts in the population are infected by a parasite at all times ($f_p + f_q - f_{pq} < 1$), (3) that hosts often have overlapping reproductive generation times, and (4) that parasites typically have shorter generation times than their hosts ($n_{pg} > 1$). Note that the $n_{pg} = 1$ scenario corresponds to the reproductive scenario case considered by many authors previously, with the exception that we make the realistic assumption that not all hosts interact with a parasite. Mathematically speaking, the host and parasite frequencies from parasite generation t to $t + 1$ are updated as follows:

$$\begin{aligned} f_i^h(t+1) &= \mathcal{S}^h\left[\left(1 - \frac{1}{n_{pg}}\right) f_i^h(t) + \left(\frac{1}{n_{pg}}\right) \mathcal{R}\left[\mathcal{M}^h\left[f_i^h(t)\right]\right]\right] \\ f_j^{p_i}(t+1) &= \mathcal{S}^{Pi}\left[\mathcal{M}^{Pi}\left[f_j^{p_i}(t)\right]\right], \end{aligned} \tag{5}$$

where $\mathcal{R}, \mathcal{M}^h, \mathcal{S}^h$ denote the successive application of the recombination step, the mutation step, and the selection step in the host population, respectively. Likewise, $\mathcal{M}^{Pi}, \mathcal{S}^{Pi}$ denote the successive application of the mutation step and the selection step, respectively, in any parasite population.

To quantify selection on the modifier, the frequency of the M allele is measured at the end of the simulation (i.e., after 1000 host-generations of the burnin phase and 1000 host generations of the competition between the M and m allele, referred to as the modifier phase) for selection coefficients ranging from 0.01 to 0.99 with 0.01 gradation. As interaction between the host population and the two parasite populations is a function of four parameters $\{s_{P_1}, s_{P_2}, s_{H_1}, s_{H_2}\}$, we assume (unless stated otherwise) that both parasites have equal virulence, namely $s_{H_1} = s_{H_2} = s_H$, and are equally affected by the host, i.e., $s_{P_1} = s_{P_2} = s_P$. For simulations that do not assume symmetry between interactions with the two parasites please refer to Figure S4.

INTERACTIONS

A central aspect in describing host/multiple-parasite systems is the way in which the species interact with each other. One of the fundamental assumptions of the RQH is that fitness is affected by genetic combinations of the coevolving species. In particular, if a parasite species p_i manages to infect a host, it decreases the host fitness by s_{H_i}. On the other hand, if the parasite fails to infect the host, its fitness is decreased by the amount s_{P_i}.

In general, RQH models assume that the efficacy of infection is proportional to the degree by which the host and parasite genotypes are matched; the more the genotypes match, the more severe the infection is. In our model, the interactions between host and each parasite species are taken into account by the use of a generalized interaction model (GI, see Table 1). This model depends on the number of matched alleles between host and parasite, and a parameter ϵ_w, which determines the strength of epistasis between host loci on which the parasite acts. The nonepistatic model, also known as the multiplicative matching allele (MMA) model, is retrieved for $\epsilon_w = 0$. (Note that this model is nonepistatic only in the sense that there is no built-in epistasis, as the actual epistasis can arise through LD in the parasite populations.) The fully epistatic models, namely the matching allele (MA) model and the opposite of MA (OMA) are obtained for the maximal values of ϵ_w, namely $\epsilon_w = -0.5$ and $\epsilon_w = 0.5$, respectively. The fitness values corresponding to each interaction pattern are shown in Table 1.

Another fundamental aspect regarding the interactions in a multiparasite species model is the way in which the parasite species interact together. The simplest assumption is that the overall fitness cost on the host is the product of the fitness costs due to each parasite population individually. This implies that there are no epistatic interactions between the two parasite populations.

Table 1. Epistasis within parasite. The interactions within each parasite population of genome of length 2. The parameter $\epsilon_w \in [-0.5, +0.5]$ denotes the within-epistasis coefficient, and s_{H_i} and s_{P_i} are the virulence of *i*th parasite population, and selection imposed on the *i*th parasite population, respectively. The parasite fitness values are independent of each other.

Number of matched alleles		0	1	2
Host fitness	GI	1	$(1-s_{H_i})^{1/2+\epsilon_w}$	$1-s_{H_i}$
	MMA	1	$(1-s_{H_i})^{1/2}$	$1-s_{H_i}$
	MA	1	1	$1-s_{H_i}$
	OMA	1	$1-s_{H_i}$	$1-s_{H_i}$
Parasite fitness	GI	$1-s_{P_i}$	$(1-s_{P_i})^{1/2-\epsilon_w}$	1
	MMA	$1-s_{P_i}$	$(1-s_{P_i})^{1/2}$	1
	MA	$1-s_{P_i}$	$1-s_{P_i}$	1
	OMA	$1-s_{P_i}$	1	1

Table 2. Epistasis between parasites. The interactions between the two parasite populations and their effect on host's fitness. In the case of a single infection, the host fitness is decreased by the quantity s_{H_i}. In the case of coinfection, the multiplicative effect of the two parasite species is less ($\epsilon_b>0$) or more ($\epsilon_b<0$) detrimental for the host.

	No infection	Single infection	Coinfection
Host fitness (GI)	1	$1-s_{H_i}$	$[(1-s_{H_1})(1-s_{H_2})]^{1-\epsilon_b}$

If, on the other hand, coinfection by two parasite species is more or less detrimental for the host than expected on the effects of each parasite species individually, then these are deviations from the multiplicative scheme, and hence epistasis. We define here a between-parasite epistasis parameter ϵ_b, which denotes the strength of the deviation from multiplicativity of the parasite effects on the host. Table 2 describes the effect of coinfection based on the parameter ϵ_b.

Finally, we describe a third way of interaction in a multiparasite model: the fraction of hosts exposed to both parasite populations f_{pq} might be higher or lower than expected on the basis of products of single infection ratios, f_p and f_q. In other words, a double infection might be more likely than expected on the basis of individual parasite frequencies, i.e., $f_{pq} > f_p f_q$, or a double infection might be less likely than expected on the basis of individual parasite frequencies, i.e., $f_{pq} < f_p f_q$. For that purpose, we define a parameter δf_{pq} which describes the deviation from the expected rate of double infection such that

$$f_{pq} = f_p f_q + \delta f_{pq}. \tag{6}$$

Hence, $\delta f_{pq} > 0$ corresponds to an excess of double infections in the host population, and $\delta f_{pq} < 0$ corresponds to an underrepresentation of double infections in the host population. The condition of nonnegativity of infection rates yields the following constraint on δf_{pq}:

$$\begin{aligned} -\min[(1-f_p)(1-f_q), f_p f_q] &\leqslant \delta f_{pq} \\ &\leqslant \min[f_p(1-f_q), (1-f_p)f_q]. \end{aligned} \tag{7}$$

Thus, our model represents a generalized version of the standard host–parasite models in the literature. In particular, it generalizes the interaction between the two parasites considered in Kouyos et al. (2009), where host coevolving with the two parasites could never be infected only by one parasite (here obtained by setting $f_p = f_q = f,\ \delta f_{pq} = f - f^2$), but it also generalizes the multiparasite species case considered in Otto and Nuismer (2004), where the results were derived for the case of independence of the two parasites (here obtained by setting $f_p = f_q = 1,\ \delta f_{pq} = 0$).

HIGH COMPLEMENATARITY BEHAVIOR

Kouyos et al. (2009) observed that host–parasite models can, for very specific parameter settings (e.g., continuous time, no recombination), converge toward equilibrium points with strong LD that are either positive or negative, depending on the initial conditions. For a much broader range of parameters (e.g., nonvanishing recombination rates), these models demonstrate a qualitatively similar behavior. Specifically, they are characterized by potent LD of constant sign but instead of converging to an equilibrium point, they exhibit limit-cycle behavior.

Such behavior relates to a similar phenomenon in single-species population genetics: a number of studies found that certain diploid models can reach equilibrium states with strong LD, and these states have been termed high complementarity equilibria, or HCE for short (Bodmer and Felsenstein 1967; Franklin and Lewontin 1970; Feldman et al. 1974). However, the related limit cycle behavior around constant positive/negative values has been so far reported solely in the context of coevolutionary models (Kouyos et al. 2007, 2009), and has been termed high complementarity behavior (HCB).

The mechanism of selection for sex/recombination underlying the RQH is based on fluctuations of LD. In particular, it has been argued that one of the necessary conditions for the RQH to work is that LD change sign every few generations (Peters and Lively 1999; Gandon and Otto 2007). Because both phenomena, HCE and HCB, are characterized by strong LD of constant sign, it is natural to expect selection against sex/recombination in parameter regions that are characterized by HCE or HCB. This indeed has been shown in Kouyos et al. (2009).

To quantify the HCB in this study, we follow the value and sign of all six pairwise LD between the set {PPQQ} of loci involved in host–parasite interactions in the modifier-phase. If the LD does not change sign for at least 100 generations, we integrate its value in this period and divide by the length of the modifier-phase (second 1000 generations). For every subsequent occurrence of such a behavior the calculation is repeated. Mathematically speaking, if Δt_k denotes the duration time of the kth HCB phase in the modifier phase, and $\mathcal{D}_{ij}(t)$ denotes the measure of LD for the locus pair ij at time t, then HCB between the loci i and j is measured as follows:

$$\mathcal{D}^{ij}_{\text{abs}} = \frac{a}{1000}\left|\int_{t\in\Delta t_1}\mathcal{D}_{ij}(t)\,dt\right| + \frac{a}{1000}\left|\int_{t\in\Delta t_2}\mathcal{D}_{ij}(t)\,dt\right| + \ldots, \tag{8}$$

where a is the normalization constant chosen such that the highest observed $\mathcal{D}^{ij}_{\text{abs}}$ yields 0.25. The overall measure of HCB, $\mathcal{D}_{\text{abs}}$, is simply the sum of individual contributions

$$\mathcal{D}_{\text{abs}} = \sum_{ij\in\{\text{PPQQ}\}}\mathcal{D}^{ij}_{\text{abs}}. \tag{9}$$

Results

The aim of this study was to examine the evolution of sex when a host population is exposed to two parasite populations. Specifically, we wanted to dissect the selection for recombination in a multiple parasite model into (1) the part coming from the interaction of the host with each parasite individually and (2) the part coming from the simultaneous presence of the two parasite species, as implemented in equation (2). The factors selecting for sex in host/single-parasite models have been determined in detail in several studies (Peters and Lively 1999; Otto and Nuismer 2004; Kouyos et al. 2007; Salathé et al. 2008b). However, the effect of the simultaneous presence of multiple parasites has been studied less extensively, and depending on the result can range from simple additivity of effects (Otto and Nuismer 2004) to a strong selection against sex (Kouyos et al. 2009). We therefore divided the results into three subparts. In the first part, we examined whether the simultaneous presence of two parasite species in a host population selects against sexual reproduction in spite of the beneficial interactions with each parasite individually (via the MA model), and then surveyed the generality of the obtained results with respect to changes in the parameters. In the second part, we asked how strong the forces selecting against sex are (described and quantified in the first part) in the context of the full host/multiparasite model defined in the previous section, where the simultaneous presence of two parasite species occurs with a given probability. In the third part, we examined to what extent the evolution of sexual reproduction can be determined based solely on interactions with single parasite species, or to put it differently, how additive the effects of the two parasites are.

HCB AND SELECTION AGAINST SEX

To quantify the impact of the interaction between both parasites on the evolution of sex, we compared three special cases of the host/multiparasite model: (1) *Exclusive infection scenario* (EIS): The host is exposed to both parasite species, but can never be infected by two parasites at the same time; we assume that $f_{pq} = 0$ and $f_p = f_q = f_{\mathrm{EIS}}$. (2) *Independent infection scenario* (IIS): The host is exposed to both parasite species, but both infections occur independently of each other; we assume that $f_{pq} = f_p f_q$ and $f_p = f_q = f_{\mathrm{IIS}}$. (3) *Simultaneous infection scenario* (SIS): The host is exposed to both parasite species, but can be infected either by two parasites at the same time or none; we assume that $f_p = f_q = f_{pq} = f_{\mathrm{SIS}}$.

For each of these three scenarios, we plotted the final frequency of the M allele, f_M^{final}, as a function of the selection coefficients: s_H (parasite virulence), and s_P (selection imposed on the parasites by the host). To describe interactions within each parasite, we have chosen the MA model (i.e., $\epsilon_w = -0.5$ see Table 1), which has been shown to typically favor sexual over asexual reproduction (Kouyos et al. 2007; Salathé et al. 2008b). The infection rates are assumed to be $f_{\mathrm{EIS}} = f_{\mathrm{IIS}} = f_{\mathrm{SIS}} = 0.5$.

The results for all three scenarios are shown in Figure 1. Panel A shows the selection for/against sex in the case of the EIS scenario, panel B shows the selection for/against sex in the case of the IIS scenario, and panel C shows the selection for/against sex in the case of the SIS scenario. The legend in all three panels is the same: the red-to-white regions indicate increasing selection for sex ($f_M^{\mathrm{final}} > 0.5$), and the blue-to-black regions indicate increasing selection against sex ($f_M^{\mathrm{final}} \leqslant 0.5$). In particular, panel A shows that when the two parasites do not interact together in the sense that they never appear at the same time (EIS), the coevolution generally favors sex. Likewise, panel B shows that when there is no interaction between the two parasites in the sense that both infections are fully independent (IIS), the system favors sexual over asexual reproduction. In contrast, panel C shows that when the simultaneous infection occurs (SIS), parameter areas emerge with strong selection against higher recombination in the region of the parameter space that selected for increased recombination in models with exclusive infection and IISs (panels A and B). Remarkably, the region for which strong selection for sex occurred in EIS and IIS models (where the qualitative pattern is very similar to interactions with only one parasite—results not shown) changed into a region of the strongest selection against sex in the SIS scenario is shown in panel A.

Figure 1D shows examples of LD dynamics for three-parameter settings, highlighted in Figure 1C as points 1, 2, and 3. Each color corresponds to one of the six pairwise LD between the interacting loci of the host population. For the parameter setting 1 in panel D1 (low s_H and s_P), we see typical Red Queen dynamics with (almost all) LD oscillating between positive and negative values. For the parameter setting 2 in panel D2 (intermediate s_H and s_P), we observe that strong associations built up between some of the loci in the initial phase, which then were, for the most part, destroyed once recombination was introduced into the host population. Finally, for the parameter setting 3 in panel D3 (high s_H and s_P), we observe more stable types of dynamics with two of six LD converging to high, constant values. Such dynamics, that is, strong LDs of constant sign, have been shown to occur in single host–parasite models (Kouyos et al. 2007, 2009), and have been termed HCB in analogy to the qualitatively similar phenomenon of HCE (Bodmer and Felsenstein 1967; Franklin and Lewontin 1970). In the context of the RQH, any LD of constant sign, as HCE/HCB, is typically expected to lead to selection against sex due to a detrimental short-term effect. (More precisely, selection against sex is expected if the detrimental short-term effect is not compensated by a long-term effect that results from directional selection on allele frequencies.)

To assess how much of the selection against sex seen in Figure 1C can be explained by the HCB, we chose the following

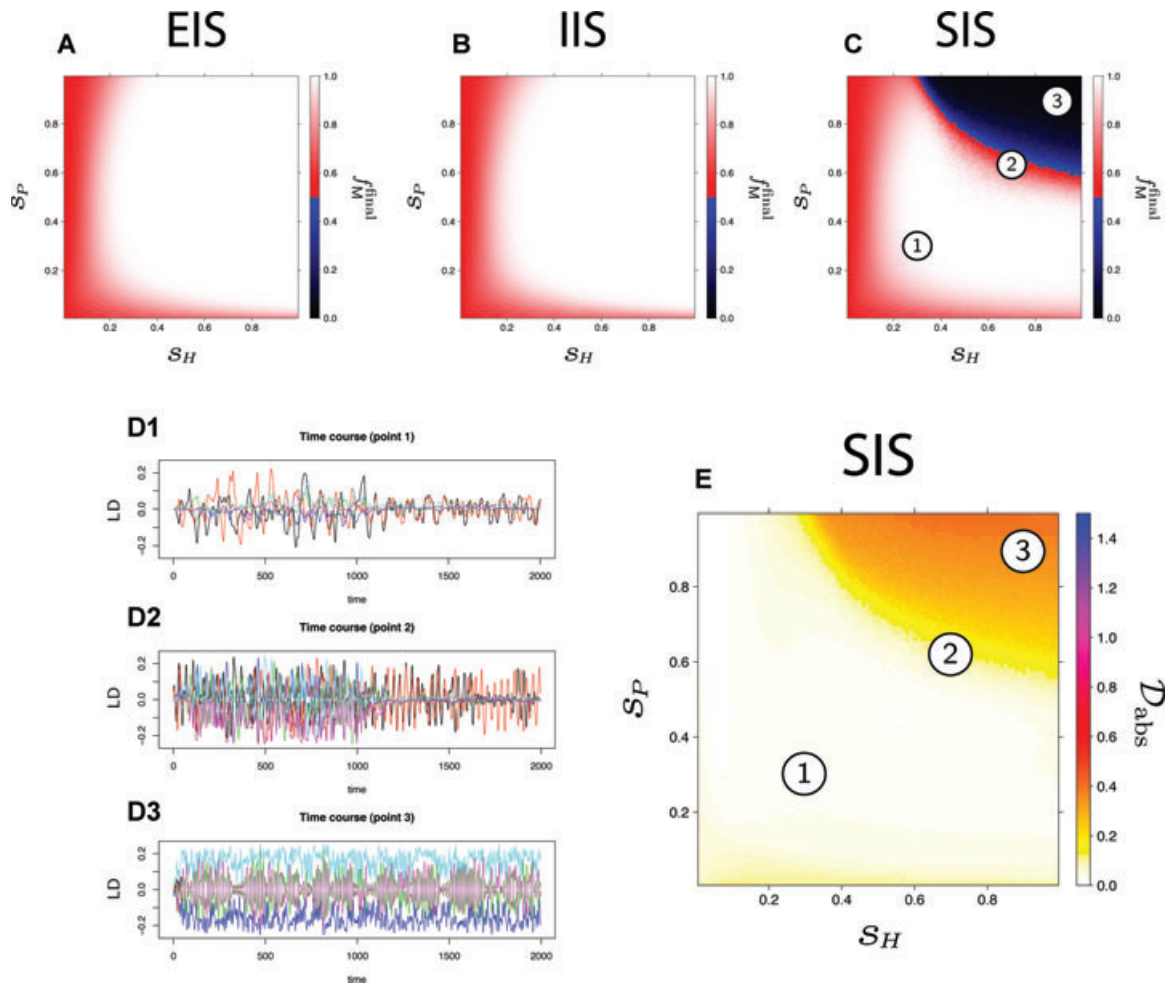

Figure 1. LD dynamics and the evolution of sex. The first three panels show f_M^{final} as a function of selection coefficients s_H and s_P, showing regions of selection for sex (red to white) and regions of selection against sex (blue to black). (A) The host is exposed to both parasites but never infected by both at the same time; the system generally selects for increased recombination rates. (B) The host is exposed to both parasites but the two infections are fully independent and the system also generally selects for increased recombination rates. As mentioned later, in this situation the effects of the two parasites are additive for any value of s_H and s_P. (C) The host is exposed to both parasites but when infected then with two parasites at the same time. In this case, areas of strong selection against sex emerge for high selection values. (D1) Time course of LD dynamics for parameter combination 1 in panel C, an example that results in selection for sex, shows domination of rapid LD oscillations around zero (within parasites) over the LDs that oscillate very weakly (between parasites). (D2) Time course of LD dynamics for parameter combination 2 in panel C, an example that also results in selection for sex, shows that associations between parasites build up initially, and then are destroyed once recombination is introduced. (D3) Time course of LD dynamics for parameter combination 3 in panel C, an example that results in selection against sex in a fraction of parameter space, shows the strong LDs of constant sign (HCB). (E) Strength of high complementarity equilibria $\mathcal{D}_{abs}$ for the same parameter setting as in panel C (see also main text). The color scale corresponds to the measure of the intensity of the HCB. Comparison of the black to blue region in panel C with regions of HCB in panel E ($\mathcal{D}_{abs} > 0.2$) indicates that regions with HCB result in strong selection against sex. The parameters used are $r_{MM} = 0.10$, $r_{Mm} = r_{mm} = 0$, $\epsilon_b = 0$ and $\epsilon_w = -0.5$, $f_{EIS} = f_{IIS} = f_{SIS} = 0.5$, and $n_{pg} = 2$. We assume $s_H = s_{H_1} = s_{H_2}$, $s_P = s_{P_1} = s_{P_2}$. Graphs in panel A, B, C, and E show the average results of 1000 simulation runs.

approach. First, we measured the average strength of HCB in the period between the 1000th and 2000th generation for each possible pair of interacting loci in the host. The overall measure $\mathcal{D}_{abs}$ is the sum over contributions from all such possible pairs (see the model section for details). The results can be seen in Figure 1E, where the intensity of color corresponds to the value of $\mathcal{D}_{abs}$. As an HCB contribution from a single pair of loci can reach maximally 0.25 in absolute value, and in the analyzed case only two of six pairwise LD produced HCB (always loci between the two parasites, see Fig. S1), the value of $\mathcal{D}_{abs}$ does not exceed 0.5. The comparison of Figure 1C and E shows that HCB (i.e., $\mathcal{D}_{abs} \gtrsim 0.2$) can be observed in those regions where $f_M^{final} < 0.5$.

Therefore, generally the entire parameter area where selection against sex is observed in Figure 1C can be attributed to HCB. In contrast, if the detrimental LD dynamics become weak enough (parameter settings 1 and 2), the same phenomenon that selected against sex in the parameter region of strong selection can now be outweighed by the beneficial, epistatic effects, leading to selection for sex/recombination.

Thus, we observed that if the host population can only be infected by two parasite species simultaneously or by none, then (1) an interaction between the two parasites leads to the emergence of parameter areas with strong selection against sex, and (2) the observed parameter areas with selection against sex are caused by HCB. We next examined the robustness of the good agreement between the areas of selection against sex and the areas with HCB with respect to changes in the interaction pattern between the host and the two parasite species, namely the changes in the epistasis-within ϵ_w and the epistasis-between ϵ_b. We chose the parameter range $-0.5 \leqslant \epsilon_w \leqslant 0$ (between the maximally epistatic MA model and the nonepistatic MMA model; results for $0 \leqslant \epsilon_w \leqslant 0.5$ were qualitatively similar) and $-1 \leqslant \epsilon_b \leqslant 0.5$; see the model section and Table 2 for a detailed description of these two parameters.

Figure 2 shows the influence of the two types of epistasis on the evolution of recombination rate. Specifically, Figure 2A shows the percentage of the total parameter space $0 < s_H, s_P < 1$ in which sex is selected against (i.e., $f_M^{\text{final}} < 0.5$) as a function of ϵ_b and ϵ_w (note that the case of $\epsilon_w = -0.5$ and $\epsilon_b = 0$ corresponds to the situation in Fig. 1A). Figure 2B shows an equivalent plot regarding the influence of HCB. In particular, it shows the percentage of the total parameter space $0 < s_H, s_P < 1$ where HCB is observed (here defined as $\mathcal{D}_{\text{abs}} > 0.2$). For parameter values close to the MMA model ($\epsilon_w \to 0$), we observe very strong selection against sex (Fig. 2A) and, accordingly, stronger impact of HCB in that region (Fig. 2B). Likewise, in the parameter areas where epistasis-between had an additional influence on the strength of HCB, as seen in Figure 2B, we see an increased percentage of the parameter space where selection against sex is observed. Thus, we generally observe a good agreement between the regions with strong HCB and the regions where selection against sex/recombination is observed. In particular, sex is selected against if HCB is strong. It needs to be emphasized, however, that while HCB always causes selection against sex, selection against sex need not be caused by HCB, and this is responsible for the slight discrepancies between Figure 2A and B. The emergence of parameter areas with selection against recombination not caused by HCB was extensively discussed in the study by Salathé et al. (2008b) and can be explicitly seen in runs with higher recombination rates, which we show in Figures S2 and S3.

We analyzed numerous variants of the SIS model, including for example different n_{pg} and f_{SIS} values (satisfying $f_{\text{SIS}} < 1$,

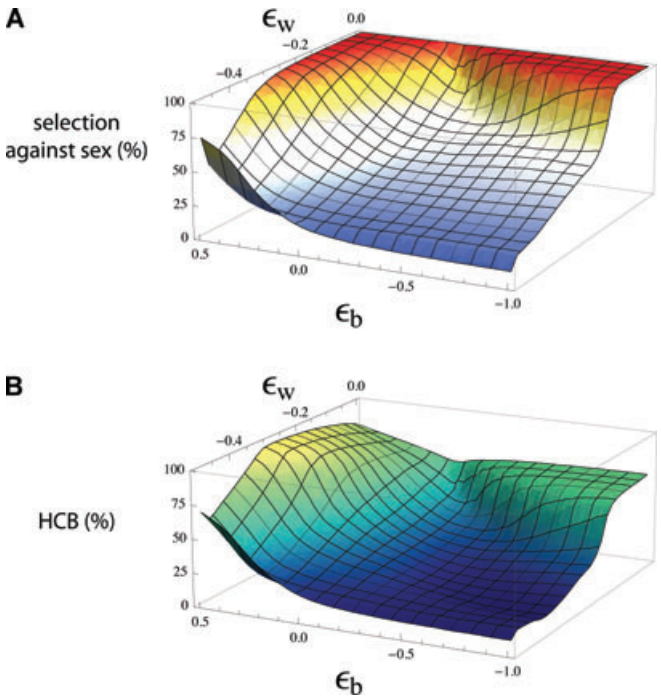

Figure 2. The role of epistasis. (A) The percentage of the s_H, s_P-parameter space where sex is selected against ($f_M \leqslant 0.5$), as a function of ϵ_b (epistasis between parasites) and ϵ_w (epistasis within parasites). (B) The percentage of the same parameter space where the high complementarity behavior (HCB) is observed, as a function of ϵ_b and ϵ_w. In the majority of parameter space, one observes a good agreement between the two regions, suggesting that the emerging HCB is a good determinant of selection against sex. In the remaining parameter areas, especially of small ϵ_w, there is less agreement between the two regions, which is due to the fact that additional regions of selection against sex, not caused by HCB, arise. The parameters used are $r_{MM} = 0.10$, $r_{Mm} = r_{mm} = 0$, $f_p = f_q = f_{pq} = 0.5$, $n_{pg} = 2$, and $s_H = s_{H_1} = s_{H_2}$, $s_P = s_{P_1} = s_{P_2}$. Note that the ϵ_b-axis has been reversed. Results are averaged over 10 simulations.

see the Supporting information for a detailed discussion), different recombination and mutation rates, finite population sizes, asymmetries between the two parasites, altered host genome architecture, inhibition/facilitation between the two parasites, and introduction of the modifier at low frequencies (see Supporting information for simulations of some of these modifications). Many of these alterations affected the HCB in a way that is predictable, for example, higher recombination rates, uneven parasite mutation rates, or a diminished role of one of the parasites ($s_{H_1} > s_{H_2}$ and $s_{P_1} > s_{P_2}$) reduce the strength of the HCB, while a faster parasite evolution (higher n_{pg} or larger parasite mutation rate) increases the strength of the HCB. On the other hand, some changes such as the order of the selected loci and the modifier locus affected the occurrence of HCB in a way that cannot be easily generalized. However, in all examined scenarios, we observed a systematic

accordance between parameter regions with strong selection against recombination and parameter regions with strong HCB, like between Figure 1C and E. Therefore, we conclude that the HCB is a very powerful determinant of selection against sex in the host/multiparasite models.

THE IMPACT OF HCB IN THE RED QUEEN MODELS

Having observed a rather substantial impact of HCB in models that assumed that hosts, when infected, are infected by both parasites at the same time, we next examined the role of such behavior in the context of the full host/multiparasite model defined in the model section, that is., when the simultaneous infection happens with a given probability f_{pq}. We assumed that $f_p = f_q = 0.5$, that f_{pq} is given by equation (6), and examined the dependence of the model on the two following parameters: ϵ_b (epistasis between the two parasites) and δf_{pq} (deviation from the expected coinfection rate). The remaining parameters were identical to the ones used in Figures 1 and 2.

The results are shown in Figure 3. Because for the recombination rates used here HCB, as measured in Figures 1 and 2, proved a good measure of selection against sex, we quantified the behavior of the host/multiparasite model solely based on the percentage of parameter space $0 < s_H, s_P < 1$ where HCB is observed. One can see that HCB becomes significant in the parameter region where $\delta f_{pq} > 0$ and $\epsilon_b > 0$, but it also occurs for $\delta f_{pq} > 0$ and $\epsilon_b < 0$ although to a lesser extent. On the other hand, HCB weakens significantly when $\delta f_{pq} < 0$, and as a result we observe little or no selection against sex. (In the Supporting information, we provide an intuitive explanation for why it is so; see also the discussion.)

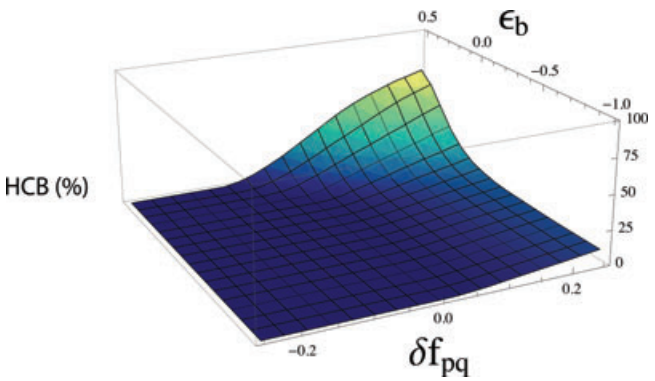

Figure 3. The impact of HCB in the host/multiparasite RQ model. The percentage of the s_H, s_P-parameter space where the high complementarity behavior (HCB) is observed, as a function of δf_{pq} and ϵ_b. One observes the most extensive HCB, and hence the largest areas with selection against sex, in the area of $\delta f_{pq} > 0$ (excess of double infections) and $\epsilon_b > 0$ (positive epistasis between parasites). The parameters used are $r_{MM} = 0.10$, $r_{Mm} = r_{mm} = 0$, $f_p = f_q = 0.5$, $n_{pg} = 2$, and $s_H = s_{H_1} = s_{H_2}$, $s_P = s_{P_1} = s_{P_2}$. Results are averaged over 10 simulations.

In short, we observed that sexual reproduction in the host/multiparasite model is the least favored when there is (1) strong positive epistasis between the two parasites affecting the host fitness ($\epsilon_b > 0$) and (2) an excess of double infections in the population ($\delta f_{pq} > 0$); this result is generally robust to variations in parameters. Moreover, the occurrence of HCB is typically higher in the region $\delta f_{pq} > 0, \epsilon_b < 0$ than in the region $\delta f_{pq} < 0, \epsilon_b > 0$ (however, we saw some exceptions to this, especially for high f_p and f_q; see Fig. S7). We also observed less HCB when the effects of the two parasites are asymmetric, namely when infection by parasite p is more virulent than the infection by parasite q (see Fig. S4), and when the infection by parasite p is more likely than the infection by parasite q (see Fig. S8).

HOW ADDITIVE ARE THE EFFECTS OF INDIVIDUAL PARASITES?

Otto and Nuismer (2004) showed that, within the limitations of the quasi-linkage equilibrium approximation, the total selection on recombination modifier is the sum of the effects of each individual parasite. In contrast, our analysis shows that in spite of highly beneficial effects stemming from the interactions with individual parasites, strong detrimental effects on the modifier can emerge as a result of interactions between the two pathogens. The HCB responsible for this effect can overwhelm any other advantageous effects, and eventually cause strong selection against sex in a considerable region of the parameter space. As this result implies that the effects of individual parasites are not additive in at least part of the parameter space, we investigated more thoroughly departure from additivity in the selection for sexual reproduction.

To this end, we calculated the effect of coevolution (selection) on the recombination modifier in a host population exposed to both parasites, s_M^{pq} and the effect of coevolution on the recombination modifier in a host population exposed to solely parasite p plus the effect solely due to parasite q, $s_M^{p+q} = s_M^p + s_M^q$. We were interested in two measures of additivity in the context of our model: (1) the relative deviation from additivity: $D_{\text{val}} = |s_M^{p+q} - s_M^{pq}|/|s_M^{pq}|$, and (2) the direction of deviation from additivity: $D_{\text{dir}} = \text{sgn}(s_M^{pq} - s_M^{p+q})$. D_{val} thus quantifies the deviation from additivity in the system, and D_{dir} measures whether exposure to two parasite species led to more selection against sex ($D_{\text{dir}} > 0$) or to less selection against sex ($D_{\text{dir}} < 0$) than expected on the basis of individual effects.

The results are shown in Figure 4. Panel A shows the percentage of the parameter space $0 < s_P, s_H < 1$ in which deviation from additivity was observed ($D_{\text{val}} > 0.1$) as a function of δf_{pq} and ϵ_b. In contrast, panel B shows the percentage of the parameter space in which there is less sex than expected on the basis of individual

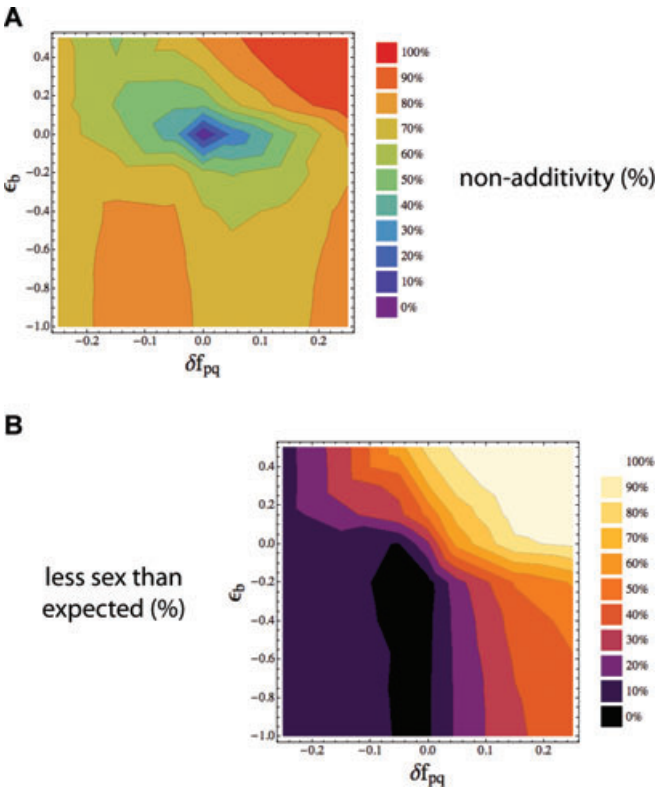

Figure 4. Nonadditivity of parasite effects. (A) The percentage of the s_H, s_P-parameter space where the effects of the two parasites deviate from additivity, as a function of δf_{pq} and ϵ_b. Various colors correspond to different extents of nonadditivity (defined as $D_{val} < 0.1$, see main text), ranging from violet (additive for all selection values) to red (nonadditive for all selection values). As the model shows, the host/multiparasite systems are the most additive for independent infections ($\delta f_{pq} = \epsilon_b = 0$), and the area of deviation from additivity increases once the two parasites become nonindependent vanishes. (B) The direction of deviation from additivity, and more precisely, the percentage of the s_H, s_P-parameter space where the two parasites select for sex less than expected on the basis of the individual effects. The colors range from white (less sex than expected for all selection values) to black (more sex than expected for all selection values). The parameters used are $r_{MM} = 0.10$, $r_{Mm} = 0$, $r_{mm} = 0$, $\epsilon_w = -0.5$, $f_p = f_q = 0.5$, $n_{pg} = 2$, and $s_H = s_{H_1} = s_{H_2}$, $s_P = s_{P_1} = s_{P_2}$. Results are averaged over 200 simulations.

parasite effects ($D_{\text{dir}} = -1$) as a function of δf_{pq} and ϵ_b. One can see that the effects of the two parasites show the least deviation from additivity when the two infections are fully independent ($\delta f_{pq} = \epsilon_b = 0$). However, as soon as epistasis-between or excess/underrepresentation of double infections become nonzero, the host/multiparasite system departs from additivity and selects for/against sex more or less than expected on the basis of single parasite effects. In particular, we observed the strongest departure from additivity in the region of positive ϵ_b and δf_{pq}, which is also the parameter region where HCB is relatively common (cf. Fig. 3). There we also see that the exposure to two parasites led to less sex than expected. In the region where little or no HCB was observed ($\delta f_{pq} < 0$), one can also see a deviation from additivity of parasite effects. In this case, however, the exposure to two parasites led to more sex than expected. This is, in large part, also true for the region of $\delta f_{pq} > 0$, $\epsilon_b < 0$, where we saw a mixture of the two kinds of deviations: a positive deviation from additivity (more sex than expected) in the region with HCB, and a negative deviation from additivity (less sex than expected) in the region with low s_H (results not shown). The corresponding results for higher recombination rates are shown in Figure S9.

That the host/multiparasite models are the most additive in the region of $\delta f_{pq} = \epsilon_b = 0$ (full independence) is intuitive as in this case host does not experience any epistasis (see Supporting information). What is more surprising is that, under the assumption of independence of parasites, our results showed that the additivity of the parasite effects on the modifier holds for any value of s_P and s_H (not shown). Interestingly, this result not only supports the intuition of the study by Otto and Nuismer (2004), which was derived on the basis of the quasi-linkage equilibrium approximation, but also shows that, in some special cases, the additivity can extend to the regions that are thought to violate the quasi-linkage equilibrium approximation.

Discussion

In this study, we have extended previous RQH models (Peters and Lively 1999; Otto and Nuismer 2004; Kouyos et al. 2007; Salathé et al. 2008b) to investigate the interactions of multiple parasite species with one host species, and the effect of these interactions on the evolution of sex/recombination. Generally speaking, we have shown that the host/multiparasite models differ nontrivially from the models that consider interactions with a single parasite. The reason for this is that the nonindependency of the two infecting parasites, either in terms of epistatic interactions between the two parasites or in terms of infection probabilities, can very often drastically affect the outcome of the model expected on the basis of a single-parasite model. The appearance of the second parasite in a host population can therefore lead to significantly more or less sex than expected on the basis of the individual parasite effects.

Although host/multiparasite interactions have been a subject of study by a few authors previously (Hamilton et al. 1990; Otto and Nuismer 2004; Kouyos et al. 2009), our treatment of the host/multiparasite coevolution goes beyond previous work in that it dissects the selection for/against recombination into the part coming from the interaction of the host with each parasite individually, and the part coming from the simultaneous presence of the two parasite species. This allows for an explicit analysis of the factors selecting for and against sex in such a system.

Moreover, our model allows for parasitic coinfection at a given rate (probability), which reflects the ecological aspect of host–parasite coevolution that not all hosts exposed to more than one parasite population will be infected by more than one parasite (or even be infected at all).

In accordance with the study by Kouyos et al. (2009), we observed the emergence of HCB as a result of an interaction between the two parasites. There, the HCB, which is identified with strong LD of constant sign, had been shown to select against sex in a two-locus host/multiparasite model. Here we found that, in spite of strongly beneficial interactions with each parasite individually (through the MA model), the simultaneous presence of two parasite species can similarly lead to the emergence of parameter regions with strong selection against sex. This selection against sex is caused by the same force—strong LDs of constant sign (HCB). This phenomenon can be best seen when one assumes that a fraction of hosts remains uninfected while the rest of the host population is simultaneously coinfected by two parasites. In that case, HCB generally emerges in the parameter regions in which selection values are the highest (especially with high parasite virulence s_{H_1}, s_{H_2}). These regions (characterized by strong selection for sex in models with exclusive and independent infections, and hence no HCB; see Fig. 1A, B) exhibited a strong selection against sex in models with the simultaneous presence of the two parasites (Fig. 1C). However, in the parameter regions with low parasite virulence the effect of the detrimental LD dynamics became weaker and was generally compensated by the positive effects coming from the interactions within each of the parasites; selection for sex occurred quite frequently under these conditions. Finally, we have seen that HCB is a good determinant of selection against sex.

In reality, however, not all hosts in a population are infected, and parasitic infections in the population can be assumed to occur with a certain probability. Therefore, we examined the impact of HCB on the evolution of recombination in the host when infected by only one of the two parasites, coinfected by both parasites, or remains uninfected, all with a given probability. For that purpose we introduced rates of infection, as defined in equations (2) and (6), and assumed again that interaction with a single parasite is highly beneficial for sexual reproduction. The analysis showed that also under these assumptions we could still observe substantial parameter areas with HCB, and hence selection against sex. In particular, the HCB turned out to be the strongest when two conditions were met: (1) the parasitic coinfection was more severe than expected on the basis of individual virulence measures, and (2) the parasitic coinfection was more likely to happen than expected on the basis of the single infection rates. Notably, the HCB was always absent when the coinfection was less severe and less common than expected (see Fig. 3 as well as Figs. S7 and S8).

To understand why HCB is most likely to arise for positive epistatic interactions between the two parasites, one has to understand how HCB arises in general. This question is difficult to address even in the context of a simpler, two locus, biallelic host-parasite model. Therefore, we only provide an intuitive argument for why HCB emerges in host–parasite systems. The argument is described in detail in Supporting information of this study, and, in a nutshell, it shows how HCB could be explained by a positive feedback loop between the LD in the host and the correlation between the allele frequencies in the two parasites. (e.g., a positive correlation between the parasite allele frequencies generates positive epistasis, which, through positive LD, leads again to a positive allele correlation; hence a positive HCB.) Importantly, however, for such a feedback to occur, a specific set of conditions is required. In particular, the feedback always occurs if $\delta f_{pq} > 0$ and $\epsilon_b > 0$. Therefore, our results show that positive epistatic interactions between the two parasites usually generate HCB, and HCB in turn produces a detrimental effect on the evolution of sexual reproduction during the host/multiparasite coevolution.

The fact that the emergence of an additional parasite species can lead to the appearance of strongly disadvantageous effects for sexual reproduction speaks against an intuitive null-model that the individual parasite effects should add up with each additional parasite acting on the host, an intuition supported by the analytical approximations in Otto and Nuismer (2004). Indeed, in this study we have shown that such additivity is characteristic of only a small fraction of the parameter space (full independence of the two parasites). In the regions where HCB was present (positive epistatic interactions and excess of double infections), the coevolution with two parasites led to less sex than expected on the basis of individual parasite effects. Interestingly, the nonadditivity was also characteristic of regions where HCB was absent (negative epistatic interactions and underrepresentation of double infections). Surprisingly, there the coevolution with two parasites led to more sex than expected on the basis of individual parasite effects. A closer examination showed that the effects responsible for this were additional rapid LD oscillations around zero between loci involved in interactions between the two parasites (results not shown). As a complete independence of parasitic infections, however, is quite rare due to many known interactions between the parasites (Cox 2001; Woolhouse et al. 2002; Graham et al. 2007), we show that the presence of another parasite is vital to the evolution of sex.

The presented study thus points to an important role of coevolution with more than one parasite in the evolution of host's sexual reproduction (but also in a more general context), an aspect which, with few exceptions, has been largely ignored in the host–parasite theoretical literature. Our study shows that the interactive dynamics, resulting from the presence of an additional parasite, can be crucial to the fate of the recombination modifier

and thus can decide whether the system selects for or against sexual reproduction. In some cases (e.g., for positive epistasis between the parasites and excess of double infections) interactions between the parasites can result in strong LD states of constant sign that are detrimental for sex (HCB), whereas in other cases (e.g., for negative epistasis between the parasites and deficiency of double infections) it can give rise to additional rapid LD oscillations around zero that favor genetic shuffling. Our results qualify those derived by Kouyos et al. (2009), who suggested that the host/multiparasite models are less likely to explain the evolution of sex than the single-parasite models. Here, we have shown that this result strictly depends on the parameter range one examines. Moreover, our study confirms and extends the insight provided by Otto and Nuismer (2004), where the authors have suggested that the effects of the independent parasites should add up in the quasi-linkage equilibrium parameter regime. Here, we have shown that additivity can also occur in areas that are thought to violate such regime (e.g., strong selection), and that it breaks down almost immediately as soon as the independence assumption is violated.

In summary, we provide two important reasons for why inclusion of more than one parasite species is fundamental for understanding the role of sex in host–parasite systems: (1) an interaction between the simultaneously infecting parasite species could lead to strong detrimental effects that select against increased recombination, and (2) the host–parasite systems with more than one parasite are likely to behave differently from what one would expect based on effects due to a single parasite. Taken together with the empirical observations that hosts coevolving with multiple parasite species seem to be the rule rather than the exception (Petney and Andrews 1998; Cox 2001; Woolhouse et al. 2002) suggests that studying interactions of multiple parasite species with their hosts is crucially important. Thus, to advance our understanding of the coevolution of a host population in an environment of multiple parasite species, as well as understanding the role and importance of HCB in real populations, there is a clear need for further theoretical investigations alongside experimental studies to enable narrowing down the relevant areas of the parameter space.

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Supporting Information

The following supporting information is available for this article:

Figure S1. The contribution of individual pairwise linkage disequilibria in the production of the high complementarity behavior.
Figure S2. Selection of sex/recombination versus high complementarity behavior for higher recombination rates.
Figure S3. The impact of epistasis-within and epistasis-between on the evolution of sex/recombination for intermediate and high recombination rates.
Figure S4. Selection of sex/recombination and asymmetric effects of the two parasites.
Figure S5. Selection of sex/recombination and the alternative order of loci.
Figure S6. Selection of sex/recombination for various infection ratios.
Figure S7. The extent of the high complementarity behavior for various single-infection rates.
Figure S8. The extent of the high complementarity behavior for asymmetric single-infection rates.
Figure S9. Nonadditivity of parasite effects for higher recombination rates.

Supporting Information may be found in the online version of this article.

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