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The maintenance of sex in parasites

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The maintenance of sex is an unresolved paradox in evolutionary biology, given the inherent twofold fitness advantage for asexuals. Parasitic helminths offer a unique opportunity to address this enigma. Parasites that can create novel antigenic strains are able to escape pre-existing host immunity. Viruses produce diversity through mutation with rapid clonal proliferation. The long generation times of helminth parasites prevent them from adopting this strategy. Instead, we argue that sexual reproduction enables parasitic helminths to rapidly generate strain diversity. We use both a stochastic, individual-based model and a simple analytical model to assess the selective value of sexual versus asexual reproduction in helminth parasites. We demonstrate that sexual reproduction can more easily produce and maintain strain diversity than asexual reproduction for long-lived parasites. We also show that sexual parasite populations are resistant to invasion by rare asexual mutants. These results are robust to high levels of cross-immunity between strains. We suggest that the enhancement of strain diversity, despite stochastic extinction of strains, may be critical to the evolutionary success of sex in long-lived parasites.

Keywords: epidemiology; extinction; genetic diversity; helminth parasites; sex; stochastic model

1. INTRODUCTION

The enigma of sexual reproduction arises from its widespread occurrence despite substantial fitness costs (Maynard Smith 1971). In particular, females forfeit half of their genome to that of their mate in the production of offspring, generating a twofold cost for sexual reproduction relative to asexual reproduction (Williams 1975), everything else being equal. Previous models have suggested that intense negative frequency-dependent selection can favour sex (Bell & Smith 1987; Hamilton *et al.* 1990). Such dynamics can arise from host–parasite coevolution (Nee 1989). However, previous work has tended to focus exclusively on the evolution of sex in hosts challenged by parasites (Jaenike 1978; Hamilton 1980; Bell 1982). These models have assumed that parasites are asexual, haploid and have short generation times. By comparison, there has been much less consideration of the sexual reproduction of helminth parasites (but see Gemmill *et al.* 1997; Lythgoe 2000; Galvani *et al.* 2001). To develop a broadly applicable theory for the evolution of sex, it is necessary to consider the selective value of sex from the perspective of the parasite.

We use a stochastic, individual-based simulation model to compare asexual versus sexual reproduction of long-lived, diploid parasites in a host population that develops protective strain-specific immunity, for which there is considerable evidence (Nadler 1987; Wassom *et al.* 1988; Dobson & Jian-Ming 1991; Fraser & Kennedy 1991; Goyal & Wakelin 1993; Zvelebil *et al.* 1993; Bellaby *et al.* 1995; Tang *et al.* 1995; Currie *et al.* 1998; reviewed by Galvani & Gupta 1998). We also allow varying levels of cross-immunity between antigenically related strains (Hackett *et al.* 1987; Quinnell *et al.* 1991). Our approach integrates population genetics with realistic epidemiological dynamics (May & Anderson 1983; Galvani 2003). The advantage for sex in many early models required that a given parasite genotype could infect only one specific host genotype, that is, 'gene-for-gene' host–parasite interactions (Hamilton 1980). However, empirical data indicate that parasites tend to be much more broadly infective (reviewed by Woolhouse & Webster 2000). Our model reveals an advantage for sex in parasites without gene-for-gene matching. In addition, ecological interactions and infection dynamics are stochastic processes involving discrete individuals. Therefore, stochastic individual-based models, such as the one we employ here, are well suited for realistically modelling epidemiological and ecological processes (Levin & Durrett 1996; Keeling & Grenfell 2000).

It has been suggested that sex enhances the maintenance of genetic diversity (Bürger 1999) and population persistence (Maynard Smith 1978; Galvani *et al.* 2001; Getz 2001). In a previous paper, we demonstrated that antigenic variation enhances the persistence of parasite populations, for which there is also empirical support (Palmer *et al.* 2000). We showed that sexual parasites are able to generate greater strain diversity from a set of homozygous strains through Mendelian segregation, compared with asexual parasites that produce sufficient diversity from mutation alone (Galvani *et al.* 2001). A new parasite strain initially has a fitness advantage, derived from the evasion of pre-existing host immunity. This advantage diminishes as the parasite strain proliferates and hosts develop immunity against it. In this way, the density-dependent relationship between host population immunity and infection levels of particular parasite strains selects for strain diversity in parasites. Thus, strain diversity is evolutionarily advantageous for parasites (Ferguson & Galvani 2002). As cross-immunity increases, the advantage of strain diversity decreases.

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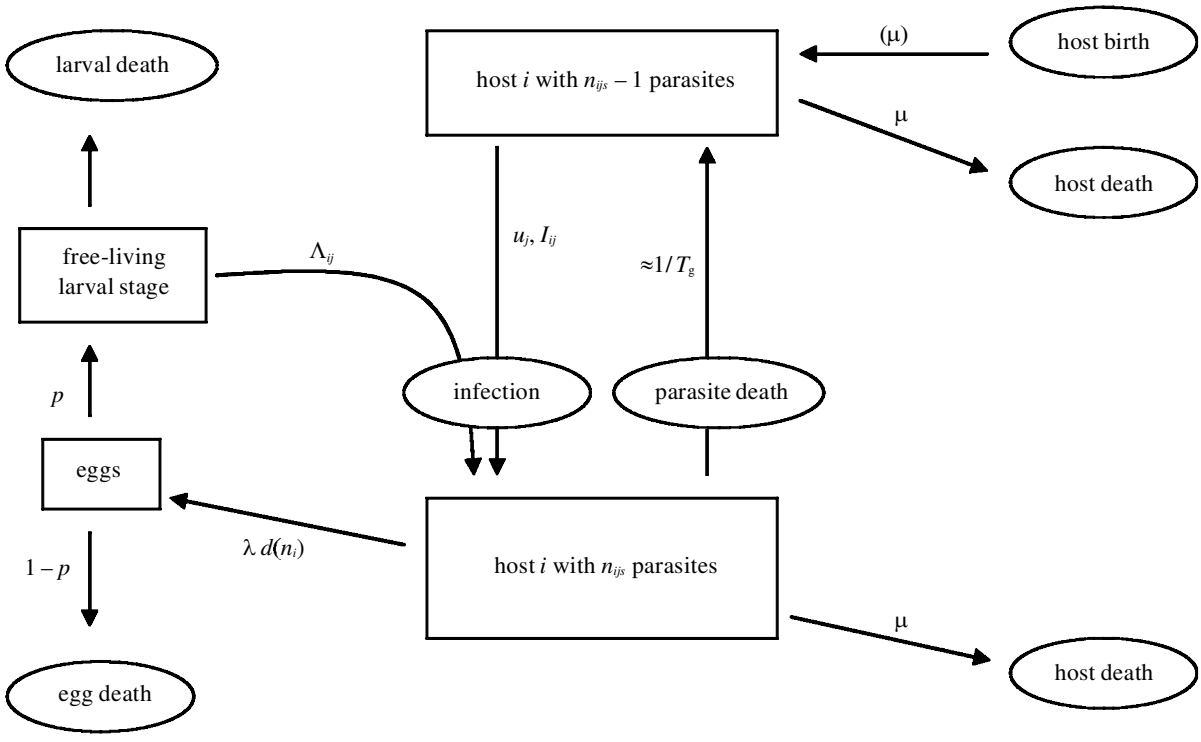

Figure 1. Flow diagram representing the major features of the model. Boxes denote host/parasite states and ovals denote birth, death or infection events. Relevant parameters and variables that influence the probability of each process are also given. See table 1 for definitions of model parameters and variables.

Table 1. Model parameters and variables.

	definition
parameters	
γ	cross-immunity
T_g	parasite generation time
β	probability that a susceptible host exposed to a parasite larvae will become infected
c_i	predisposition to infection of host i
λ	maximum parasite fecundity, subject to density-dependent competition $d(n_i)$
p	proportion of eggs that survive to become infective larvae
Θ_{jk}	measure of antigenic relatedness between strains j and k
H_{jlq}	proportion of offspring of matings between genotypes l and q that produce genotype j
δ	mutation rate
Ω_{jf}	proportion of antigenic mutations of parasites with genotype f that generate genotype j
N	size of host population
μ	rate of host mortality
L	number of strains in the system
variables	
n_{ijs}	number of parasites of genotype j of gender s in host i
I_{ij}	strain-specific immunity of host i against parasite strain j
Λ_{ij}	rate of exposure of host i to larvae of strain j

Previous models have suggested that fluctuating epistasis contributes to the maintenance of sex in hosts (Barton 1995; Peters & Lively 1999). Furthermore, a previous deterministic model of the evolution of sex in parasites showed that fluctuating epistasis can maintain polymorphism between sexual and asexual parasites, but was not able to generate a selective advantage for sex that overcame its twofold cost (Lythgoe 2000). Here we demonstrate that the advantages of sexual reproduction can surpass those of asexual reproduction. Results from both our individual-based simulation model and our simpler analytically tractable model show that sexual parasites subject to strain-specific immunity can achieve a larger population size (i.e. higher levels of infection), greater levels of strain diversity and better persistence than asexual parasites. We examine the sensitivity of this strain diversity and the size of the parasite population to the generation time of the parasite, as well as the degree of cross-immunity elicited between antigenically related strains.

We also question whether sexual reproduction is resistant to invasion from a rare asexual mutant for different levels of cross-immunity. We consider the probability of asexual invasion for different generation times of the parasite, rates of parasite reproduction and host population

sizes. We suggest that the superior ability to generate and maintain strain diversity can account for the evolutionary success of sexual helminth parasites, even for high levels of cross-immunity between strains.

2. MODEL STRUCTURE

Our stochastic, individual-based model incorporates the reproduction and transmission dynamics of a directly transmitted helminth parasite, such as the whipworm *Trichuris trichiura*. Model parameters are described below, represented in the flow diagram of figure 1 and summarized in table 1.

The sexually reproducing parasites are assumed to be polygamous (Anderson & May 1991). Sexual parasites are also assumed to mate outside the host (cf. Galvani *et al.* 2001), allowing us to examine whether the twofold fecundity cost of sex could be overcome, rather than whether finite mating probabilities (MacDonald 1965) impose further costs on sex. Furthermore, previous epidemiological modelling has suggested that the mating probability is unimportant to helminth epidemiology in practice (May 1977).

Parasite strains are defined by polymorphism at an antigenic locus. We assume that when a parasite strain infects a host, the host develops immunity specific to the antigenic genotype of that strain. This strain-specific immunity prevents reinfection by the same strain. Between strains that share an antigenic allele, the model also incorporates a degree of cross-immunity (γ) which ranges from 0 to 1. Thus, if γ is 0, immunity is assumed to be entirely strain-specific, and if γ is 1, host immunity is antigen-specific and hence develops across all strains that share an antigen.

The model is formulated as an individual-based, stochastic simulation, with time step Δt (10 days). Let Δn_{ijs} represent the changes in one time step of the numbers of parasites of a genotype j of gender s (M for male or F for female) in an individual host i, and let ΔI_{ij} represent the consequent immunity to strain j in host i. The change in number of infections of host i with parasites of strain j and sex s occurring in a time-step is denoted as ΔY_{ijs}. A binomially distributed random deviate generated from B Bernoulli trials, each with probability q of success, is denoted as Bin(q, B). Defining Λ_{ij} as the rate of exposure of host i to larvae of parasite strain j, and I_{ij} as the level of strain-specific immunity (assumed to be lifelong) of host i against parasite strain j, the model dynamics are given by

$$\Delta n_{ijs} = \Delta Y_{ijs} - \mathrm{Bin}\left(\frac{\Delta t}{T_\mathrm{g}}, n_{ijs}\right),$$

$$\Delta I_{ij} = 1 - I_{ij} \quad \text{if} \quad \sum_s \Delta Y_{ijs} > 0$$

$$= 0 \qquad \text{if} \quad \sum_s \Delta Y_{ijs} = 0,$$

where

$$\Delta Y_{ijs} = \mathrm{Bin}\left[\beta(1 - I_{ij})\left[1 - \gamma\left(1 - \prod_{k \neq j} \Theta_{jk} I_{ik}\right)\right]\Delta t/2, \Lambda_{ij}\right],$$

$$\Lambda_{ij} = c_i \lambda p \frac{\sum_w \exp\left[-\left(\sum_{l,s} n_{wls} - 1\right)\Big/\alpha\right]\sum_{z,l,q} n_{zqM} n_{wlF}\left[(1-\delta)H_{jlq} + \delta \sum_g \Omega_{jg} H_{glq}\right]}{\sum_{z,q} n_{zqM}}.$$

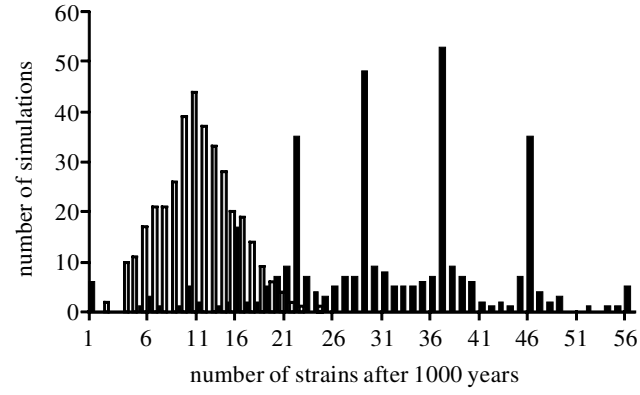

Figure 2. The distribution of the number of strains persisting after 1000 years from 365 simulations for sexual (black bars) versus asexual (open bars) systems when they evolve independently. The peaks in the sexual distribution correspond to the full range of genotypes generated by different numbers of alleles. The spread around these peaks is caused by a new mutation arising, but not yet having had enough time to combine with all other alleles through segregation and/or the temporary extinction of strains. With increasing parasite population size, we would expect the spread between peaks to decrease. Larger parasite populations support faster rates of generation of the full range of genotypes thence segregation thence the coming together of genotypes with all possible alleles. Larger parasite populations also reduce extinction probabilities. Here, strain-specific immunity confers complete protection against reinfection, there is no cross-immunity ($\gamma = 0$), parasite generation time is $T_\mathrm{g} = 36$ months and host population size is 1000. Similar diversity patterns are seen for all $\gamma < 0.9$, though overall parasite abundance decreases as γ increases.

In these equations, T_g (36 months) is the average lifespan of adult parasites; β (0.1; giving $R_0 = 13$) is the probability that a susceptible host exposed to a parasite larva will become infected (all strains are assumed equally transmissible); λ (12 000 per day) is the maximal output of larvae by adult females; and p (10^{-5}) is the proportion of larvae surviving to encounter hosts. Heterogeneity in predisposition to exposure between hosts is represented by c_i, drawn from a negative binomial distribution. In turn, the predisposition heterogeneity generates an aggregated distribution of parasite burdens in the host population, which empirical studies repeatedly reveal in communities infected by helminth parasites (Anderson & May 1991). Parasite fecundity is assumed to be reduced by density-dependent competition between parasites (Keymer 1982), through a negative exponential function of parasite burden, $d(n_i) = \Sigma_w \exp[-(\Sigma_{l,s} n_{wls} - 1)/\alpha]$, consistent with empirical data (Cheever 1968; Anderson & May 1985; Medley & Anderson 1985), where α (= 2) is an inverse measure of the intensity of density dependence. (We also test the effect of removing density dependence to the selective advantage of sexual reproduction.) The host population size is assumed to be constant, with host survival being exponentially distributed with an average of 50 years.

Parameter Θ_{jk} specifies the antigenic relatedness between strains j and k (= 0 if j and k share an antigenic allele, = 1 otherwise). H_{jlq} is the proportion of offspring of matings between genotypes l and q that produce genotype j, δ is the rate of antigenic mutation per offspring pro-

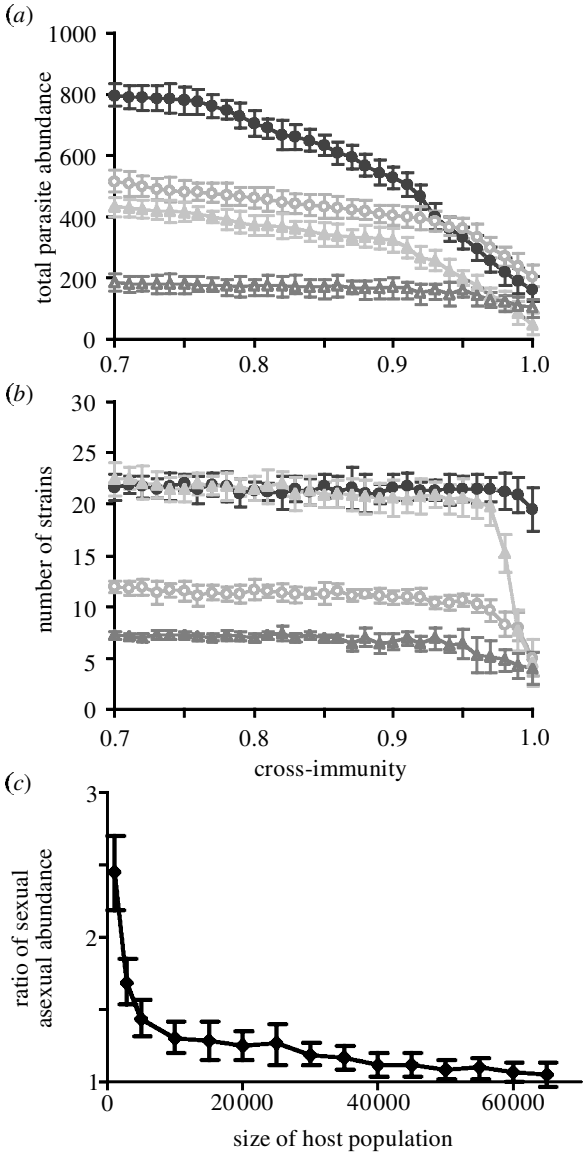

Figure 3. Change in strain diversity and population size with varying cross-immunity for sexual versus asexual parasite populations. (*a*) Equilibrium parasite population sizes, and (*b*) number of strains for sexual versus asexual parasites as a function of cross-immunity, γ, for parasite generation times of $T_g = 36$ and 18 months, respectively. Standard errors are plotted. Increasing cross-immunity (γ) reduces parasite population size more rapidly for systems with higher diversity, due to the greater antigenic overlap between strains. Sexual systems maintain near maximal diversity (all 21 genotypes of the six alleles originally introduced) for $\gamma = 0.95$, while asexuals rapidly lose diversity. Parasite population size and diversity fall more rapidly for $\gamma = 0.95$, and in this regime the greater innate fecundity of asexuals allows them to outcompete sexuals. Increasing parasite generation time (while maintaining R_0) increases equilibrium parasite population size, and asexual diversity. For computational efficiency, the duration of each simulation was 50 years—sufficient for quasi-equilibrium of infection levels to be reached, but not for diversity to fully equilibrate given the long time-scale of mutation. True asexual diversity equilibrium is lower than shown here, and sexual diversity higher. When varying parasite generation time, parasite fecundity was adjusted to keep R_0 constant. Other parameters were as in figure 2. Filled circles denote 38 months sexual generation time; open circles denote 36 months asexual generation time; filled triangles denote 18 months sexual generation time; and open triangles denote 18 months asexual generation time. (*c*) The ratio of population sizes of sexual to asexual parasites circulating in separate host populations as a function of host population size. $T_g = 36$ months and $\gamma = 0.5$.

duced (10^{-6}), and Ω_{jf} is the proportion of antigenic mutations of parasites with genotype f that generate genotype j. Up to ten alleles of a single antigenic locus can be modelled simultaneously, giving a maximum of 55 co-circulating diploid parasite strains.

The dynamics of asexual parasite populations are given by a simplified form of the equations above, without gender or segregation. To assess the resistance of the sexual population to invasion from a rare asexual mutant, a randomly selected reproductively mature sexual parasite was assumed to mutate to asexual reproduction. The asexual and sexual parasites then compete within the same host population.

Simulations are run for 1000 years (or in the case of the invasion results, until either sexuals or asexuals are driven extinct). For most of our results, we initiate the system with 55 strains (arising from 10 alleles at the diploid locus), but we also compare this with an initial system of 21 strains (arising from six alleles).

(a) *Analytical consideration of parasite population persistence*

For sexual versus asexual parasites, we derive approximate analytical results for the critical community size (CCS), which we define (in an evolutionary rather than epidemiological context) as the host population size for which there is a 0.001 probability of extinction of all parasite strains within 1000 years. We employ an approximate measure of time to extinction of an SIR (i.e. hosts can be either susceptible, infected or recovered) epidemic model derived by Nasell (1999) (see Appendix A).

If $T(N,\mu,\sigma,R_0)$ is the expected time to extinction of a single strain in an SIR model with host population size N, host mortality μ and parasite clearance σ, then the expected time to extinction of a system in which L immunologically distinct and independent strains are co-circulating (representing an asexual system) is given by

$$T_A(L,N,\mu,\sigma,R_0) = (\Gamma + \ln(L))T(N,\mu,\sigma,R_0),$$

where there is no genetic mixing between strains and Γ is Euler's constant (see Appendix A).

If one approximates the equivalent sexual system as an L-strain SIR model with complete random genetic mixing between strains (i.e. the force of infection for any one strain is given by the average force of infection of all strains $\lambda_i = \beta\Sigma_j y_j/LN$), then the persistence properties of the system are equivalent to those of a one-strain system with a population size of $L \times N$ (see Appendix A). Hence, the expected time to extinction of such a sexual system is given by

$$T_S(L,N,\mu,\sigma,R_0) = T(LN,\mu,\sigma,R_0/2),$$

where immunity is entirely strain-specific and the sexual parasite system has half the R_0 of the asexual system to account for the twofold cost of sex, assuming a 1 : 1 sex

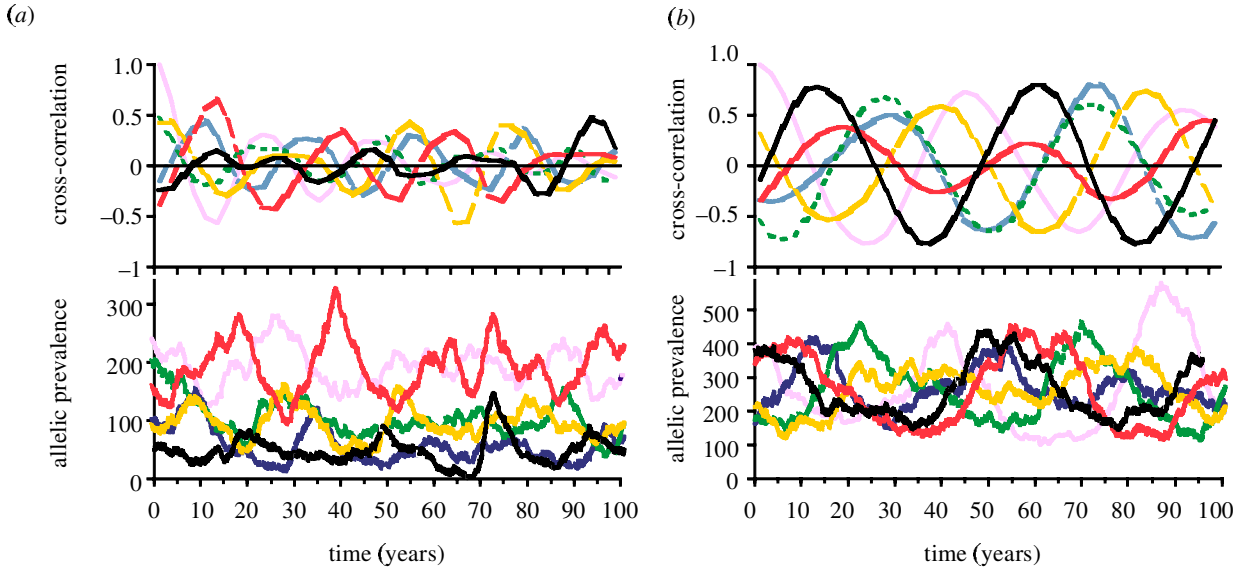

Figure 4. Time-series of the prevalence of different alleles in sexual versus asexual parasite populations. Allelic prevalence is defined as the number of alleles in a parasite population, that is, the sum of the number of heterozygous parasites and double the number of homozygous parasites for a given allele. (*a*) Prevalence of six antigenic alleles in a population of asexual parasites within a host population of 1000 after 500 years equilibration, in addition to the cross-correlation function (calculated from 500 years of time-series) between allele 1 (pink line) and alleles 2–6 (2, blue line; 3, green dashed line; 4, yellow dashed line; 5, red dashed line; 6, black line). Allelic (and genotypic) frequencies exhibit limit cycles with a period of *ca*. 25 years (half the host lifespan), although stochasticity disrupts their regularity to some extent. (*b*) Sexual system. More defined cycles are seen, with a period of 50 years (the host lifespan), higher and more consistent allelic frequencies (a result of the much greater genetic mixing induced by recombination) and thus higher trough frequencies, giving better overall persistence. Other parameters were as in figure 2.

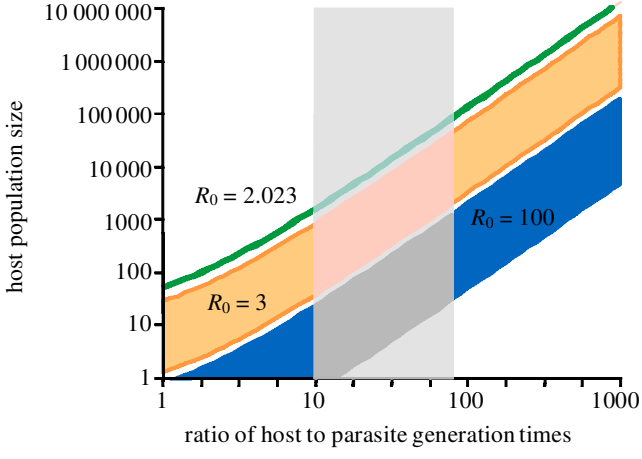

Figure 5. The effect of host population size and the ratio of host to parasite generation times on the CCS of antigenically diverse sexual and asexual parasite populations. The shaded diagonal bands represent ranges of host population size where the sexual population is above its CCS but the asexual population is not, as a function of the ratio of host to parasite generation time, σ/μ, and for three values of R_0 (thin green band represents $R_0 = 2.023$; orange band represents $R_0 = 3$; blue band represents $R_0 = 100$). For some values of R_0 between 3 and 100 (e.g. 15), the CCS region would partially overlap with both orange and blue bands. The vertical grey region represents ratios of host to parasite generation times that are typical of helminth parasites. Here, $L = 100$. There is a general trend for both sexual and asexual persistence to improve with increasing R_0.

ratio. Values of the CCS for these two systems are then obtained by solving the following equations for N:

$$1 - \exp\{-1000/T_S(L,N,\mu,\sigma,R_0)\} = 0.001,$$

$$1 - \exp\{-1000/T_A(L,N,\mu,\sigma,R_0)\} = 0.001.$$

These transcendental equations were solved numerically, using an approximate functional form of the time to extinction for an SIR system derived by Nasell (1999):

$$T(N,\mu,\sigma,R_0) \approx \frac{\sqrt{2\pi(R_0-1)N}}{(\mu+\sigma)R_0}\exp\left[\frac{\mu^2(R_0-1)N}{2(\mu+\sigma)^2}\right].$$

In the limit of $R_0 \gg 1$, it can be shown analytically that the ratio of the CCSs of the sexual versus asexual systems tends to $L/2$. For $L \gg 2$, the sexual system has consistently better persistence than the asexual system ($T_S \gg T_A$). Due to the sigmoidal relationship between the probability of extinction in a given time interval and population size, we formulate this analysis in terms of a threshold population size above which extinction is infrequent (0.001 probability in 1000 years)—our CCS concept. Stochastic extinction has the most significant impact on observed dynamics in the region of the CCS.

3. RESULTS

(a) *Parasite population size and strain diversity*

We find that sexual parasites outperform asexual parasites in terms of higher infection levels and greater strain diversity. First, we initiate the system with all of the possible 21 genotypes that can arise from six antigenic alleles

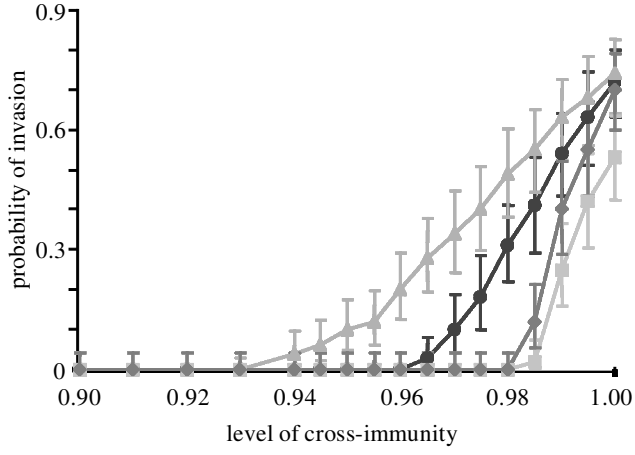

Figure 6. The probability of successful asexual invasion of a sexual parasite population as a function of cross-immunity when (i) parasites have an average generation time of $T_g = 36$ months in a population of $N = 1000$ hosts (circles); (ii) $T_g = 18$ months and $N = 1000$ (triangles); (iii) $T_g = 36$ months and $N = 1500$ (squares), and (iv) $T_g = 36$ months and $N = 1000$, with R_0 increased by 50% to 19.5 (diamonds). Binomial confidence intervals of 95% are shown. At least 100 realizations were performed for every point, each being run for *ca.* 50 years before an asexual mutant was introduced into a host. The dynamics are then tracked until either the sexual or asexual population became extinct. Invasion probabilities were highest when sexual parasite abundance was lowest and thus extinction rates highest, that is, brought about by reducing parasite generation time (T_g), decreasing R_0 or increasing host population size (N).

at the single diploid locus. We find that sexual reproduction maintain significantly higher levels of diversity than asexual reproduction (average of 30 ± 1 (s.e.) versus 9 ± 0.4 strains; figure 2). The asexual population reaches a strain diversity equilibrium resulting from the balance between the rates of emergence and extinction of strains. Parasite strains are much more likely to be retained in a sexual population. If a genotype is lost from a sexual population, it can be recreated by the sexual reproduction of persisting related strains. These differences in strain diversity also results in differences in parasite population size between sexuals and asexuals (average of 1120 ± 50 versus 390 ± 20 parasites). Enhanced strain diversity allow parasites to escape pre-existing host immunity, thereby boosting levels of infection.

Second, we initiate the system with all 55 strains. We find that sexual populations are able to maintain the full set of 55, again because strains that temporarily go extinct can be recreated by sexual reproduction. This strain diversity gives rise to an average sexual population size of 1770 ± 60 (compared with 1120 ± 50 when the system is initiated with 21 strains). By contrast, the asexual population loses strains from the initial 55 to reach an equilibrium of an average 9 ± 0.4 strains and 400 ± 30 parasites, which is very similar to the equilibrium of the asexual system initiated with 21 strains. Thus, the asexual system appears less sensitive to the initial level of antigenic diversity than the sexual system.

We find that the higher population size and diversity of sexual systems relative to asexuals is robust to (albeit reduced by) substantial cross-immunity (figure 3). The sexual advantage increases as the generation time of the parasite is lengthened (from 18 to 36 months).

(b) *The selective value of sex and stochastic extinction*

Both sexual and asexual systems exhibit limit cycles in allele frequencies with a time-scale determined by host generation time (figure 4), an expected result of the intense frequency-dependent selection induced by the lifelong, strain-specific host immunity. Low troughs in genotype frequencies increase the rates of stochastic extinction of alleles in asexual populations. By contrast, sex reduces temporal variability in average genotype and allele frequencies, resulting in more regular cycles, with higher trough frequencies and a lower probability of allelic extinction compared with asexuals.

We prevent stochastic extinction of individual strains in our model to test the importance of this factor to the selective value of the reproductive strategies. (This is achieved by making no copies of a particular allele a reflecting boundary rather than an absorbing boundary.) We find that the population size of asexuals increases significantly from 400 ± 30 to 1980 ± 90 parasites when extinction is prevented. However, the average size of the sexual population (1770 ± 60 versus 1780 ± 50) is not affected significantly by preventing extinction, because Mendelian segregation recreates strains that have gone temporarily extinct. Without stochastic extinction, asexual reproduction becomes more successful (in terms of population size) than sexual reproduction, although by less than twofold.

We also explore extinction probabilities of sexual versus asexual parasite populations analytically. We compare CCS for parasite populations as R_0 and the ratio of host to parasite generation times are increased, respectively. The sexual population is above its CCS while the asexual population is below its CCS over a considerable region of parameter space (figure 5). For fixed R_0, sexual populations continue to maintain a higher probability of persistence than asexual populations, even in relatively large host population sizes, as the ratio of host to parasite generation times increases. While stochasticity in small populations favours sex, sexual populations maintain greater infection levels than asexual populations even when the host population is increased considerably (figures 3*c* and 5).

Finally, when we remove density-dependent constraints on fecundity (while decreasing β to maintain previous numbers of sexual parasites, *ca.* 1770), the average population size of asexual parasites rises from 400 to 1070. Removing density dependence reduces the probability of stochastic extinction. This benefits asexual parasites more than sexual parasites, the latter being intrinsically resistant to stochastic extinction. Furthermore, the sensitivity of the selection balance of reproductive strategies to the inclusion of density dependence indicates that biological realism (of which density dependence is a component) is an important component of models that attempt to account for the maintenance of sex.

(c) *Resistance to invasion*

If sex is an evolutionarily stable strategy (Maynard Smith & Price 1973) for helminth parasites, the sexual

population will resist invasion from an asexual mutant. We find that the probability of asexual invasion increases with cross-immunity (figure 6). Cross-immunity generates competition between strains, and thus lowers the selective value of strain diversity. The probability of invasion rises significantly at very high levels of cross-immunity, mirroring figure 3. This is consistent with the erosion of the advantage of antigenic diversity with increasing cross-immunity.

We find that the probability of establishment for an asexual parasite entering an uninfected population (i.e. with no sexual parasites) was virtually 100%. By comparison, when a sexual population equilibrium is already present, the probability of invasion is much less than unity, except for very high levels of cross-immunity, confirming that competition from the sexual population prevents establishment of asexual parasites.

As the basic reproductive rate, R_0 (Anderson & May 1991), of the parasite population is increased, the probability of asexual invasion falls (figure 6). Increasing R_0 has two effects on the ability of sexual populations to resist invasion, both related to increasing parasite population size with R_0. First, a larger parasite population results in a greater proportion of immune hosts at the time of invasion. Second, the net mutation rate also increases with parasite population size, generating greater strain diversity of the sexual population before mutation to asexuality. Likewise, an increase in the size of the host population also reduces the probability of sexual displacement, because larger host populations supported a larger sexual parasite population, making extinction of the sexual population less likely. Consistent with figure 4, a longer parasite generation time is correlated with enhanced evolutionary stability of sex.

4. DISCUSSION

We have seen that sex benefits from the ability to regenerate lost genotypes (Hamilton *et al.* 1981). This has been termed the 'gene-storage effect' (Hackett *et al.* 1987), in which sex uses alleles 'stored' in other genotypes to recreate a lost genotype (Otto & Michalakis 1998). Furthermore, Mendelian segregation allows a new mutation in a single-locus system that already has n alleles to give rise to a further $n + 1$ strains (n new heterozygotes and one new homozygote). Therefore, the number of strains at equilibrium in a sexual system rises quadratically with the number of alleles, compared with a linear increase for an asexual system.

Results from both our analytical model and our individual-based model highlight the importance of stochastic persistence of antigenic diversity to the selective value of sex. The role that stochasticity has in generating a relative advantage for sex may also explain the discrepancy between our results and those of previous deterministic models that did not overcome the twofold cost of sex (Lythgoe 2000). Stochasticity is also fundamental to the 'demographic balance hypothesis' (Getz 2001) that identifies the importance of population persistence to the distribution of sexual and asexual reproduction in endeostigmatid mites.

Stochasticity becomes relatively less important in larger populations, but spatial structuring of infection dynamics in larger host populations may also contribute to sexual advantage (Keeling & Rand 1995). Alternatively, spatial structuring could also lead to inbreeding in sexual populations, resulting in the local fixation of homozygous genotypes. Explicit modelling of spatial structuring in host populations is required to determine which of these factors dominates.

Fluctuating epistasis can make gene combinations that are adaptive in one generation rapidly less favourable (Charlesworth 1976; Maynard Smith 1978; Hamilton 1980; Hutson & Law 1981; May & Anderson 1983; Sasaki & Iwasa 1987; Barton 1995). It has been proposed that fluctuating selection of different host genotypes favours sex in hosts (Jaenike 1978; Bremermann 1980; Hamilton 1980; Seger & Hamilton 1988) and reciprocally for parasites (Lythgoe 2000). Fluctuating epistasis requires a selective interaction among loci in a multiple-locus system. Consequently, the selective value of sex that we observe cannot be due to fluctuating epistasis, because our model is based on segregation at a single diploid locus. Moreover, our results show that fluctuating epistasis is not necessary to generate a selective advantage for sex. It would be interesting to incorporate multiple loci in our model framework to identify the effects of recombination among loci, linkage disequilibrium and fluctuating epistasis, relative to the effects of segregation of alleles within a locus. As the number of loci increases, we would expect that segregation would become more important in generating genetic diversity than recombination.

Competing theories, such as the mutational-related hypotheses, are being discredited, specifically in helminths, by recent estimates of mutation rates (Keightley & Eyre-Walker 2000), the nature of synergistic interactions between mutations (Elena & Lenski 1997; Moore *et al.* 2000; Peters & Keightley 2000) and by population genetic models (Kondrashov & Kondrashov 2001). However, our results do not exclude contributions from other factors to the selective value of sex (Howard & Lively 1994; Otto & Michalakis 1998; Peters & Lively 1999; West *et al.* 1999). A threshold of initial genetic diversity is required to give sex the selective advantage that we identify here. It is feasible that the origin of sexual reproduction was driven by selection mechanisms that are now less important in its continued maintenance. Further elaborations of the model could explore the repercussions of deleterious mutations. A model that incorporates a proportion of mutations that reduce fitness in addition to a proportion that permit immune evasion could be used to address the interaction between the selection pressures considered here and the purging of deleterious mutations (Kondrashov 1988).

Our results provide an explanation for observed correlations between parasite reproductive strategy and ecological niche. Free-living helminth species are more often sexual than parasitic species (Glesener & Tilman 1978; Maynard Smith & Szathmary 1995). Helminth parasites of host species with a poor immune response are also less likely to be sexual, or to exchange genetic material, than those that mount a highly protective response (Poinar & Hansen 1983). For instance, the helminth *Stronglygoides ratti* is more likely to reproduce sexually in hosts expressing a strong immune response and asexually in immunocompromised hosts (Gemmill *et al.* 1997; West *et al.* 2001). Furthermore, our finding that sexual advantage

increases with generation time of the parasite may help to explain the distribution of asexuality in pathogens. Viruses with very short generation times, high mutation rates and large population sizes are able to generate sufficient diversity without sexual reproduction. Bacteria with generation times that are slower than those of viruses are asexual, but have mechanisms for exchanging genetic material. Helminths with longer generation times still employ complete sexual reproduction. In addition, the helminth *Toxoplasma gondii* is sexual in its definitive host but is asexual in the range of other species it also infects (Berdoy *et al.* 2000). From an evolutionary perspective, it can be expected that definitive hosts are selected to develop more effective immunity against parasites to which they are frequently exposed than hosts that are rarely exposed. Indeed, experimental studies demonstrate that cats develop protective strain-specific immunity against *T. gondii* (Omata *et al.* 1996). Our theory is consistent with these observations and the occurrence of the sexual phase of the parasite in the definitive feline host only, in which evasion of effective immunity will be most important.

Concomitant immunity (where adult parasites persist within hosts despite immunity against reinfection) is a parasite adaptation to evade host immunity, minimize within-host competition between strains and enhance persistence (Brown & Grenfell 2001). Interestingly, schistosomes exploit concomitant immunity to the greatest extent of the helminth parasites and, uniquely among helminths, have a monogamous mating system (Anderson & May 1991). Our proposal may help to explain these observations. Promiscuity generates greater genetic mixing than monogamy. Greater reliance on an alternative mechanism to minimize intraspecific competition (i.e. concomitant immunity) would be expected to reduce selection on the production of diverse offspring. Also consistent with the immune-evasion value of sex for parasites is the observation that the sex ratio of the malarial parasite *Plasmodium gallinaceum* switches from predominately female early in infection to 1 : 1 later in infection (Paul *et al.* 2000). This finding suggests that the production of genetically diverse offspring becomes particularly important later in infection once immunity has developed against the parental strains.

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APPENDIX A: CRITICAL COMMUNITY SIZE OF SIMPLE MULTI-STRAIN EPIDEMIC MODELS WITH GENETIC MIXING

Consider a pathogen with L strains, each of which generates complete strain-specific immunity when it infects a host, but no cross-immunity between strains. We employ an SIR model modified to represent multi-strain systems (after Gupta *et al.* 1996). Unlike the standard SIR model, the S and I classes are overlapping. That is, y_i (number of hosts infected with strain i $(1 \leqslant i \leqslant L)$) is a subset of x_i (the number of hosts that have been exposed to strain i). Consequently, $x_i - y_i$ is equivalent to the R class in a standard SIR model. Likewise, if N is the total host population size, then $N - x_i$ is the number of hosts that are susceptible (not exposed) to strain i. Assuming homogenous mixing of the host population, the deterministic state equations of the system are

$$\frac{\mathrm{d}x_i}{\mathrm{d}t} = \lambda_i(N - x_i) - \mu x_i,$$

$$\frac{\mathrm{d}y_i}{\mathrm{d}t} = \lambda_i(N - x_i) - (\sigma + \mu)y_i.$$

If there is no genetic exchange between strains (representing asexual reproduction), the force of infection of strain i is given by $\lambda_i = \beta y_i/N$, while complete genetic mixing (sexual genetic mixing) can be approximated by assuming $\lambda_i = \beta\Sigma_{j=1}^{L} y_j/NL$. The latter expression describes a system with maximal genetic mixing; that is, the composition of secondary infections is given by random genetic mixing of all strains in the system. Clearly, this is a highly simplified approximation of the sexual reproduction of our individual-based model. In addition, the expected persistence of this system will be somewhat longer than that of a system with Mendelian segregation (as in our individual-based model), because extinction of a strain requires not only extinction of all strains sharing alleles with the given strain, but that of all strains in the system. The basic reproductive ratio of a single strain is given by $R_0 = \beta/(\sigma + \mu)$ for this model, as for our individual-based model.

We examine the extinction properties of the stochastic versions of these models. For the asexual system, if extinction of a single strain in isolation occurs as a Poisson process, with rate $1/T$ and average time to extinction T, the independence of strains implies that the average time to extinction of the first strain in an L-strain system is T/L; the time to extinction of the next strain is $T/(L - 1)$; and the time to extinction of the last strain is T. Thus, the overall time to extinction of all L strains is given by the harmonic series sum $T\Sigma_{j=1}^{L}(1/j) \approx (\Gamma + \ln(L))T$, where $\Gamma(= 0.577...)$ is Euler's constant.

For the sexual system, we note that summation over the equations describing the dynamics of all L strains gives a single-strain SIR model with a host population size of $L \times N$. Thus, the expected time to extinction of L sexual strains circulating in a population of N hosts is the same as the expected time to extinction of a single strain circulating in an L-fold larger host population.

This analysis, together with the approximate expression for the expected extinction time of the simple SIR model derived by Nasell (1999) are used to produce the results presented in the main text.

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