High levels of mitotic gene conversion are needed to effectively purge deleterious mutations in asexual organisms

Dominik Kopčak[1,2,3], Matthew Hartfield[1,*]

1) Institute of Ecology and Evolution, The University of Edinburgh, Edinburgh EH9 3FL, UK.
2) Institute of Science and Technology Austria, 3400 Klosterneuburg, Austria.
3) Okinawa Institute of Science and Technology Graduate University, Onna-son, Okinawa, Japan.
* Corresponding author: m.hartfield@ed.ac.uk

Key words: Self-fertilisation, facultative sex, gene conversion, deleterious mutation, dominance, mutation load.

Data availability statement: Simulation code and Conda environments with all used packages, together with all scripts have been archived on GitHub (https://github.com/dominik-kopcak/SelfClonGC).

Author contributions: MH designed the project with input from DK. DK wrote and ran the simulations, and analysed the resulting outputs with feedback from MH. MH developed and analysed the single-locus model. MH and DK wrote and edited the manuscript.

Funding: Matthew Hartfield is supported by a NERC Independent Research Fellowship (NE/R015686/1) and a UKRI Frontier Research Guarantee Grant (EP/X027570/1).

Conflict of interest statement: Nothing to declare.

Acknowledgements. We would like to thank Anders Poulsen Charmouh for advising on how to implement the non-constant *s*, *h* model, and members of the Hartfield lab and three anonymous reviewers for providing feedback on the work.

ORCID links:
Dominik Kopčak: https://orcid.org/0009-0005-3758-4390
Matthew Hartfield: https://orcid.org/0000-0001-6941-0647

Abstract

Self-fertilisation and asexual reproduction are both hypothesised to cause long-term extinction due to inefficient selection against deleterious mutations. Self-fertilisation can counter these effects through creating homozygous genotypes and purging deleterious mutations. Although complete asexuality lacks meiotic gene exchange, mitotic gene conversion creates homozygous regions that could limit deleterious mutation accumulation in an analogous manner. We compare mutation accumulation in self-fertilising and facultative sexual populations subject to mitotic gene conversion, and quantify the efficacy of purging in the latter. We first show analytically that purging is most effective with high levels of asexuality and gene conversion, and when deleterious mutations are recessive. We further show using simulations that, when mitotic gene conversion becomes sufficiently high in obligate asexuals, there is a reduction in the mutation count and a jump in homozygosity, reflecting purging. These effects are stronger than predicted from single-locus models, demonstrating how multi-locus purging can further reduce mutation prevalence in asexuals. However, this mechanism is not necessarily as efficient at purging under high self-fertilisation, and elevated rates of mitotic gene conversion seem to be needed for widespread purging compared to empirical estimates.

Teaser Text

Asexuality has been argued to be an evolutionary ‘dead end’, due to a lack of gene exchange causing inefficient selection acting against deleterious mutations. It has been proposed that asexuals can counter these negative effects through mitotic gene conversion, which exposes mutations to selection within individual lineages. Here, we theoretically investigate how effective this mechanism is. We compare results to those obtained when individuals reproduce by self-fertilisation, which has similar effects on exposing deleterious variants. While mitotic gene conversion can be effective in removing recessive deleterious mutations, high rates are required and it is not always as effective as selfing.

Introduction

Mutations are the ultimate source of genetic novelty in all organisms and are essential for biological evolution (Loewe and Hill 2010). However, the generation of variation is a double-edged sword; although favourable mutations may emerge that are crucial for long-term survival, it may also result in a burden of deleterious mutations, which are much more common than favourable ones (Eyre-Walker and Keightley 2007).

The reproductive system, which determines how offspring are generated in a population, has a profound impact on the prevalence of deleterious variation. Reproduction can either be with or without sex. Sex usually involves two mechanisms (Hartfield and Keightley 2012): recombination (the exchange of genetic information between homologous DNA sequences, most commonly as meiotic crossing-over), and fusion of gametes (specialised cells with halved ploidy and DNA content). In the simplest case when asexuality is equivalent to clonal reproduction, the genotype of an offspring is identical to that of a parent, except for *de novo* mutations. Clonal reproduction encompasses diverse mechanisms, including fission or budding in unicellular eukaryotes, or budding (e.g., hydra, jellyfishes), fragmentation (e.g., corals) and whole-body fission (e.g., flatworms, starfish) in animals, or many forms of vegetative propagation in plants (De Meeûs et al. 2007). Nevertheless, most eukaryotic organisms undergo some variant of sexual reproduction, at least occasionally (Speijer et al. 2015). Facultatively sexual organisms reproduce via a mixture of sexual and asexual reproduction, so that genetic evolution is affected by the frequency of both recombination and clonal reproduction (Hartfield 2016).

Gametes from which offspring originate may also be derived from the same individual, leading to self-fertilisation or selfing. The opposite of selfing is panmictic outcrossing, with anyone in the population having an equal chance of being a mate of anyone else, and self-mating being rare or even prohibited (if, e.g., self-incompatibility mechanisms are present; Nettancourt 2001). Cloning and selfing are two types of uniparental reproduction, which forms a basis of their similarity (e.g., the lack of mate finding) and potential ecological advantages

95 (e.g., reproductive assurance; Baker 1955, 1967; Theologidis et al. 2014; Mráz and Mrázová
96 2021).

97 Despite these apparent similarities, cloning and selfing differ in the manner in which
98 they affect selection acting against deleterious mutations. Muller's ratchet operates in asexual
99 populations leading to stochastic mutation accumulation, due to their inability to generate
100 offspring that have fewer deleterious alleles than those of their ancestors (Muller 1964; Haigh
101 1978). This mechanism is a form of selection interference, which refers to the effect of strong
102 genetic linkage on the dynamics of variants under selection (Felsenstein 1974). In completely
103 clonal populations all sites are in absolute genetic linkage, therefore the presence of multiple
104 deleterious alleles selectively interferes with one another having a strong impact on mutation
105 accumulation (Hill and Robertson 1966; Roze and Barton 2006; Hartfield and Keightley 2012).
106 Recombination breaks this linkage and allows the dynamics of variants to respond to selection
107 independently, i.e., more efficient removal of deleterious mutations and restoration of the
108 most-fit genotype. Another important consequence due to the lack of gamete fusion in diploid
109 asexuals is the divergence of homologous chromosomes found within one individual. The two
110 genome copies are always transmitted together across the lineage, gradually and
111 independently accumulating mutations leading to a high level of heterozygosity; a process
112 known as 'allelic sequence divergence' or the Meselson effect (Mark Welch and Meselson
113 2000; Butlin 2002; Hartfield 2016).

114 In contrast to cloning, the main genetic consequence of selfing is genome
115 homogenization. Under complete selfing, parents with heterozygous genotypes *Aa* have a
116 50% probability of losing heterozygosity, independent of the allele frequencies in the rest of
117 the population. The probability that heterozygosity is retained decreases exponentially every
118 generation, rapidly leading to overall genome homogenisation. This homogenisation also
119 occurs, albeit at a lower rate, under partial selfing (Wright 1951). As a consequence, the
120 effective recombination rate is reduced since recombination creates variability only when
121 heterozygosity is present (Nordborg 2000). An additional consequence of increased
122 homozygosity is the exposure of recessive deleterious alleles to selection, giving rise to

123 inbreeding depression (Charlesworth and Willis 2009). An outcome of selfing broadly shared
124 with cloning is the removal of neutral variation through extensive linked selection acting
125 against deleterious mutations, reducing the effective population size (Agrawal and Hartfield
126 2016).

127 Both reproductive systems have been called 'dead-end' strategies, meaning that
128 species using them have an increased extinction probability. In asexual and selfing
129 populations, this phenomenon has often been ascribed to mutational meltdown, i.e., the
130 accumulation of deleterious mutations leading to extinction (Lynch et al. 1993, 1995; Olofsson
131 et al. 2023), but note that other mechanisms have been proposed for the phylogenetically
132 short-lived appearance of asexual species (Engelstädter 2008; Janko et al. 2008; Schwander
133 and Crespi 2009). While the 'dead-end' hypothesis has also been invoked for selfing species
134 (Wright et al. 2013), they can increase their fitness over time through purging deleterious
135 mutations (Glémin 2003; Abu Awad and Billiard 2017); this purging can also stop Muller's
136 ratchet from operating (Charlesworth et al. 1993). Furthermore, there exist ancient asexuals
137 that have seemingly existed for long periods of time in the absence of sexual reproduction,
138 including species of bdelloid rotifers (Flot et al. 2013), oribatid mites (Brandt et al. 2021),
139 ostracods (Tran Van et al. 2021), and trypanosomes (Weir et al. 2016). The persistence of
140 these ancient asexuals begs the question of whether there are underexplored mechanisms at
141 play that prevent these species from undergoing excessive accumulation of deleterious
142 variants, similar to what occurs in self-fertilising species.

143 Although genome exchange is an intrinsic part of sexual reproduction, gene conversion
144 is a mechanism of genome exchange that acts in both sexuals and asexuals in the form of
145 mitotic recombination (Lee et al. 2009). These gene conversion events can result in the
146 creation of homozygous tracts, either over localised regions or over large parts of
147 chromosomes depending on the underlying mechanism (Smukowski Heil 2023). In addition, it
148 is becoming increasingly known that many asexual species modify their meiosis cycle when
149 reproducing, which also results in increased runs of homozygosity (through, e.g., fusing
150 meiotic products; Blanc et al. 2025). This creation of homozygosity is similar to what happens

under selfing, and in asexual species it has been shown to have an important impact on neutral diversity when sex is rare (Hartfield et al. 2016; Hartfield 2021). Mitotic gene conversion has been argued to affect deleterious mutation accumulation in asexual species, including the bdelloid rotifer *Adineta vaga* (Flot et al. 2013; although see Vakhrusheva et al. 2020 who argue that these species show evidence of inter-individual gene exchange), the crustacean *Daphnia pulex* (Tucker et al. 2013), and the planarian worm *Schmidtea mediterranea* (Kershenbaum et al. 2024). Khakhlova and Bock (2006) also show experimental evidence of gene conversion eliminating deleterious mutations in plastids of tobacco plants, which are uniparentally inherited and are effectively asexual. Most of these studies argue that this gene conversion is beneficial for asexual species by removing deleterious mutations (an exception being Tucker et al. (2013) that argued that mitotic gene conversion reduces fitness by creating homozygous mutations). However, most of these arguments are verbal, and there exist little theoretical predictions to determine whether realistic levels of gene conversion can purge deleterious mutations, and hence maintain high fitnesses in asexual populations in an analogous manner to self-fertilisation. One exception is a model in Flot et al. (2013), showing how gene conversion slows down Muller's ratchet in asexual populations.

The overall goal of this study is to investigate the efficacy of mitotic gene conversion in facultative sexual and asexual species in purging deleterious mutations through creating homozygous regions, and compare its effects to self-fertilisation to determine to what extent these mechanisms of genetic homogenisation are similar. We will use two complementary approaches: first, we develop single-locus analytical models to quantify the frequency of deleterious mutations under facultative sex and gene conversion, and compare results to equivalent models under self-fertilisation. We will then use multi-locus simulations to determine how both reproductive mechanisms influence the genome-wide prevalence of deleterious mutations. Furthermore, we present simulation outputs in a way so that they can be compared with genome data, to enable empirical comparisons with the theories presented here.

Our principal questions are: (i) Under facultative sex, how does the frequency of deleterious variants (and hence also the population's mean fitness) scale with the prevalence of sex and gene conversion, both at individual loci and genome-wide? (ii) What values of sexual reproduction and gene conversion produce a level of mutation purging similar to a given level of self-fertilisation? (iii) Can realistic levels of mitotic gene conversion hence be as effective as self-fertilisation at purging deleterious mutations? The findings of this study will provide important quantification of a mechanism for maintaining the fitness of facultative sexuals and asexuals that has been often discussed but seldom investigated theoretically. It also provides insights as to which mode of uniparental reproduction might be favoured by evolution due to its effects on removing deleterious variation.

Single-Locus Mathematical Model

To gain some insight into how mitotic gene conversion affects the purging of deleterious mutations and how it compares to self-fertilisation, we investigate a single-locus model of deleterious allele frequencies in a deterministic diploid Wright-Fisher population subject to either facultative sex with sexual reproduction occurring at frequency σ (i.e., every generation, a fraction σ of offspring are generated sexually, and the remaining $1-\sigma$ are produced asexually), or self-fertilisation at rate S (i.e. a fraction S of reproductions are produced by selfing). We consider the frequency of a deleterious allele (denoted A) that arose from mutation from a wild-type allele a with probability μ; heterozygous mutant genotypes have fitness $1-hs$ and homozygotes $1-s$, for h the dominance coefficient and s the selection coefficient. Mitotic gene conversion occurs with frequency γ, which converts heterozygotes to either wild-type or derived homozygote with equal frequency. Note that gene conversion acts under both sexual and asexual reproduction; for completeness we will derive equations considering the effects of gene conversion acting in both facultative sexuals and self-fertilising species, although the greater interest will be the effects under infrequent sex.

We can track the prevalence of deleterious mutations by creating numerical recursions of how the frequency is affected by selection, reproduction, mutation and gene conversion in non-overlapping generations (following the methods of Agrawal and Hartfield 2016, Hartfield 2021; see Supplementary *Mathematica* file for the full recursion equations). Although these recursions calculate the change in genotype frequencies over time, we can convert these genotype frequencies to allele frequencies; specifically, if the frequency of the mutant allele is p_A, then the frequency of the mutant homozygote is $p_A^2 + Fp_A(1 - p_A)$ (with similar equations for the wild-type homozygote), and the heterozygote has frequency $2\,p_A\,(1 - p_A)\,(1 - F)$. Here, *F* denotes the reduction in homozygosity compared to Hardy-Weinberg equilibrium (Wright 1951), and shows to what extent either gene conversion under facultative sex or self-fertilisation affects genotype composition by reducing heterozygote frequencies. After making these transformations, we linearise the equations assuming $\mu, p_A \ll 1$ to obtain steady-state values of p_A and *F* (see Otto 2003 for a similar approach). Appendix 1 in the Supplementary Text outlines the derivations used to obtain analytical solutions.

After these algebraic calculations, we obtain the general equation for the deleterious allele frequency:

$$\hat{p}_A = \frac{\mu}{s(F+h-Fh)} \tag{1}$$

This is a known form for mutation-selection balance under non-random mating (Ohta and Cockerham 1974; Glémin 2003). The inbreeding coefficient *F* in Equation 1 differs depending on the form of uniparental reproduction. Under selfing:

$$\hat{F}_{self} = \frac{((1 - h\,s)\,S\,(1 - \gamma) + 2\gamma)}{(2 - (1 - (2 - h)\,s)\,S\,(1 - \gamma))} \tag{2}$$

While under facultative sex:

$$\hat{F}_{asex} = \frac{\gamma}{\gamma + (1-\gamma)(s(1-\sigma)+\sigma)} \tag{3}$$

We can use heuristic arguments to compare *F* in each case to understand which reproductive mode has the greatest effect on homozygosity. In the absence of gene conversion and selection, so we only focus on the effects of selfing, then $\hat{F}_{self} \approx S/(2-S)$ in

line with classic results on how selfing affects homozygosity (e.g., Nordborg and Donnelly 1997). Under facultative sex (Equation 3), positive $\hat{F}_{asex}$ is only formed when gene conversion is present; interestingly, dominance does not appear in Equation 3 and instead affects allele frequencies through the mutation-selection balance term (Equation 1). In the absence of selection and assuming that $\gamma << \sigma$ so the denominator approximates to σ, then $\hat{F}_{asex} \approx \frac{\gamma}{\sigma}$ (as also found by Hartfield 2021). By comparing these two approximations, we can show that the inbreeding values are equivalent if S = 2/(1+ϕ) for $\phi = \sigma/\gamma$ (Hartfield et al. 2016; Hartfield 2021). As sex is much more common than gene conversion under these assumptions (so ϕ >> 1) then we see that gene conversion acting under frequent sex is only equivalent to a small selfing rate. Note that this approximation for *S* will break down once gene conversion and sex act at the same rate. Specifically, gene conversion becomes more effective at making homozygous genotypes once sex becomes rarer due to the enforced pairing of diploid gene copies, which are subsequently maintained by asexual reproduction (Hartfield et al. 2016). Hence, the genetic outcome becomes more similar to that expected under higher levels of self-fertilisation.

If we assume that alleles act independently, and that selection and mutation parameters are the same for all sites, then we can use these equations to predict the total number of deleterious mutations and deleterious homozygotes across a genetic region, which we can then compare to simulation results. If there are *N* diploid individuals in a sample (so there are a maximum of 2*N* alleles per site) over a stretch of genome covering *L* sites, then the total number of deleterious mutations will equal $2NL\, p_A$, and the number of deleterious homozygotes will equal $2NL\, [p_A^2 + F p_A (1 - p_A)]$.

Figure 1 illustrates these derived equations for a single locus and different values of the rate of sex and mitotic gene conversion. If mutations are extremely recessive (*h* = 0.01, Figure 1a) then even small amounts of gene conversion can reduce the prevalence of deleterious mutations compared to outcrossing expectations, even for intermediate frequencies of sex. This purging effect is weakened as the dominance coefficient increases,

so that the frequency of additive alleles becomes closer to outcrossing expectations (Figure 1b, c). These results illustrate that mitotic gene conversion has the strongest effect on purging deleterious mutations if they are highly recessive, with the purging effect magnified as sex becomes rarer. Under self-fertilisation (Figures 1d–f), the same general results arise, but we note that only a small amount of selfing is needed to remove highly recessive deleterious mutations, reflecting its stronger effect on creating homozygosity. Mitotic gene conversion also only has a minor effect on allele frequencies (note that the contour lines are nearly vertical).

Plots of $\hat{F}$ (Figure 2a, b) show that while mitotic gene conversion under facultative sex leads to elevated homozygosity, the increase is mostly modest unless sex is rare and deleterious mutations have a weak selection effect. In contrast, under self-fertilisation then $\hat{F}$ values tend to be much higher (Figure 2c, d).

To test the accuracy of these approximate analytical solutions (Equations 1-3), we compared them to exact values obtained from numerical recursions over 50,000 generations, where the deleterious heterozygous had an initial frequency of 0.001 (Supplementary Figures S1 and S2, with code in Supplementary *Mathematica* file). We see that the two match over a wide range of parameters. One caveat of these models is that they do not consider drift, and hence processes like Muller's ratchet (Muller 1964; Haigh 1978) that can arise when sex is rare.

Multi-Locus Simulation Methods

Simulation Outline

While the single-locus model provides insight into how rare sex and gene conversion affect deleterious mutation frequencies, it does not show how gene conversion affects longer genome regions (e.g., through longer gene-conversion events purging mutations at multiple loci, and its effects on the resulting disequilibrium between them) and hence how effective it is in purging mutations across larger, linked stretches of the genome. Hence, we next use

282 simulations to compare how gene conversion leads to mutation removal in asexuals,
283 compared to selfing species. Note that we do not consider gene conversion in selfers, as the
284 single-locus results demonstrate that it only has a minor effect under this reproductive mode.

285 Simulations were written using SLiM v4.0.1 (Haller and Messer 2023), where we used
286 one of two types of simulation framework. We first used the Wright-Fisher framework
287 (hereafter 'WF model') when exploring population fitness and mutation counts for general
288 levels of uniparental reproduction, in the absence of mitotic gene conversion. By default in
289 these simulations, the population size is fixed and the probability of choosing parents are
290 proportional to their fitness. We next ran simulations using the non-Wright-Fisher framework
291 (hereafter 'non-WF model') when investigating the effects of mitotic gene conversion in fully
292 asexual populations. By default in non-WF models, the population size can change depending
293 on which parents reproduce or not. However, as explained below, we modify the non-WF
294 behaviour in SLiM so that it acts like a comparable WF model.

295 Simulated populations consist of 5,000 diploid individuals, each with a genome size of
296 L = 25 Mb that approximates the length of *Arabidopsis thaliana* chromosome 1 (The
297 Arabidopsis Genome Initiative 2000). Deleterious mutations can arise at all nucleotides, and
298 individuals do not carry mutations at the start of the simulations. We let populations evolve for
299 60,000 generations, which is sufficient for the population variance to reach a steady-state
300 (Supplementary Figure S3). After this time, 50 individuals are sampled and their genotypes
301 were saved in a VCF file format for downstream analysis. The mutation rate was set to
302 4×10^{-9}, which is on the same order as that inferred from *Arabidopsis thaliana* (Ossowski et al.
303 2010). Per-base-position recombination rate was set to 4×10^{-8}, reflecting estimates obtained
304 from crosses of *Arabidopsis thaliana* (Salome et al. 2012).

305 Mutations were deleterious and mutation stacking is allowed by default. Two types of
306 fitness models were investigated. They differ in whether selection and dominance coefficients
307 were constant or drawn from a distribution (Table 1). In the first type, fixed values of deleterious
308 mutations and dominance coefficients were chosen. In the second type, the selection and
309 dominance coefficients were drawn from a distribution based on Spigler et al. (2017) (see

Table 1). Selection coefficient is modelled by a gamma distribution with shape parameter α and mean $\bar{s}$. The dominance coefficient of a mutation with selection coefficient *s* is then drawn from a uniform distribution bounded by 0 and e^{-ks} , where *k* is a constant (see Table 1 for exact values). Free parameters in the model, *k*, $\bar{s}$, and α, were defined so *h* matches that used in main simulations (0.2) when $\bar{s}$ = 0.01 using a shape parameter α = 0.02 that reflects values inferred by Chen *et al.* (2017); note that this parameterisation produces $\bar{h} \approx 0.35$. This model reflects empirical observations that strongly deleterious mutations tend to be more recessive (Simmons and Crow 1977; Agrawal and Whitlock 2011).

Implementing mitotic gene conversion in asexual reproduction

By default, recombination in SLiM (including gene conversion) only acts exclusively when meiosis is involved. Therefore, the reproduction cycle was manually modified through a reproduction callback (i.e., a SLiM function for governing reproduction), so that mitotic gene conversion in asexuals can be simulated; this can only be done in non-WF model. For simplicity, the model applies only to a completely asexual population. The development of the code was facilitated by a post on the SLiM discussion board (Dussert and Haller 2019) and was extended based on the gene conversion model under SLiM Wright-Fisher simulations (Haller and Messer 2016).

When an offspring is generated, then the number of gene conversion events that acts within it is drawn from a Poisson distribution with a mean $L\gamma$ where γ denotes the rate of gene conversion initiating at a site. For each gene conversion event, the central position is first drawn uniformly through the chromosome. Then, the tract is extended to the left and right independently by drawing each length from a geometric distribution with parameter *p:*

$$p = \frac{-\lambda + \sqrt{\lambda}\sqrt{8+\lambda}}{2\lambda} \qquad (2)$$

335 where λ is the total mean tract length we want to use in simulations. This parameterization is
336 used to account for the fact that gene conversion tracts with length zero are discarded. This
337 redefined value of *p* means that the mean tract length is λ after accounting for discarding these
338 tracts (see Supplementary *Mathematica* notebook for further information). Gene conversion
339 events that overlap with the chromosome ends are truncated so they stop at the chromosome
340 edges.

341 After the gene conversion start- and end-points are drawn, they are checked and
342 potentially modified as follows: tracts of length 0 are removed; gene conversion tracts may
343 have the same start or end positions, in which case, the start (or end) of one of them is shifted
344 to the right (left), so that gene conversion tracts overlap; or two gene conversion tracts overlap,
345 in which case two interior gene conversion positions are removed so that the resulting gene
346 conversion tract is a union of the two smaller ones. Since multiple gene conversion tracts can
347 overlap, this refinement process is repeated until all start and end positions are unique and
348 alternate after sorting.

349 Gene conversion can occur on either of the homologous chromosomes with equal
350 probability and results in the selected part of one chromosome being copied onto the same
351 position at the other chromosome. Note that for each offspring-production event, all gene
352 conversion tracts are in the same direction, i.e., material is transferred solely from one strand
353 to another during each reproduction event. Since the 'initiation' strand is randomly chosen for
354 each reproduction event, then this effect should not lead to a major bias in results. The mean
355 length of a gene conversion event is set to 4,000 base pairs; this value accounts for the fact
356 that empirical studies of mitotic gene conversion tends to find that gene conversion tracts are
357 bimodally distributed, with a combination of short (on the order of 1kb) and long tracts (on the
358 order of 10kb) (Yim et al. 2014; Chumki et al. 2016; Jia et al. 2021). Using a mean length of
359 4,000bp will thus enable a wide distribution of tract lengths to be drawn that cover both these
360 possibilities. We report simulation results in terms of the 'mean gene conversion rate per site',
361 which is equal to the initiation rate times the mean tract length. This value is approximately
362 equivalent to the gene conversion parameter γ in the single-locus model (Frisse et al. 2001).

To enable fair comparison between simulations, further changes to the non-WF model were made so that it behaves as a standard Wright-Fisher model with a fixed population size over time (which is explained further in the SLiM Manual; Haller and Messer 2016). Specifically, a survival callback (i.e., a SLiM function for regulating individual mortality) limits the age of an individual to a single generation, and a reproduction callback samples a constant number of parents with probability weighted by their fitness. By comparing measurements of population fitness (Supplementary Figure S4) and mutation count (Supplementary Figure S5) between the two types of simulations, we see that this new routine yields similar behaviour under low gene conversion rates to the original WF model with complete asexuality.

100 replicate simulations were run if *s*, *h* were fixed; 20 replicate simulations were run for WF simulations (i.e., with partial selfing and cloning) where *s*, *h* are drawn from distributions; and 10 replicate simulations were run for non-WF simulations (i.e., with asexuality and gene conversion) where *s*, *h* are drawn from distributions. Fewer simulations replicates were used in the latter two cases due to their longer runtime.

Mutational Load Measurement

Although we can measure exact genetic load measurements from our simulations, in practice they are hard to measure from empirical data, as they usually require assumptions of the underlying parameters (such as selection and dominance coefficients) that have considerable uncertainty around them (Henn et al. 2016; Robinson et al. 2023; Kyriazis and Lohmueller 2024). Instead, empirical studies often use proxy measurements for load, specifically the total number of deleterious mutations, and those present as homozygotes (Marsden et al. 2015; Henn et al. 2016; Robinson et al. 2016; Laenen et al. 2018; Grossen et al. 2020; Hoffman et al. 2024). These measurements are simple ways to gauge deleterious mutation prevalence, and their approximate contributions to fitness reductions in a way that can be easily measured from genome data and compared across species with different reproductive modes in nature. For example, Laenen et al. (2018) used these measurements

389 in an analysis of *Arabis alpina* populations across Europe, to compare deleterious mutation
390 prevalence in populations with different degrees of inbreeding via self-fertilisation. Hence, we
391 present deleterious mutation prevalence from simulations in the same way here, to better
392 enable empirical comparisons with these theoretical results and to understand the relative
393 contributions of heterozygotes and homozygotes to deleterious mutation prevalence.

394 We calculated (i) the total number of mutations present in the population, and (ii) the
395 number of homozygous mutant genotypes. These measurements are proportional to the
396 population's genetic load, assuming a fixed selection coefficient, and the realised load for
397 recessive mutations with a dominance coefficient of zero (Bertorelle et al. 2022). We also
398 calculated the relative homozygosity for each simulation, defined as 2*(number of
399 homozygous mutations)/(total number of mutations), which can be interpreted as the
400 proportion of deleterious mutations that are in homozygous state, across all sites and
401 individuals. Simulation outputs were analysed using R 4.2.1 (R Core Team 2024).

402 Measuring linkage and identity disequilibrium

403 Linkage disequilibrium was measured from simulations using similar methods to those
404 in Hartfield et al. (2018) and Hartfield and Glémin (2024). Briefly, r^2 was measured using
405 VCFtools (Danecek et al. 2011) after thinning SNPs so measurements were taken between
406 sites that are 10kb apart and had a minor allele frequency of 5%. LD decay was plotted for
407 each simulation replicate in bins of 0.5Mb up to 12.5Mb apart, while subsampling the results
408 per bin so that they all contain the same number as in the lowest bin, but no more than 200
409 (in case of simulations with fixed fitness effects) or 100 (for simulations with variable fitness
410 effects). Only replicates with the minimum number of points per bin larger or equal to 5 were
411 retained.

412 Identity disequilibrium was measured using the g_2 statistic (David et al. 2007) as
413 implemented in the 'inbreedR' package (Stoffel et al. 2016). This statistic measures the extent
414 heterozygosity across a genome is correlated, relative to an expectation under independent

415 sites. For all simulations, the output VCF file was converted to inbreedR format as described
416 in the package's manual. The 'g2_snps' function was then used with 100 bootstraps for
417 generating the mean g_2 estimate along with 95% confidence intervals.

418 Results

419 Comparing self-fertilisation and facultative sex

420 We first compared the mean fitness and mutation counts under different levels of self-
421 fertilisation or asexual reproduction, to determine how mutation accumulation differs between
422 the two mating systems. These simulations were run using the WF model. All results in the
423 main text assume a fixed *s* and *h*; we will present results with varying *s*, *h* values at the end of
424 the results section.

425 Figure 3 shows how the mean fitness changes over time. Under self-fertilisation
426 (Figure 3a), while the mean fitness is fairly high overall, it is lowest when there is complete
427 outcrossing. It generally increases with the selfing rate, reaching a maximum for around 95%
428 self-fertilisation, but then drops for even higher rates. In contrast, the mean fitness drops
429 dramatically for highly asexual populations (Figure 3b).

430 Figure 4 (and Supplementary Figure S6) shows mutation counts under these
431 reproductive regimes. Selfing populations undergo mutation purging, as evidenced by a
432 decreasing number of mutations in the population (approximately linearly with the selfing rate)
433 and an increase in homozygous mutations reflecting higher inbreeding. In contrast, asexual
434 populations have approximately the same mutation counts (both total and homozygote) as
435 outcrossing populations, until the population becomes around 95% asexual. At this point, there
436 is a spike in homozygosity that is then reduced with even higher levels of asexuality, while the
437 total mutation count rapidly increases. The high mutation count reflects inefficient selection
438 acting once asexuality becomes prevalent, as reflected in the drop in mean fitness (Figure 3).
439 The spike in homozygosity likely reflects the fact that for high-but-incomplete asexuality,

440 mutation accumulation starts acting but there is still sufficient segregation to create
441 homozygous genotypes. In populations that become completely asexual (i.e., the level of
442 clonal reproduction approaches 1), there is a lack of genetic segregation so genotypes only
443 form as heterozygotes (the signature of 'allelic sequence divergence'). The relative
444 homozygosity (Supplementary Figure S7) also reflects these outcomes; it increases towards
445 1 with higher selfing, reflecting greater homozygosity due to inbreeding. In contrast, it stays
446 constant with increased asexuality up to 95% asexual reproduction, but then decreases with
447 higher rates. No apparent spike at 95% asexuality is observed using this measure.

448 Impact of mitotic gene conversion

449 Given this increased mutation count under low rates of sex, we next investigated to
450 what extent mitotic gene conversion can reduce it. Here, we used the non-WF model with the
451 new gene conversion routine implemented. Figure 5 plots the mean fitness for different per-
452 base-pair initiation rates of gene conversion. For most values the mean fitness remains low,
453 similar to results in the absence of gene conversion (Figure 3b). However, once mean gene
454 conversion per site becomes sufficiently high (in this case, $4x10^{-4}$ and above) the mean fitness
455 increases and starts reflecting that of outcrossing populations. These results imply that purging
456 is indeed occurring.

457 Purging is further validated when looking at mutation counts (Figure 6). Here, the
458 mutation count drops while the homozygosity increases (as reflected with heightened relative
459 homozygosity; Supplementary Figure S8), in line with purging reducing the total mutation
460 prevalence and increasing the mean fitness. We also compared our simulations to
461 expectations obtained by extending the single-locus results across multiple loci, assuming
462 they act independently. We see that this simple expectation overestimates both the total
463 number of deleterious mutations present, as well as the number of homozygotes. Some of this
464 discrepancy can be explained by the sample of 50 individuals not capturing all mutations.
465 However, the tailing-off of homozygous genotypes with high GC rates suggests that multi-

466 locus purging has reached a maximum level, resulting in lower mutation counts (both total and
467 homozygotes) than expected from single-locus expectations. We will discuss this discrepancy
468 further in the Discussion.

469 Comparing mitotic gene conversion to self-fertilisation

470 We next compare the effects of mitotic gene conversion in asexuals to self-fertilisation,
471 to determine to what extent the two effects are comparable. Figure 7 compares gene
472 conversion in asexuals against a range of selfing rates. When looking at homozygosity, it
473 seems that asexuality with the highest gene conversion rates ($4x10^{-2}$) yield the same
474 homozygous counts as complete selfing, implying that the two are equivalent. However, the
475 total mutation count is lower for high levels of self-fertilisation (80% and above), compared to
476 the highest gene conversion rate used. Hence, it seems that while gene conversion in
477 asexuals can purge mutations, it does not produce the same benefits in terms of reducing
478 deleterious mutation prevalence as observed under high self-fertilisation.
479 In Appendix 2 in the Supplementary Text (and Supplementary Figures S9–S12) we
480 show how facultative sex and self-fertilisation affect linkage and identity disequilibrium; these
481 results reflect previously-investigated properties of uniparental reproductive modes.
482

483 Exploring results under different parameters

484 Results with different population sizes and rescaled parameters

485 To test if these simulation results hold more generally, we next explored deleterious
486 mutation prevalence under different parameters. Under classic population-genetic models,
487 evolutionary outcomes are not just dependent on parameters such as the mutation rate and
488 selection coefficients, but on their products with the population size (Hill and Robertson 1966;
489 Kim and Wiehe 2009; Uricchio and Hernandez 2014; but see Dabi and Schrider 2025 for an
490 investigation into when such rescaling breaks down). It has been previously shown that the

effects of mitotic gene conversion on increased homozygosity is dependent on $N\gamma$ if the rate of gene conversion is sufficiently rare (Hartfield et al. 2016, 2018; Hartfield 2021). Hence, if this scaling influences mutation purging, then we should see lower mutation counts arising for smaller gene conversion values if other compound parameters are kept constant. We therefore re-ran simulations with different population sizes (N = 1,000 and 10,000 respectively) but with the mutation rate and selection coefficients scaled so that $N\mu$, Ns are the same as for N = 5,000 case.

Results for mutation counts are shown in Supplementary Figures S13, S14. It seems that the same general levels of gene conversion (i.e. $4\text{x}10^{-4}$ and above) lead to the largest drops in deleterious mutation count. However, for N = 10,000 greatly increased homozygosity is observed for $4\text{x}10^{-5}$, albeit with only a modest reduction in total mutation count. This result hints that purging does depend on the compound parameter $N\gamma$. Hence, it seems that the observed results for N = 5,000 still generally hold if other compound parameters ($N\mu$, Ns) are kept constant, but there are indications that gene conversion can be more effective in much larger populations due to more gene conversion events in a population (larger $N\gamma$).

Results where *s*, *h* are drawn from distributions

We also investigate simulations where s, h are drawn from probability distributions, using the same mutation rate as before. In these cases the fitness variance also reaches a steady-state within 60,000 generations (Supplementary Figure S15). Mean fitness results are generally similar to the fixed s, h case although, if looking just at selfing populations, the highest fitness is realised for complete self-fertilisation (Supplementary Figure S16). Mutation accumulation in the absence of gene conversion acts slightly differently; while the highest mutation count occurs under high levels of sexual reproduction, the number of homozygous genotypes is reduced under both high levels of selfing and asexual reproduction (Supplementary Figures S17, S18). The relative homozygosity is higher than that for fixed s, h cases under low rates of sex, but drastically drops once the rate of asexuality reaches 95%

and higher (Supplementary Figure S19). Gene conversion in asexuals restores population fitness in a similar manner to the fixed *s, h* simulations (Supplementary Figure S20), with slightly lower levels of gene conversion needed to purge deleterious mutations (Supplementary Figure S21 – S23). LD and ID measurements are also qualitatively similar to before (Supplementary Figure S24 – S27). Hence, it seems that the main results do not strongly depend on the fitness model used.

Discussion

Summary of results

In this study, we theoretically explored to what extent mitotic gene conversion in facultative sexual species purge deleterious mutations. We additionally compared this process to self-fertilisation, as the latter is known to purge deleterious mutations so we can compare how effective the two are. Using a single-locus model (Figure 1), we first demonstrated that mitotic gene conversion has the greatest effect in reducing mutation frequency when asexuality is frequent, but also when mutations are recessive. We also show that, while the process leads to elevated homozygosity (as measured by the inbreeding coefficient *F,* Figure 2), it will not be substantially elevated and match the effects of selfing unless populations are fully asexual and the gene conversion rate is high. We subsequently used multi-locus simulations to explore the effect of mitotic gene conversion acting on a genome-wide level. In the absence of gene conversion (Figures 3, 4), highly-asexual species accumulate many mutations as heterozygotes, leading to a drastic drop in fitness. When gene conversion is present in fully asexual populations (Figures 5, 6), then we do see purging: once the gene conversion rate exceeds a critical value (generally a mean rate of 4×10^{-4} per site) the total mutation count drops and homozygosity increases. These levels of gene conversion also dramatically increase the fitness of asexual populations (Figure 5, Figure S18). Despite these

542 benefits of gene conversion, the total mutation count is lowest under high levels of self-
543 fertilisation (Figure 7).

544 If single-locus models are used to predict multi-locus results assuming independent
545 loci, then we see that while they can be used to predict the qualitative outcomes of mutation
546 purging, they greatly overestimate the mutation load at high rates (Figure 6). One likely reason
547 for this discrepancy is that loci do not act independently in the simulations, due to linkage
548 between sites and the fact that gene conversion events affect multiple linked loci at once. Little
549 is known about the joint effects of inbreeding and linkage on mutation purging, but
550 Charlesworth et al. (1992) found that linkage has little effect on inbreeding depression caused
551 by deleterious mutations, except under tight recombination and complete self-fertilisation
552 where there was a noticeable reduction in inbreeding depression. A similar mechanism could
553 be acting here, where a combination of asexuality and multi-locus mitotic gene conversion
554 jointly act to greatly amplify purging of deleterious mutations.

555 Overall, both the single-locus and multi-locus results indicate that while high rates of
556 gene conversion can purge deleterious mutations under asexual reproduction, these benefits
557 do not always counter the costs of inefficient selection under clonality and self-fertilisation can
558 act a stronger force at purging deleterious mutations. That said, lower levels of gene
559 conversion (rate of 10^{-5} and above) can remove some deleterious mutations and increase the
560 fitness of asexual organisms, and hence still be evolutionary favoured.

561

562 Comparing mutation accumulation in self-fertilising and asexual 563 populations

564 One notable feature of the asexual-selfing comparison is the threshold behaviour of
565 mutation counts. In the absence of gene conversion, neutral genetic diversity in asexual
566 populations is similar to outcrossers unless the rate of sex is of the order $1/N$ (Bengtsson 2003;
567 Ceplitis 2003; Hartfield et al. 2016; Hartfield 2021); background selection also incurs

568 noticeable reductions in diversity in species with infrequent sex (Agrawal and Hartfield 2016).
569 We see similar results with regards to the mutation count in Figure 4; it stays fairly stable under
570 facultative sex unless populations become highly asexual. Conversely, under high selfing we
571 generally see a drop in the mutation count and heightened homozygosity, in line with mutation
572 purging. With very high levels of selfing, we see an elevation in both the homozygous mutation
573 count and in the total mutation number. These increases likely arise due to an additional
574 increase in mutation number due to genetic drift, as N_e can be greatly reduced in selfing
575 populations due to background selection (Kamran-Disfani and Agrawal 2014; Roze 2016).
576 Note that this behaviour can change with the fitness model assumptions; under a model where
577 *s* and *h* are drawn from distributions, we instead see a large reduction in the mutation count
578 under high selfing, in line with a model where genetic variation is purged (Charlesworth and
579 Charlesworth 1995; Lande and Porcher 2015; Abu Awad and Roze 2018).
580

581 Effects of mitotic gene conversion, and comparison with 582 empirical estimates

583 Throughout this study, we compared purging via mitotic gene conversion in asexuals
584 to that via self-fertilisation. We showed that despite the benefits of mitotic gene conversion,
585 lower mutation counts were observed under high levels of self-fertilisation (see, e.g., Figure
586 7). These results indicate that self-fertilisation can be a more efficient means of purging
587 deleterious mutations, likely because it affects the whole genome and homozygous regions
588 can be formed more rapidly. In contrast, mitotic gene conversion as employed here only
589 affects a fraction of the genome every generation, so purging acts in a piecemeal manner.
590 This comparison seems to hold over different mutation models, selfing and gene conversion
591 rates; one caveat is that there could be a specific, untested combination of mutation and
592 recombination rates in selfers that might lead to different results.

593 Our simulation model shows that a mean mitotic gene conversion rate above $4x10^{-4}$
594 per site can start facilitating the purging of deleterious mutations. This is in line with previous
595 theoretical work showing that, once gene conversion becomes sufficiently high, then it can
596 substantially reduce genetic variation in asexual populations (Hartfield et al. 2016; Hartfield
597 2021). Although empirical estimates are sparse, they are usually much lower than this value,
598 ranging between 10^{-7} and 10^{-5} depending on the study organism (Mandegar and Otto 2007;
599 Xu et al. 2011; Flot et al. 2013; Sharp and Agrawal 2016). Potentially higher rates were
600 observed in rice; Jia et al. (2021) inferred a mitotic recombination rate of 10^{-6} with mean tract
601 lengths ranging between 30-50kb, leading to a per-locus rate on the order of 10^{-2}. Hence, it
602 seems that while some larger empirical estimates are sufficiently high to cause extensive
603 deleterious mutation purging, on the whole mitotic gene conversion is usually too weak a force
604 to lead to extensive purging in nature. However, the estimates of Jia et al. (2021) consider all
605 mitotic recombination events, including crossover and non-crossover events. Our simulation
606 procedure only considers non-crossover events, which converts more localised genomic
607 regions. Mitotic crossover events, in contrast, can convert much larger fractions of the genome
608 (Jia et al. 2021). Even though the net conversion rate per site seems the most important
609 parameter, longer conversion regions could have a stronger effect through affecting multiple
610 sites at once, in a similar manner to self-fertilisation. Other forms of asexual reproduction that
611 also create extensive homozygous regions, including automixis (Mirzaghaderi and Hörandl
612 2016; Engelstädter 2017), could also affect enough of the genome to cause large levels of
613 mutation purging. Future research can determine whether these different factors have a
614 stronger effect on asexual fitness.

615 Caveats and perspective for future work

616 Another factor not considered in our study is the effect of small and/or changing
617 population sizes. In small populations, homozygotes can form more frequently due to genetic
618 drift (Malécot 1948), which can also lead to the purging of deleterious mutations. However,

619 this process is only effective at purging highly recessive mutations (Glémin 2003). Gene
620 conversion and drift also interact to affect the evolution of sex (Roze and Michod 2010). Given
621 the large diploid population size of 5,000 used in simulations, then drift should have a minimal
622 influence on purging in the multi-locus simulations, especially since the prevalence of
623 deleterious mutations did not strongly change under different population sizes (Supplementary
624 Figures S13, S14). However, the population size could affect the baseline level of deleterious
625 mutations in the absence of gene conversion. It will be a rewarding avenue of future research
626 to determine how drift would affect these processes. Similarly, we also did not consider
627 demographic feedbacks that are induced during the purging period, where mutation purging
628 also causes a reduction in population size (Abu Awad and Billiard 2017). The presence of
629 these feedbacks could increase the extinction probability of populations during gene-
630 conversion induced purging. Another avenue for further investigation is the role of adaptive
631 evolution and/or epistasis on the relative advantage of different reproduction strategies, as
632 they could impact the relative fitness of heterozygotes and homozygotes and hence the
633 outcomes of the reproductives modes studied here (Kondrashov and Kondrashov 2001; Abu
634 Awad and Roze 2020).

635 An interesting case where gene conversion may be important under clonal
636 reproduction is tumour evolution. Here, cells reproduce asexually by mitosis and can
637 potentially accumulate deleterious mutations due to a lack of recombination (Ní Leathlobhair
638 and Lenski 2022). Hence, the gene conversion mechanisms investigated here may prevent
639 transmissible cancers from experiencing mutation meltdown (Ní Leathlobhair and Lenski
640 2022), and also create homozygote loss-of-function tumor-suppressing genes that enable
641 carcinogenesis (Takahashi and Innan 2022).

Table 1. Summary of model parameters and values used for simulations in the main text. Note that in the Supplementary Material, we show results with different population sizes (*N* equalling 1,000 and 10,000) but with the mutation rate and selection coefficients scaled so $N\mu$, Ns are the same as for N = 5,000 simulations.

Parameter	Values for simulations with f. e.*	
	constant	non-constant
Genome size	Diploid, L = 25 Mbp	
Per-base mutation rate μ	4×10^{-9}	
Per-position recombination rate *r*	4×10^{-8}	
Population size *N*	5 000	
Sample size	50	
Selection	s = 0.01	s ~ Gamma($\bar{s}$, α) mean $\bar{s}$ = 0.01 shape α = 0.2
Dominance	h = 0.2	h ~ U(0, e^{-ks}) k = 91.63
Mean gene conversion tract length	λ = 4 000 bp	

* f. e. - fitness effects; selection and dominance coefficients

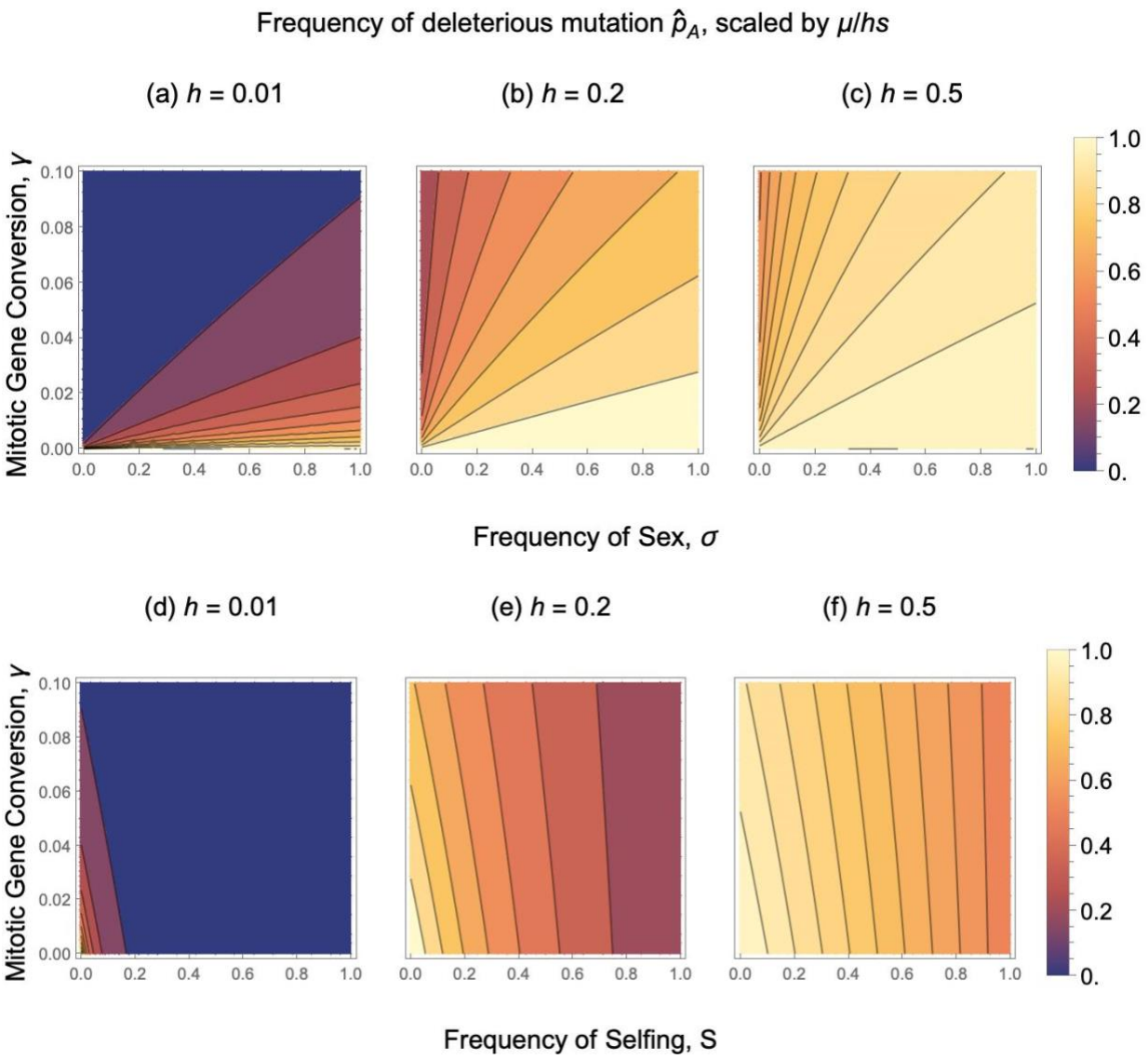


Figure 1: Plots of single-locus analytical results for the steady-state frequency of a deleterious mutation. *Results show Equation 1 for different dominance values, scaled to the expected result under outcrossing (μ/hs). Note that μ is not present in this scaled equation. Panels (a) – (c) are for different frequencies of sex (with $\hat{F}$ given by Equation 3), while (d) – (f) are for different frequencies of self-fertilisation (with $\hat{F}$ given by Equation 2).*

Alt text: *Contour plots showing deleterious mutation frequency with subfigures labelled a to f, illustrating effects of reproductive modes and mitotic gene conversion.*

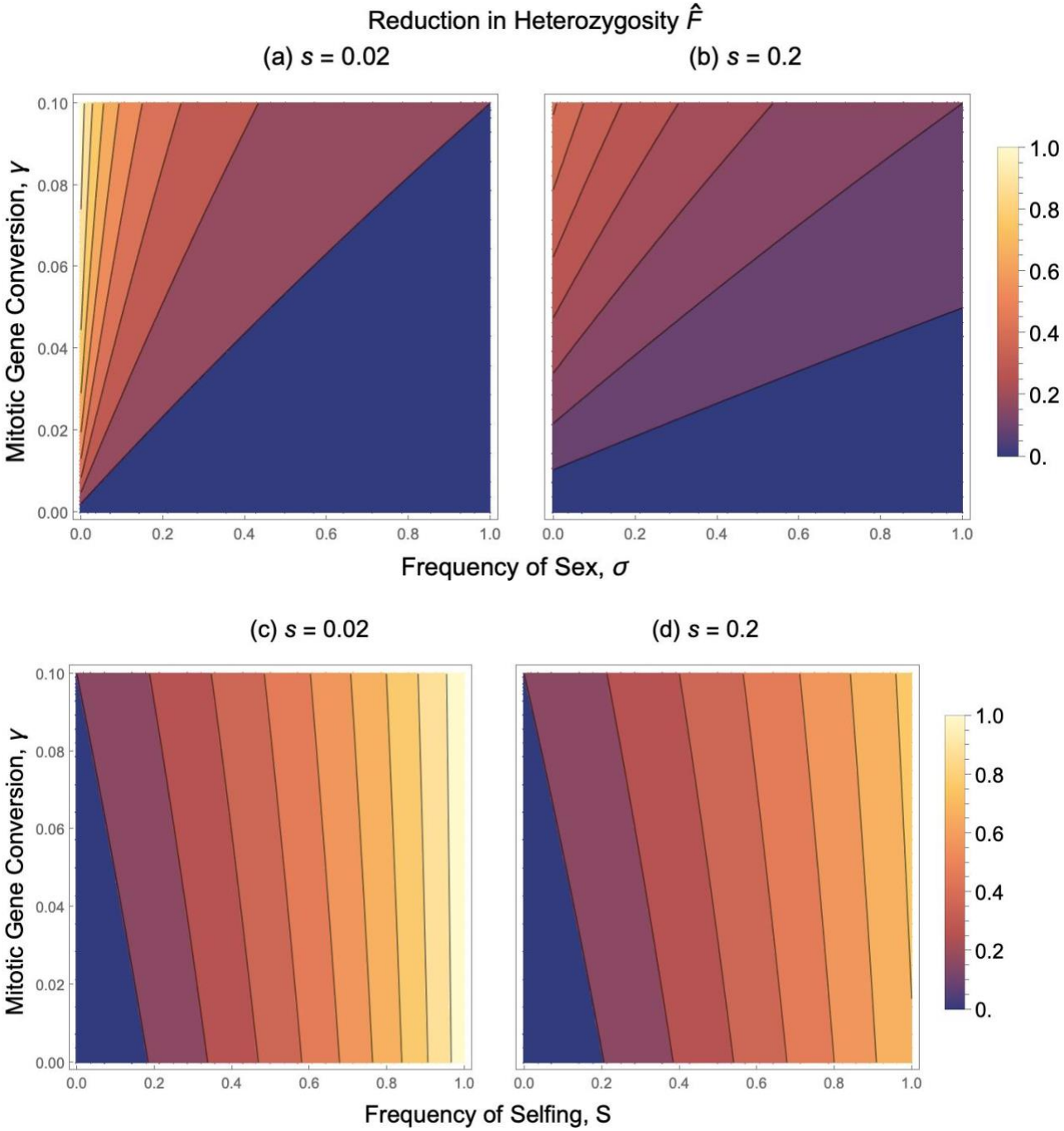


Figure 2: Plots of single-locus analytical results for the reduction in heterozygosity, $\hat{F}$. *Results show either Equation 2 under facultative sex (panels a, b), or Equation 3 under self-fertilisation (panels c, d).*

Alt text: *Contour plots showing reduction in heterozygosity with subfigures labelled a to d, illustrating effects of reproductive modes and mitotic gene conversion.*

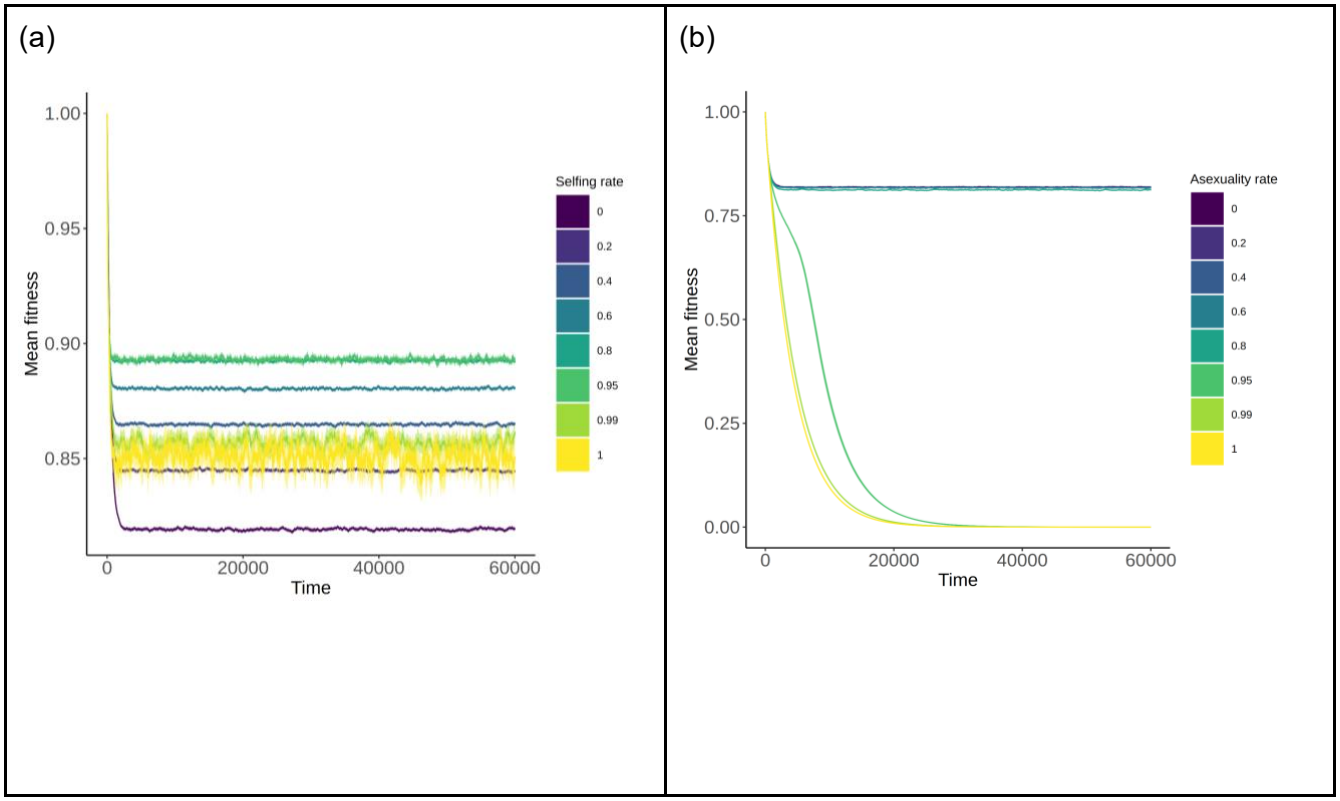
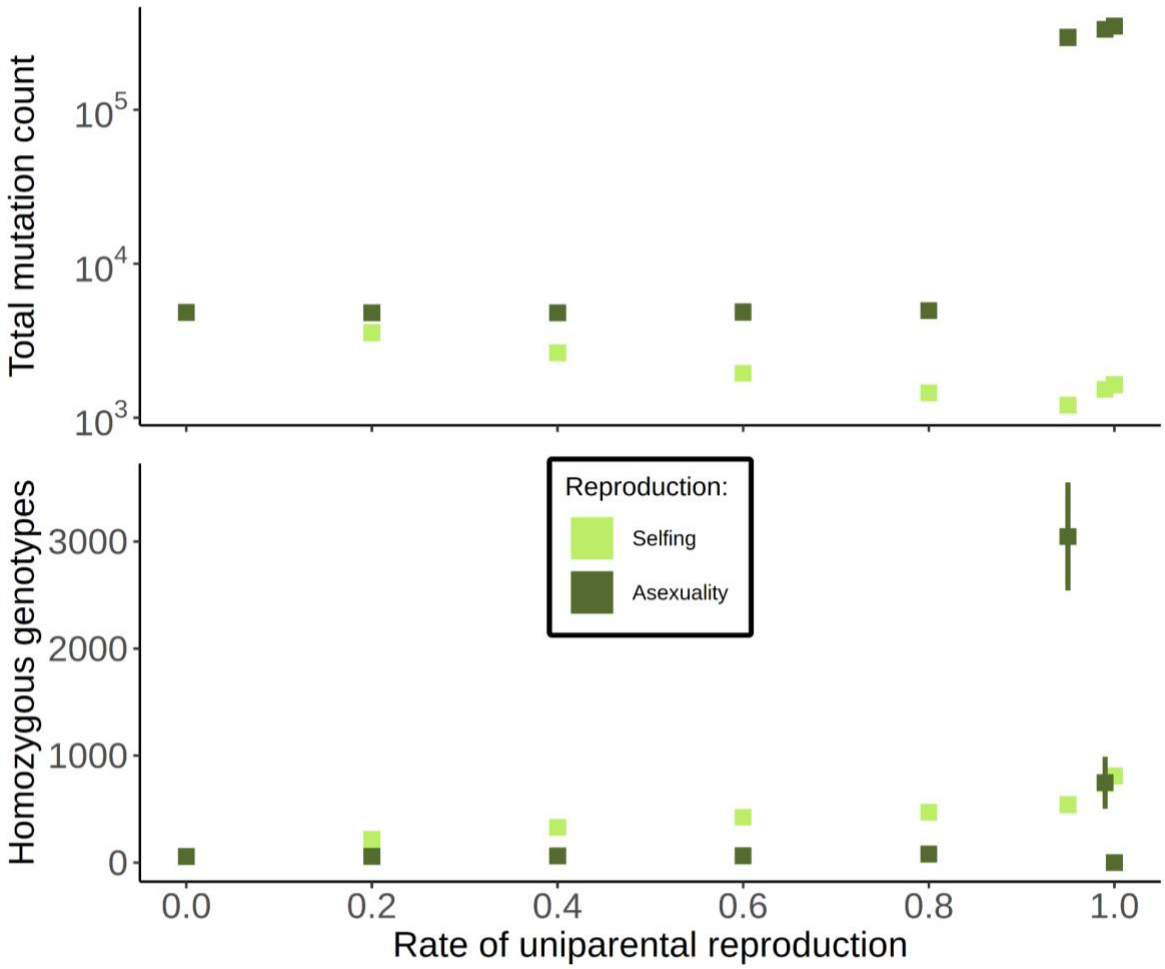


Figure 3: Mean fitness for different modes of uniparental reproduction. *Results are shown as a function of time, for (a) different rates of self-fertilisation or (b) asexual reproduction. Thick lines are mean values, while thinner shading around lines show 95% confidence intervals. Notice the change of scale on the y-axis between (a) and (b).*

Alt text: *Graphs showing the mean fitness over time for either self-fertilisation or partial asexual reproduction, with different colour lines showing different rates.*



676

677 ***Figure 4: Deleterious mutation counts under different uniparental reproductive modes.***
678 *As a function of the rate of uniparental reproduction (either selfing or asexual reproduction),*
679 *these plots show (top) the total mutation count present in the sample of 50 individuals, and*
680 *(bottom) the number of homozygous genotypes present in the sample. Error bars represent*
681 *95% confidence intervals; if they are not shown then they lie within a point. See Supplementary*
682 *Figure S6 for the total count on a linear scale.*

683 ***Alt text:*** *Graphs showing total mutation count and number of homozygote genotypes, with*
684 *different points indicating different rates of either self-fertilisation or partial asexual*
685 *reproduction.*

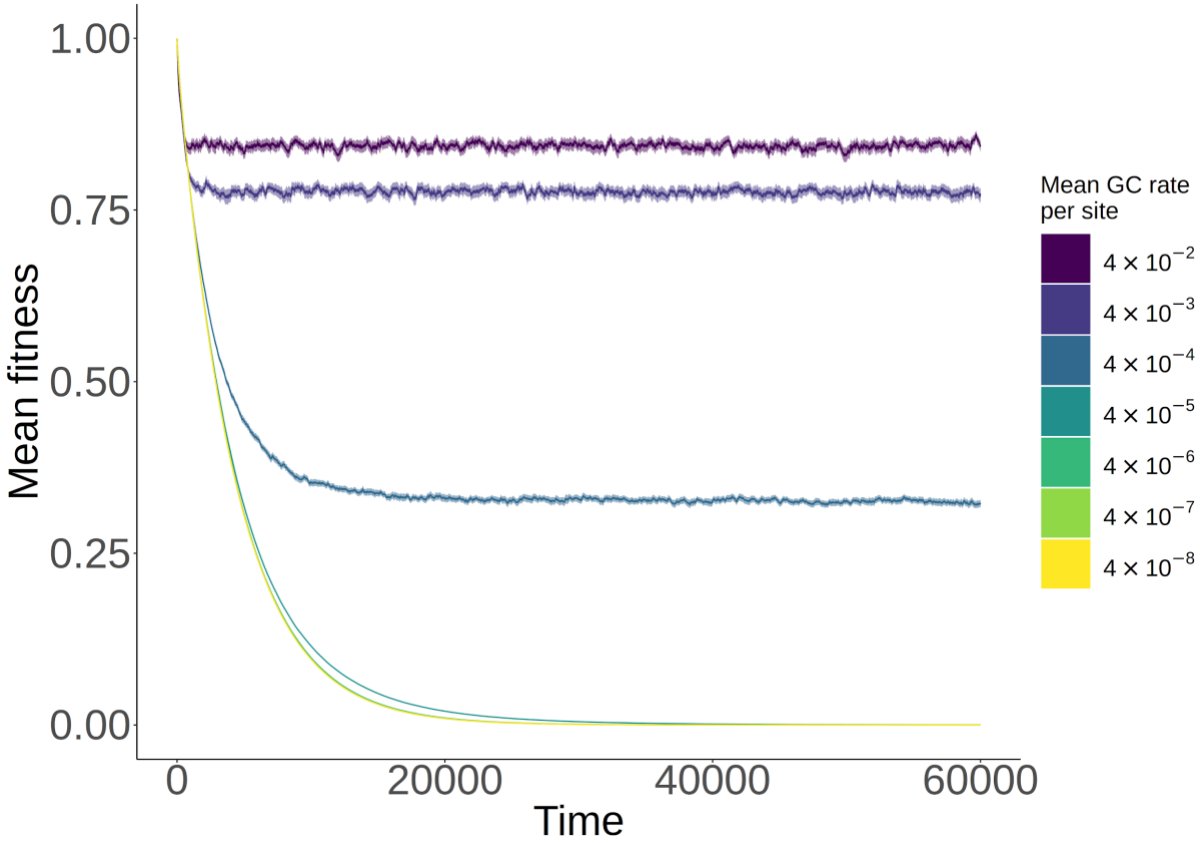
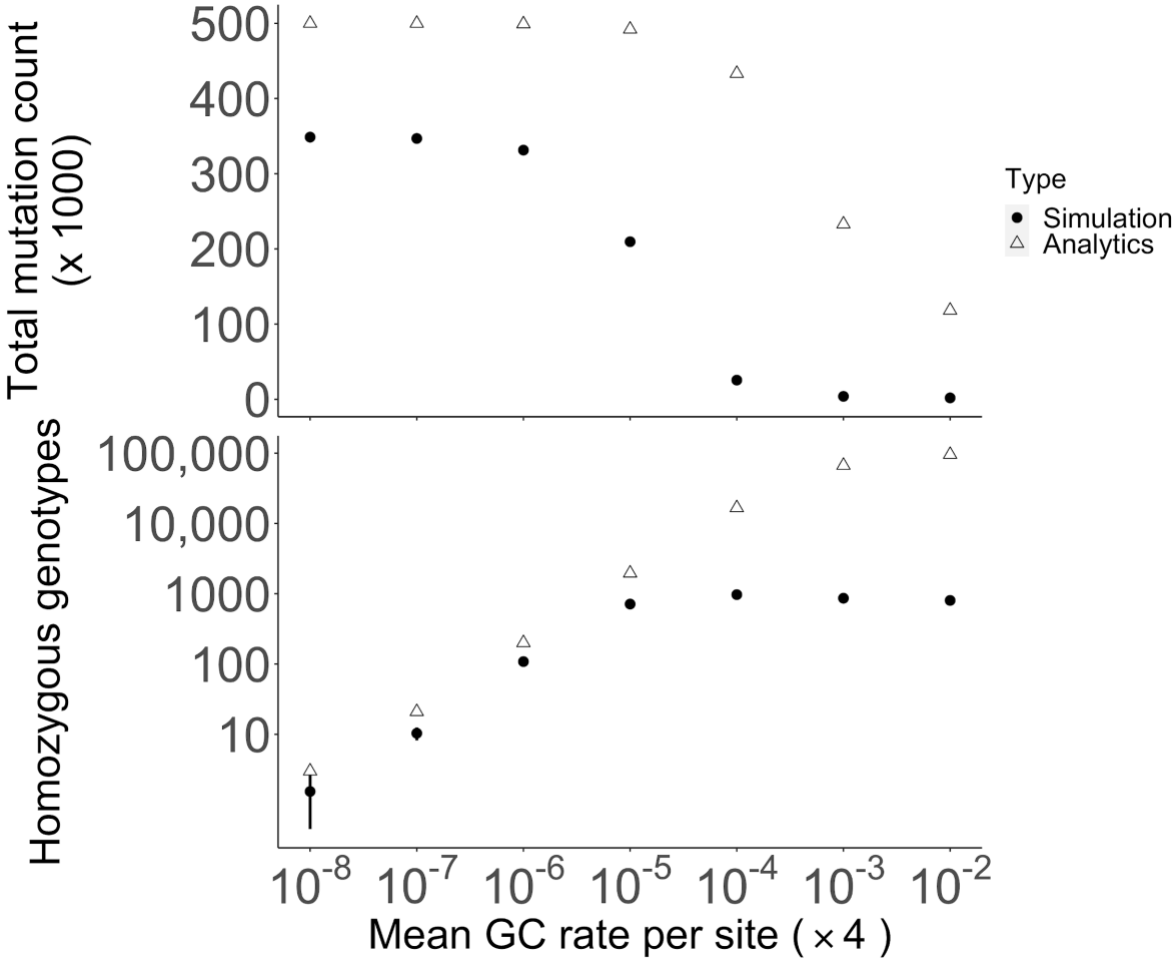


Figure 5: Mean fitness for obligately asexual populations when mitotic gene conversion ***is present at different rates.*** *Results are shown as a function of time. Thick lines are mean values, while thinner shading around lines show 95% confidence intervals.*

Alt text: *Graph showing the mean fitness over time for complete asexual reproduction, with different colour lines showing different rates of mitotic gene conversion.*



692

693 ***Figure 6: Deleterious mutation counts for obligate asexual populations with mitotic***
694 ***gene conversion present.*** *Results are shown for both simulations and analytic expectations*
695 *assuming no interactions between loci, as a function of the rate of gene conversion initiation*
696 *at a single site. Plots show (top) the total mutation count present in the sample of 50*
697 *individuals, and (bottom) the number of homozygous genotypes present in the sample. Error*
698 *bars for simulation results represent 95% confidence intervals; if they are not shown then they*
699 *lie within a point.*

700 ***Alt text:*** *Graph showing total mutation count and number of homozygote genotypes from both*
701 *simulations and analytical expectations, with different points indicating different rates of gene*
702 *conversion under complete asexual reproduction.*

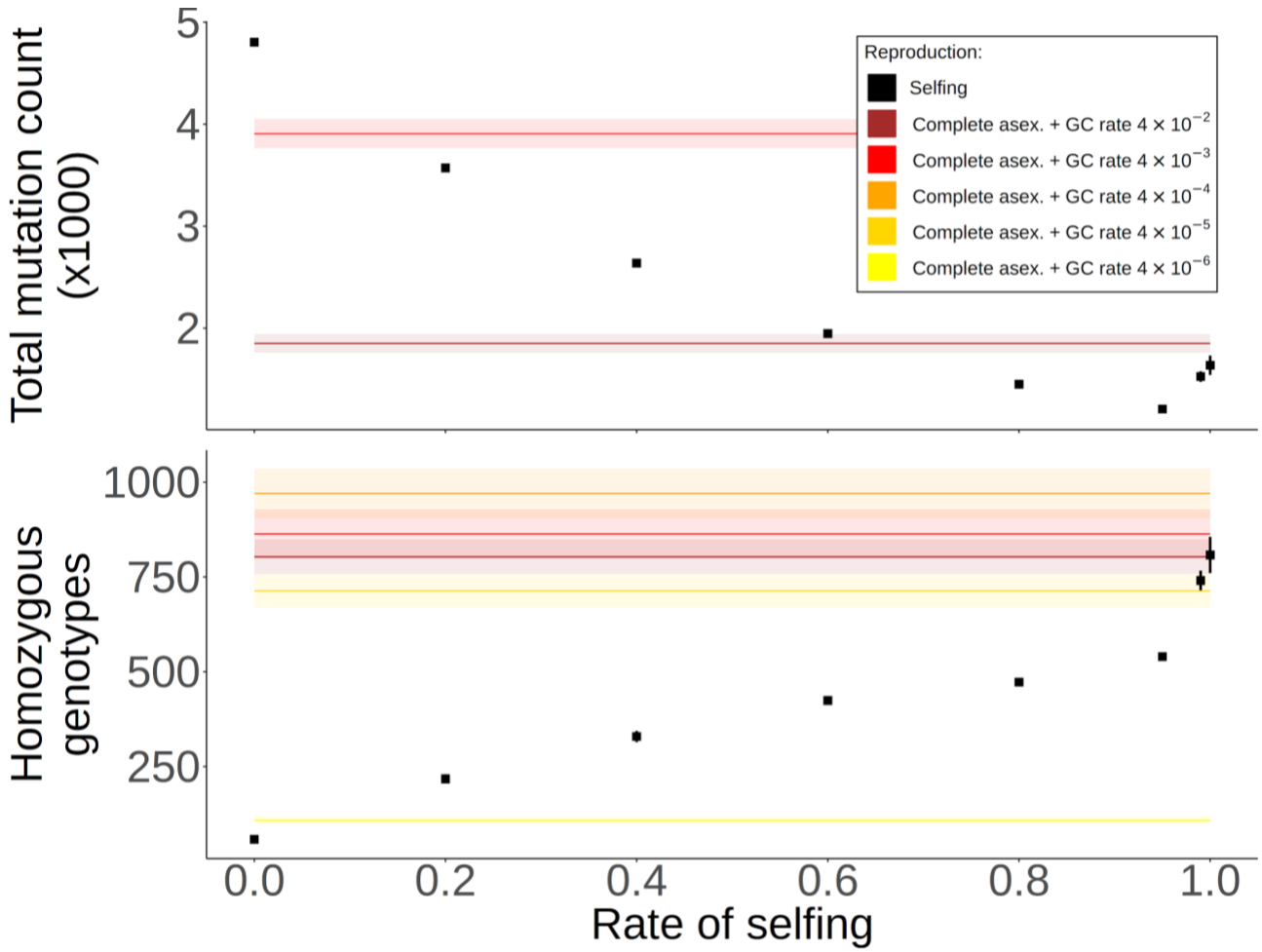


Figure 7: Comparing mutation counts under different levels of self-fertilisation to asexual populations subject to mitotic gene conversion***.*** *Plots show (top) the total mutation count present in the sample of 50 individuals, and (bottom) the number of homozygous genotypes present in the sample. Note that only* gene conversion *rates of* $4x10^{-2}$ *and* $4x10^{-3}$ *are shown in the top panel. The points show results under different levels of self-fertilisation; error bars represent 95% confidence intervals, and if they are not shown then they lie within a point. Coloured lines are results for asexuality with different levels of mitotic* gene conversion*, as indicated by the legend; shaded bands around lines indicate 95% confidence intervals.*

Alt text: *Graph comparing total mutation count and number of homozygote genotypes, with points indicating different selfing rates, and horizontal lines are results for complete asexuality with different gene conversion rates.*

References

Abu Awad, D., and S. Billiard. 2017. The double edged sword: The demographic consequences of the evolution of self-fertilization. Evolution 71:1178–1190.

Abu Awad, D., and D. Roze. 2018. Effects of partial selfing on the equilibrium genetic variance, mutation load, and inbreeding depression under stabilizing selection. Evolution 72:751–769.

Abu Awad, D., and D. Roze. 2020. Epistasis, inbreeding depression, and the evolution of self-fertilization. Evolution 74:1301–1320.

Agrawal, A. F., and M. Hartfield. 2016. Coalescence with Background and Balancing Selection in Systems with Bi- and Uniparental Reproduction: Contrasting Partial Asexuality and Selfing. Genetics 202:313–326.

Agrawal, A. F., and M. C. Whitlock. 2011. Inferences About the Distribution of Dominance Drawn From Yeast Gene Knockout Data. Genetics 187:553–566.

Baker, H. G. 1955. Self-Compatibility and Establishment After ‘Long-Distance’ Dispersal. Evolution 9:347–349.

Baker, H. G. 1967. Support for Baker’s Law–As a Rule. Evolution 21:853–856.

Bengtsson, B. O. 2003. Genetic variation in organisms with sexual and asexual reproduction. J. Evol. Biol. 16:189–199.

Bertorelle, G., F. Raffini, M. Bosse, C. Bortoluzzi, A. Iannucci, E. Trucchi, H. E. Morales, and C. Van Oosterhout. 2022. Genetic load: genomic estimates and applications in non-model animals. Nat Rev Genet 23:492–503.

Blanc, C. S., M. Delattre, and K. Van Doninck. 2025. Cracking the meiotic secrets behind animals asexuality: when cytology seduces genomics to decode asexual meiosis. J. Evol. Biol. 38:832–850.

Brandt, A., P. Tran Van, C. Bluhm, Y. Anselmetti, Z. Dumas, E. Figuet, C. M. François, N. Galtier, B. Heimburger, K. S. Jaron, M. Labédan, M. Maraun, D. J. Parker, M. Robinson-Rechavi, I. Schaefer, P. Simion, S. Scheu, T. Schwander, and J. Bast.

742 2021. Haplotype divergence supports long-term asexuality in the oribatid mite
743 *Oppiella nova*. Proc. Natl. Acad. Sci. U.S.A. 118:e2101485118.
744 Butlin, R. 2002. The costs and benefits of sex: new insights from old asexual lineages. Nat.
745 Rev. Genet. 3:311–317.
746 Ceplitis, A. 2003. Coalescence times and the Meselson effect in asexual eukaryotes. Genet.
747 Res. 82:183–190.
748 Charlesworth, D., and B. Charlesworth. 1995. Quantitative Genetics in Plants: The Effect of
749 the Breeding System on Genetic Variability. Evolution 49:911–920.
750 Charlesworth, D., M. T. Morgan, and B. Charlesworth. 1993. Mutation accumulation in finite
751 outbreeding and inbreeding populations. Genet. Res. 61:39–56.
752 Charlesworth, D., M. T. Morgan, and B. Charlesworth. 1992. The effect of linkage and
753 population size on inbreeding depression due to mutational load. Genet. Res. 59:49–
754 61.
755 Charlesworth, D., and J. H. Willis. 2009. The genetics of inbreeding depression. Nat. Rev.
756 Genet. 10:783–796.
757 Chen, J., S. Glémin, and M. Lascoux. 2017. Genetic Diversity and the Efficacy of Purifying
758 Selection across Plant and Animal Species. Mol. Biol. Evol. 34:1417–1428.
759 Chumki, S. A., M. K. Dunn, T. F. Coates, J. D. Mishler, E. M. Younkin, and A. M. Casper.
760 2016. Remarkably Long-Tract Gene Conversion Induced by Fragile Site Instability in
761 *Saccharomyces cerevisiae*. Genetics 204:115–128.
762 Dabi, A., and D. R. Schrider. 2025. Population size rescaling significantly biases outcomes
763 of forward-in-time population genetic simulations. Genetics 229:iyae180.
764 Danecek, P., A. Auton, G. Abecasis, C. A. Albers, E. Banks, M. A. DePristo, R. E.
765 Handsaker, G. Lunter, G. T. Marth, S. T. Sherry, G. McVean, and R. Durbin. 2011.
766 The variant call format and VCFtools. Bioinformatics 27:2156–2158.
767 David, P., B. Pujol, F. Viard, V. Castella, and J. Goudet. 2007. Reliable selfing rate estimates
768 from imperfect population genetic data. Molecular Ecology 16:2474–2487.
769 De Meeûs, T., F. Prugnolle, and P. Agnew. 2007. Asexual reproduction: Genetics and

770 evolutionary aspects. Cell. Mol. Life Sci. 64:1355–1372.

771 Dussert, Y., and B. C. Haller. 2019. Mitotic recombination in clonal population. URL:
772 https://groups.google.com/g/slim-discuss/c/5bcUrRLA1cQ/m/kHg3nk4WAgAJ.
773 Accessed July 2024.

774 Engelstädter, J. 2017. Asexual but Not Clonal: Evolutionary Processes in Automictic
775 Populations. Genetics 206:993–1009.

776 Engelstädter, J. 2008. Constraints on the evolution of asexual reproduction. BioEssays
777 30:1138–1150.

778 Eyre-Walker, A., and P. D. Keightley. 2007. The distribution of fitness effects of new
779 mutations. Nat. Rev. Genet. 8:610–618.

780 Felsenstein, J. 1974. The Evolutionary Advantage of Recombination. Genetics 78:737–756.

781 Flot, J.-F., B. Hespeels, X. Li, B. Noel, I. Arkhipova, E. G. J. Danchin, A. Hejnol, B.
782 Henrissat, R. Koszul, J.-M. Aury, V. Barbe, R.-M. Barthelemy, J. Bast, G. A. Bazykin,
783 O. Chabrol, A. Couloux, M. Da Rocha, C. Da Silva, E. Gladyshev, P. Gouret, O.
784 Hallatschek, B. Hecox-Lea, K. Labadie, B. Lejeune, O. Piskurek, J. Poulain, F.
785 Rodriguez, J. F. Ryan, O. A. Vakhrusheva, E. Wajnberg, B. Wirth, I. Yushenova, M.
786 Kellis, A. S. Kondrashov, D. B. Mark Welch, P. Pontarotti, J. Weissenbach, P.
787 Wincker, O. Jaillon, and K. Van Doninck. 2013. Genomic evidence for ameiotic
788 evolution in the bdelloid rotifer *Adineta vaga*. Nature 500:453–457.

789 Frisse, L., R. R. Hudson, A. Bartoszewicz, J. D. Wall, J. Donfack, and A. Di Rienzo. 2001.
790 Gene Conversion and Different Population Histories May Explain the Contrast
791 between Polymorphism and Linkage Disequilibrium Levels. Am. J. Hum. Genet.
792 69:831–843.

793 Glémin, S. 2003. How are deleterious mutations purged? Drift versus nonrandom mating.
794 Evolution 57:2678–2687.

795 Grossen, C., F. Guillaume, L. F. Keller, and D. Croll. 2020. Purging of highly deleterious
796 mutations through severe bottlenecks in Alpine ibex. Nat Commun 11: 1001.

797 Haigh, J. 1978. The accumulation of deleterious genes in a population–Muller's Ratchet.

Theor. Popul. Biol. 14:251–267.

Haller, B. C., and P. W. Messer. 2023. SLiM 4: Multispecies Eco-Evolutionary Modeling. The American Naturalist 201:E127–E139.

Haller, B. C., and P. W. Messer. 2016. SLiM: An Evolutionary Simulation Framework. URL: http://benhaller.com/slim/SLiM_Manual.pdf. Accessed July 2024.

Hartfield, M. 2021. Approximating the Coalescent Under Facultative Sex. Journal of Heredity 112:145–154.

Hartfield, M. 2016. Evolutionary genetic consequences of facultative sex and outcrossing. J. Evol. Biol. 29:5–22.

Hartfield, M., and S. Glémin. 2024. Polygenic selection to a changing optimum under self–fertilisation. PLoS Genet 20:e1011312.

Hartfield, M., and P. D. Keightley. 2012. Current hypotheses for the evolution of sex and recombination. Integr. Zool. 7:192–209.

Hartfield, M., S. I. Wright, and A. F. Agrawal. 2018. Coalescence and Linkage Disequilibrium in Facultatively Sexual Diploids. Genetics 210:683–701.

Hartfield, M., S. I. Wright, and A. F. Agrawal. 2016. Coalescent Times and Patterns of Genetic Diversity in Species with Facultative Sex: Effects of Gene Conversion, Population Structure, and Heterogeneity. Genetics 202:297–312.

Henn, B. M., L. R. Botigué, S. Peischl, I. Dupanloup, M. Lipatov, B. K. Maples, A. R. Martin, S. Musharoff, H. Cann, M. P. Snyder, L. Excoffier, J. M. Kidd, and C. D. Bustamante. 2016. Distance from sub-Saharan Africa predicts mutational load in diverse human genomes. Proc. Natl. Acad. Sci. USA 113:E440–E449.

Hill, W. G., and A. Robertson. 1966. The effect of linkage on limits to artificial selection. Genet. Res. 8:269–294.

Hoffman, J. I., D. L. J. Vendrami, K. Hench, R. S. Chen, M. A. Stoffel, M. Kardos, W. Amos, J. Kalinowski, D. Rickert, K. Köhrer, T. Wachtmeister, M. E. Goebel, C. A. Bonin, F. M. D. Gulland, and K. K. Dasmahapatra. 2024. Genomic and fitness consequences of a near-extinction event in the northern elephant seal. Nat Ecol Evol 8:2309–2324.

826 Janko, K., P. Drozd, J. Flegr, and J. R. Pannell. 2008. Clonal Turnover Versus Clonal Decay:
827 A Null Model for Observed Patterns of Asexual Longevity, Diversity and Distribution.
828 Evolution 62:1264–1270.
829 Jia, X., Q. Zhang, M. Jiang, J. Huang, L. Yu, M. B. Traw, D. Tian, L. D. Hurst, and S. Yang.
830 2021. Mitotic gene conversion can be as important as meiotic conversion in driving
831 genetic variability in plants and other species without early germline segregation.
832 PLoS Biol 19:e3001164.
833 Kamran-Disfani, A., and A. F. Agrawal. 2014. Selfing, adaptation and background selection
834 in finite populations. J. Evol. Biol. 27:1360–1371.
835 Kershenbaum, S., M. J. Espejo, A. Griffin, and A. Aboobaker. 2024. Gene conversion limits
836 the cost of asexuality in somatically immortal worms. bioRxiv, doi:
837 10.1101/2023.03.20.533553.
838 Khakhlova, O., and R. Bock. 2006. Elimination of deleterious mutations in plastid genomes
839 by gene conversion. Plant J. 46:85–94.
840 Kim, Y., and T. Wiehe. 2009. Simulation of DNA sequence evolution under models of recent
841 directional selection. Briefings Bioinf. 10:84–96.
842 Kondrashov, F. A., and A. S. Kondrashov. 2001. Multidimensional epistasis and the
843 disadvantage of sex. Proc. Natl. Acad. Sci. USA 98:12089–12092.
844 Kyriazis, C. C., and K. E. Lohmueller. 2024. Constraining models of dominance for
845 nonsynonymous mutations in the human genome. PLoS Genet 20:e1011198.
846 Laenen, B., A. Tedder, M. D. Nowak, P. Toräng, J. Wunder, S. Wötzel, K. A. Steige, Y.
847 Kourmpetis, T. Odong, A. D. Drouzas, M. C. A. M. Bink, J. \AAgren, G. Coupland,
848 and T. Slotte. 2018. Demography and mating system shape the genome-wide impact
849 of purifying selection in *Arabis alpina*. Proc. Natl. Acad. Sci. USA 115:816–821.
850 Lande, R., and E. Porcher. 2015. Maintenance of Quantitative Genetic Variance Under
851 Partial Self-Fertilization, with Implications for Evolution of Selfing. Genetics 200:891–
852 906.
853 Lee, P. S., P. W. Greenwell, M. Dominska, M. Gawel, M. Hamilton, and T. D. Petes. 2009. A

854 Fine-Structure Map of Spontaneous Mitotic Crossovers in the Yeast Saccharomyces
855 cerevisiae. PLoS Genet 5:e1000410.
856 Loewe, L., and W. G. Hill. 2010. The population genetics of mutations: good, bad and
857 indifferent. Philos. Trans. R. Soc. Lond. B Biol. Sci. 365:1153–1167.
858 Lynch, M., R. Burger, D. Butcher, and W. Gabriel. 1993. The Mutational Meltdown in
859 Asexual Populations. J. Hered. 84:339–344.
860 Lynch, M., J. Conery, and R. Bürger. 1995. Mutational Meltdowns In Sexual Populations.
861 Evolution 49:1067–1080.
862 Malécot, G. 1948. The mathematics of heredity.
863 Mandegar, M. A., and S. P. Otto. 2007. Mitotic recombination counteracts the benefits of
864 genetic segregation. Proc. R. Soc. B 274:1301–1307.
865 Mark Welch, D. B., and M. Meselson. 2000. Evidence for the Evolution of Bdelloid Rotifers
866 Without Sexual Reproduction or Genetic Exchange. Science 288:1211–1215.
867 Marsden, C. D., D. Ortega-Del Vecchyo, D. P. O'Brien, J. F. Taylor, O. Ramirez, C. Vilà, T.
868 Marques-Bonet, R. D. Schnabel, R. K. Wayne, and K. E. Lohmueller. 2015.
869 Bottlenecks and selective sweeps during domestication have increased deleterious
870 genetic variation in dogs. Proc. Natl. Acad. Sci. USA.
871 Mirzaghaderi, G., and E. Hörandl. 2016. The evolution of meiotic sex and its alternatives.
872 Proc. R. Soc. B 283.
873 Mráz, P., and V. Mrázová. 2021. Greater reproductive assurance of asexual plant compared
874 with sexual relative in a low-density sympatric population: Experimental evidence for
875 pollen limitation. J of Evolutionary Biology 34:1503–1509.
876 Muller, H. J. 1964. The relation of recombination to mutational advance. Mutat. Res. 1:2–9.
877 Nettancourt, D. de. 2001. Incompatibility and incongruity in wild and cultivated plants. Vol. 3.
878 Springer Science & Business Media, 2001.
879 Ní Leathlobhair, M., and R. E. Lenski. 2022. Population genetics of clonally transmissible
880 cancers. Nat Ecol Evol 6:1077–1089.
881 Nordborg, M. 2000. Linkage Disequilibrium, Gene Trees and Selfing: An Ancestral

882 Recombination Graph With Partial Self-Fertilization. Genetics 154:923–929.
883 Nordborg, M., and P. Donnelly. 1997. The Coalescent Process With Selfing. Genetics
884 146:1185–1195.
885 Ohta, T., and C. C. Cockerham. 1974. Detrimental genes with partial selfing and effects on a
886 neutral locus. Genet. Res. 23:191–200.
887 Olofsson, P., L. Chipkin, R. C. Daileda, and R. B. R. Azevedo. 2023. Mutational meltdown in
888 asexual populations doomed to extinction. J. Math. Biol. 87:88.
889 Ossowski, S., K. Schneeberger, J. I. Lucas-Lledo, N. Warthmann, R. M. Clark, R. G. Shaw,
890 D. Weigel, and M. Lynch. 2010. The Rate and Molecular Spectrum of Spontaneous
891 Mutations in *Arabidopsis thaliana*. Science 327:92–94.
892 Otto, S. P. 2003. The Advantages of Segregation and the Evolution of Sex. Genetics
893 164:1099–1118.
894 R Core Team. 2024. R: A Language and Environment for Statistical Computing. R
895 Foundation for Statistical Computing, Vienna, Austria.
896 Robinson, J. A., D. Ortega-Del Vecchyo, Z. Fan, B. Y. Kim, B. M. vonHoldt, C. D. Marsden,
897 K. E. Lohmueller, and R. K. Wayne. 2016. Genomic Flatlining in the Endangered
898 Island Fox. Current Biology 26:1183–1189. Elsevier BV.
899 Robinson, J., C. C. Kyriazis, S. C. Yuan, and K. E. Lohmueller. 2023. Deleterious Variation
900 in Natural Populations and Implications for Conservation Genetics. Annu. Rev. Anim.
901 Biosci. 11:93–114. Annual Reviews.
902 Roze, D. 2016. Background Selection in Partially Selfing Populations. Genetics 203:937–
903 957.
904 Roze, D., and N. H. Barton. 2006. The Hill-Robertson Effect and the Evolution of
905 Recombination. Genetics 173:1793–1811.
906 Roze, D., and R. E. Michod. 2010. Deleterious Mutations and Selection for Sex in Finite
907 Diploid Populations. Genetics 184:1095–1112.
908 Salome, P. A., K. Bomblies, J. Fitz, R. A. E. Laitinen, N. Warthmann, L. Yant, and D. Weigel.
909 2012. The recombination landscape in *Arabidopsis thaliana* F2 populations. Heredity

910 108:447–455.
911 Schwander, T., and B. J. Crespi. 2009. Multiple direct transitions from sexual reproduction to
912 apomictic parthenogenesis in Timema stick insects. Evolution 63:84–103.
913 Sharp, N. P., and A. F. Agrawal. 2016. Low Genetic Quality Alters Key Dimensions of the
914 Mutational Spectrum. PLoS Biol. 14:e1002419.
915 Simmons, M. J., and J. F. Crow. 1977. Mutations Affecting Fitness in *Drosophila*
916 Populations. Annu. Rev. Genet. 11:49–78.
917 Smukowski Heil, C. 2023. Loss of Heterozygosity and Its Importance in Evolution. J Mol Evol
918 91:369–377.
919 Speijer, D., J. Lukeš, and M. Eliáš. 2015. Sex is a ubiquitous, ancient, and inherent attribute
920 of eukaryotic life. Proc. Natl. Acad. Sci. USA 112:8827–8834.
921 Spigler, R. B., K. Theodorou, and S. Chang. 2017. Inbreeding depression and drift load in
922 small populations at demographic disequilibrium. Evolution 71:81–94.
923 Stoffel, M. A., M. Esser, M. Kardos, E. Humble, H. Nichols, P. David, and J. I. Hoffman.
924 2016. inbreedR: an R package for the analysis of inbreeding based on genetic
925 markers. Methods Ecol Evol 7:1331–1339.
926 Takahashi, K. K., and H. Innan. 2022. Frequent somatic gene conversion as a mechanism
927 for loss of heterozygosity in tumor suppressor genes. Genome Res. 32:1017–1025.
928 The Arabidopsis Genome Initiative. 2000. Analysis of the genome sequence of the flowering
929 plant Arabidopsis thaliana. Nature 408:796–815.
930 Theologidis, I., I. M. Chelo, C. Goy, and H. Teotónio. 2014. Reproductive assurance drives
931 transitions to self-fertilization in experimental Caenorhabditis elegans. BMC Biol
932 12:93.
933 Tran Van, P., Y. Anselmetti, J. Bast, Z. Dumas, N. Galtier, K. S. Jaron, K. Martens, D. J.
934 Parker, M. Robinson-Rechavi, T. Schwander, P. Simion, and I. Schön. 2021. First
935 annotated draft genomes of nonmarine ostracods (Ostracoda, Crustacea) with
936 different reproductive modes. G3 11:jkab043.
937 Tucker, A. E., M. S. Ackerman, B. D. Eads, S. Xu, and M. Lynch. 2013. Population-genomic

938 insights into the evolutionary origin and fate of obligately asexual *Daphnia pulex*.
939 Proc. Natl. Acad. Sci. USA 110:15740–15745.
940 Uricchio, L. H., and R. D. Hernandez. 2014. Robust Forward Simulations of Recurrent
941 Hitchhiking. Genetics 197:221–236.
942 Vakhrusheva, O. A., E. A. Mnatsakanova, Y. R. Galimov, T. V. Neretina, E. S. Gerasimov, S.
943 A. Naumenko, S. G. Ozerova, A. O. Zalevsky, I. A. Yushenova, F. Rodriguez, I. R.
944 Arkhipova, A. A. Penin, M. D. Logacheva, G. A. Bazykin, and A. S. Kondrashov.
945 2020. Genomic signatures of recombination in a natural population of the bdelloid
946 rotifer Adineta vaga. Nat Commun 11:6421.
947 Weir, W., P. Capewell, B. Foth, C. Clucas, A. Pountain, P. Steketee, N. Veitch, M. Koffi, T.
948 De Meeûs, J. Kaboré, M. Camara, A. Cooper, A. Tait, V. Jamonneau, B. Bucheton,
949 M. Berriman, A. MacLeod, and D. Soldati-Favre. 2016. Population genomics reveals
950 the origin and asexual evolution of human infective trypanosomes. eLife 5:e11473.
951 Wright, S. 1951. The genetical structure of populations. Ann. Eugen. 15:323–354.
952 Wright, S. I., S. Kalisz, and T. Slotte. 2013. Evolutionary consequences of self-fertilization in
953 plants. Proc. R. Soc. B 280:20130133.
954 Xu, S., A. R. Omilian, and M. E. Cristescu. 2011. High Rate of Large-Scale Hemizygous
955 Deletions in Asexually Propagating *Daphnia*: Implications for the Evolution of Sex.
956 Mol. Biol. Evol. 28:335–342.
957 Yim, E., K. E. O'Connell, J. St. Charles, and T. D. Petes. 2014. High-Resolution Mapping of
958 Two Types of Spontaneous Mitotic Gene Conversion Events in *Saccharomyces*
959 *cerevisiae*. Genetics 198:181–192.