Evolutionary simulations of Z-linked suppression gene drives

Luke Holman*
**luke.holman@unimelb.edu.au*

Abstract

Lorem ipsum dolor sit amet, consectetur adipiscing elit, sed do eiusmod tempor incididunt ut labore et dolore magna aliqua. Ut enim ad minim veniam, quis nostrud exercitation ullamco laboris nisi ut aliquip ex ea commodo consequat. Duis aute irure dolor in reprehenderit in voluptate velit esse cillum dolore eu fugiat nulla pariatur. Excepteur sint occaecat cupidatat non proident, sunt in culpa qui officia deserunt mollit anim id est laborum.

Keywords: *W*- and *X*-shredders, Lepidopteran pests, meiotic drive, population control, schistosomiasis, selfish genes.

*School of BioSciences, The University of Melbourne, Victoria, Australia.

Introduction

Developments in biotechnology will soon make it feasible to control or eliminate populations of disease vectors, pathogens, agricultural pests, and invasive species using 'gene drives' [1–6]. Gene drives assist the propagation of engineered genes through populations using a range of mechanisms including gene conversion, poison-antidote systems, segregation distortion, and genetic incompatibility [7,8]. For example, CRISPR-Cas9 gene editing can be used to create a transgenic insertion that is transmitted to almost 100% of the offspring of heterozygous individuals instead of the usual 50%; this type of gene drive functions by inducing a double-strand break in the wild type allele, which is then repaired using the transgene as a template. Gene drives are often categorised into two types, both of which can be created with CRISPR-Cas9. 'Replacement drives' aim to propagate a human-beneficial allele that would not otherwise spread by selection, e.g. an allele that interferes with the tranmission of malaria by mosquitoes [1,9]. Conversely, 'suppression drives' aim to cause extinction (or at least a reduction in population size), for example by propagating an allele that causes lethality or sterility (MORE HERE [2,5]), or which skews the population sex ratio – typically towards males MORE HERE [10–12].

Recent theoretical papers have investigated the feasibility, efficacy, and potential negative consequencies of emerging gene drive technologies. For example, Noble et al. [6] used models to show that the basic version of a CRISPR gene drive might be highly invasive and could rapidly spread to fixation across whole meta-populations, which will sometimes be undesirable. Conversely, other models have concluded that alleles that are resistant to being cut and replaced by CRISPR gene drives could prevent them from spreading and acheiving their aims [13,14]. The issue of resistance is compounded because the standard implementation of CRISPR drives (but perhaps not updated versions; [4,5,14,15]) tends to create its own resistance alleles by deleting the Cas9 target site via non-homologous end joining (NHEJ; [1,14,16]). Given the potential safety, ethical, and sociopolitical concerns surrounding gene drives, some models have focused on gene drives that would go extinct once their job is done [11,17], would stay confined to particular populations [18], and/or could be reversed once they have spread [19].

Here, I focus on the evolutionary dynamics of Z-linked suppression gene drives. The simulation is inspired by proposals for various types of CRISPR-Cas9 Z-linked gene drives from Kevin Esvelt and colleagues (see www.sculptingevolution.org/genedrives/current/schistosomiasis; at the time of writing, these ideas have not been published in a journal or pre-print). Various Z-linked suppression drives proposed by Esvelt and colleagues are shown schematically in Figure XX. The gene drive would enjoy a transmission advantage in ZW females, and optionally also in ZZ males. Esvelt et al. propose that Z-linked drives could be used to control the trematode parasites (*Schistosoma* spp.) responsible for the deadly disease schistosomiasis, though Z-linked drives could theoretically be used to control any organism with female-heterogametic sex determination (such as Lepidopteran crop pests or invasive populations of birds).

A Z-linked gene drive could suppress populations by biasing gametogenesis in females, for example by cutting unique sequences on the W chromosome in order to destroy it; such a gene drive would be a 'W-shredder', similar to the X- and Y-shredders that are being

developed to control XY species [12,20–22]. Females carrying the gene drive would thus produce relatively few W-bearing eggs, and therefore produce mainly drive-carrying sons. Esvelt et al. point out that the evolutionary dynamics of the drive will depend on the fitness of drive carriers relative to wold types, the timing of W-shredding (e.g. in pre-meiotic cells vs mature ova), and the ecology of the target species. For example, some W-shredder designs might allow drive females to produce roughly the same number of (mostly-male) offspring as a wild-type female, because the W chromosome is destroyed early enough in oogenesis that lost W-bearing gametes can be replaced (Figure XX). Alternatively, drive-carrying females might produce half the number of offspring, e.g. if the drive works by destroying all ova or offspring that carry a W chromosome, and females cannot compensate by producing more. As an alternative to W-shredders, Esvelt et al. also proposed that one could suppress populations using a Z-linked locus that caused sterility or lethality in females. If this female-harming gene was capable of gene drive in males (see below), it could perhaps reach high enough frequencies to suppress the population.

Esvelt and colleagues also note that if the Z-linked locus caused gene drive in *males* in addition to females, it would probably spread through the population faster and be more likely to result in extinction. Male gene drive could be accomplished using 'standard' CRISPR-Cas9 gene conversion (REF), whereby the driving Z allele would convert the wild type locus using homing endonuclease activity followed by DNA repair, causing heterozygous males to produce mostly drive-carrying sperm and offspring.

Here, I present an evolutionary simulation that can accommodate all of these hypothesised types of Z-linked drives. I aimed to test which properties of the gene drive and the ecology of the target species are critical to determining the likelihood and speed with which the gene drive causes extinction. For example, the gene drive will presumably spread faster if it can bias transmission in both sexes, but perhaps a female-only gene drive (which might be easier to engineer) would be perfectly adequate. Also, since the population will become more male-biased as the gene drive invades, there will be eco-evo feedback (REF) that might affect the evolutionary outcome in non-intuitive ways. For example, the altered sex ratio might intensify the fitness advantage accruing to any resistant W chromosomes or autosomal modifiers that prevent W-shredding (due to Fisherian selection for an even sex ratio; [23]), relative to that observed in earlier models focusing on gene drives carried on autosomes [13,14]. Moreover, the change in sex ratio could affect the demographics of the population, particularly if males and females contribute differentially to density-dependent population growth [24], or have different dispersal rates [25]. The model incorporates the possibility that Z-linked resistance alleles are sometimes formed through NHEJ in males that are heterozygous for the drive allele [1,14,16]. It is not clear *a priori* whether the creation of resistant Z-linked alleles by NHEJ is a equally problematic for a Z-linked gene drive as it is for an autosomal drive, because it would only hinder gene conversion in males, assuming that NHEJ does not occur in response to W-shredding (which seems likely, because the W-shredder could be designed to target many repetitive regions of the W chromosome).

Methods

Overview

I model a finite population of dioecious diploids with ZW sex determination, living in j discrete habitat patches that are arranged linearly in a ring. The model considers the demography and evolution of a population into which $n_{release}$ males carrying a Z-linked gene drive are released. The drive allele causes either W-shredding or sterility in females, and optionally also causes gene drive in heterozygous males (e.g. via gene conversion of the non-driving Z). The generations are non-overlapping and each one proceeds as follows: birth, dispersal between patches, breeding with patches, and death of the parental generation. The species has 3 loci with 2-3 alleles each, some of which potentially show non-Mendelian inheritance. The equilibrium population size was roughly 10,000 in all simulations upon release of the gene drive. The model is a stochastic individual-based simulation written in R 3.4.0 and was run on the `Spartan` computer cluster at the University of Melbourne. An accompanying website describes the R scripts used to run, analyse and plot the model (link).

Loci and alleles

Each male in the simulation carries one Z-linked locus and two autosomal loci, each with two alleles. Each female carries a single allele at the Z-linked locus plus a W chromosome, as well as two alleles at both of the autosomal loci.

There are three possible Z-linked alleles: a gene drive allele (denoted Z^*), a wild-type allele ($Z+$) that is vulnerable to gene drive in Z^*Z+ males, and a resistant allele (Zr) that is immune to gene drive in Z^*Zr males. Similarly, there are two possible types of W chromosomes: a wild-type W chromosome ($W+$) that is vulnerable to gene drive by the Z^* allele, and a resistant W chromosome (Wr) that is immune to gene drive.

The two autosomal loci, denoted A/a and B/b, control immunity to W-shredding and gene conversion respectively. A/a and B/b could be called 'trans-acting' resistance loci, since they are at a different locus (indeed, a different chromosome) to the gene drive allele, in contrast to the 'cis-acting' resistance conferred by the Zr and Wr alleles. The A allele is dominant to a and confers immunity to Z-linked gene drive (e.g. W-shredding) in females. The B allele is dominant to b and confers immunity to Z-linked gene drive (e.g. gene conversion) in males.

Calculating female and male fitness

I assume that wild-type individuals (i.e. those lacking drive or resistance alleles) have fitness $w = 1$, while other genotypes have $0 \leq w \leq 1$. The fecundity of females carrying the gene drive is reduced by a factor $1 - C_f$. Small C_f implies minimal costs (e.g. because lost gametes/offspring are easily replaced), $C_f = 0.5$ could represent the case where all daughters die and are not replaced, and $C_f = 1$ means that females carrying Z^* are completely sterile

(which is useful for modelling a female-sterilising Z-linked drive as opposed to a W-shredder). Similarly, the fitness of males carrying the gene drive is reduced by a factor $1 - C_m$; male fitness determines mating success (see below). Furthermore, the resistant chromosomes *Wr* and *Zr* are assumed to reduce fitness by factors of $1 - C_w$ and $1 - C_z$ respectively. For brevity, I assume that the autosomal resistance alleles A and B are cost-free. All costs are multiplicative; for example, a *Z*Zr* male would have fitness $(1 - C_m)(1 - C_z)$. Additionally, all costs are assumed to be dominant, meaning that having one drive or resistance allele is equally costly as having two.

Gamete production and gene drive

I assume that the *A/a* and *B/b* loci segregate independently during meiosis and display standard Mendelian inheritance. Inheritance of the sex chromosomes is also Mendelian, except for certain genotypes carrying one *Z** allele.

Firstly, *Z*W+aaBB*, *Z*W+aaBb*, and *Z*W+aabb* females produce a fraction $\frac{1}{2}(1 + p_{shred})$ of Z-bearing gametes and $\frac{1}{2}(1 - p_{shred})$ W-bearing gametes. Therefore, these three female genotypes produce >50% sons when $p_{shred} > 0$, due to the shortage of W chromosomes in their gametes. Note that the gamete frequencies of *Z*Wr females*, or of females carrying at least one A allele, conform to the standard Mendelian expectations due to resistance.

Secondly, *Z*Z+AAbb*, *Z*Z+Aabb*, and *Z*Z+aabb* males produce a fraction $\frac{1}{2}(1 + p_{conv} - p_{conv}p_{nhej})$ of gametes carrying the *Z** allele, $\frac{1}{2}(1 - p_{conv})$ gametes carrying the *Z+* allele, and $\frac{1}{2}(p_{conv}p_{nhej}))$ gametes carrying the *Zr* allele. Thus, gene conversion occurs in males if $p_{conv} > 0$, meaning that the *Z** allele is over-represented in the gametes of these three male genotypes. The parameter p_{nhej} represents non-homologous end joining, in which an endonuclease-based gene drive fails to copy itself to the homologous chromosome, and instead deletes its target site and thereby creates a resistant allele. The gamete frequencies of *Z*Zr* males, or of males carrying at least one B allele, conform to the standard Mendelian expectations due to resistance.

Calculating female fecundity

In the breeding phase of the lifecycle, the model first determines the number of offspring produced by each female. The expected fecundity of female i (F_i) is affected by three factors: the female's genotype, the density of males and females in the local patch and/or in the full population, and some global parameters in the model, as follows:

$$F_i = (1 + w_i r(1 - (D_i/K)^{\alpha}))$$

where D_i is the 'density' experienced by female i, w_i is her fitness ($0 \leq w_i \leq 1$), K is the carrying capacity, and r and α are constants that control the maximum possible fecundity and the shape of density-dependence, respectively [26].

To ensure that the simulation captures various possible types of life history and ecology, I calculated density D_i in various ways in different simulation runs. First, I define the 'global density' d_g, which acts equally on every female in every patch, as

$$d_g = \sum_{i=1}^{N_f} w_i + \delta N_m$$

where N_f and N_m are the numbers of females and males across all patches, the first term is the summed fitnesses of all these females, and δ is a constant (range: $0 - \infty$) that scales the effect of each male on d_g relative to a female with fitness $w_i = 1$. This formulation means that females with high relative fitness (i.e. fecundity) have a stronger effect on the global density than do low-fitness females. I also assume that each male contributes a fixed amount to the global density, irrespective of his genotype/fitness (since I assume that male fitness only affects male mating success; see below). The parameter δ represents sex differences in ecological niche use and behaviour. For example, we might expect $\delta < 1$ in species where males and females utilise very different environmental niches, or $\delta > 1$ in species where males are harmful to females.

Second, we define the 'local density' d_j, which is experienced by every female in patch j, as

$$d_j = \sum_{i=1}^{n_{f,j}} w_i + \delta n_{m,j}$$

where $n_{f,j}$ and $n_{m,j}$ are the numbers of females and males in patch j. As before, this formulation means that d_j depends on the fitnesses of the females in the patch, as well as the number of males (scaled by the constant δ).

Finally, the overall density experienced by female i in patch j (D_i) is a composite of the global and local densities given by $D_i = \psi d_g + (1 - \psi) d_j$. The parameter ψ scales the importance of global and local density to female fecundity. When $\psi = 0$, only local density matters and selection on females is entirely "soft", while when $\psi = 1$ only global density matters and selection on females is completely "hard" (REFERENCE). Intermediate values of ψ produce a mixture of hard and soft selection on females, and the growth rate of population depends on density at both scales.

After calculating the expected fecundity of each female (F_i), we generate the realised fecundity of the female by randomly sampling from a Poisson distribution with $\lambda = F_i$ (allowing for stochastic variation in fecundity between females with equal F_i). If the resulting number of offspring exceeded the global carrying capacity K, the model randomly selects K surviving offspring.

Competition between males

After determining how many offspring each female produces, we determine the fathers of each of these offspring. We assume that all breeding occurs within patches, such that males

only compete for matings/fertilisations with males in the same patch. If the patch contains k different male genotypes and there are $n_1, n_2, ...n_k$ males of each genotype, the probability that a male of genotype k is the father of any given offspring is

$$p_j = \frac{n_k w_k}{\sum_{i=1}^{k} n_i w_i}$$

such that relatively common and/or high-fitness male genotypes are more likely to sire offspring. This formulation means that both sexes potentially reproduce with multiple partners.

Reproduction, mutation and dispersal

After picking the parents, the model randomly generates each offspring's genotype based on the expected gamete (and thus zygote) frequencies. Offspring are born in the same patch as their parents, and the parental generation is replaced by the offspring generation.

When an offspring is created, each *Z+* allele it carries has a chance μ_Z to mutate to a *Zr* allele, and *vice versa* (i.e. mutation in both directions is equally probable). Similarly, each *W+* allele has a chance μ_W to mutate to a *Wr* allele, and *vice versa*.

Female and male offspring disperse to another patch with probabilities x_f and x_m respectively. We model two types of dispersal, in separate simulations: local dispersal, in which offspring move to one of the two neighbouring patches with equal probability (recalling that the patches are arranged in a ring), or global dispersal, in which dispersing offspring can land in any of the other patches.

One compete run of the simulation

The model first initialises a population of 10,000 individuals (the carrying capacity, K) with low or zero frequencies of *Zr*, *Wr*, *A* and *B* alleles, higher frequencies of the wild type *Z+*, *W+*, *a*, and *b* alleles, and zero *Z** gene drive alleles. It then runs 50 generations of burn-in to allow the population to reach demographic and genotypic equilibrium. Next, $n_{release}$ males with the genotype *Z*Z*aabb* are added to the population just before fathers are selected, representing the release into the wild of a laboratory-reared strain homozygous for the driving *Z*. In some simulations, all the *Z*Z*aabb* males were released in a single patch, while in others the $n_{release}$ males were randomly and evenly divided across all k patches. The model continued until either A) the driving *Z** allele went extinct, B) the population went extinct, C) the *Wr* chromosome went to fixation (making population suppression impossible), D) the *Z** allele fixed, but failed to cause population extinction, or E) 900 generations had elapsed. The model recorded which of these five outcomes occurred, as well as the allele frequencies, population size, and sex ratio at each generation.

Investigating the parameter space

For each of the parameters in Table 1, I selected two or more possible parameter values (e.g. high versus low rates of W-shredding p_{shred}; many versus few patches k). I then ran the model once for all possible combinations of these parameter values (n = 6,000,000 model runs). The aim was to measure the effect of each parameter across a background of assumptions for the other parameters, as well as to investigate all possible 2-way interactions between the parameters.

Results

Three illustrative simulation runs

Figure 1 shows three contrasting evolutionary outcomes, illustrating some representative evolutionary dynamics from among the 6,000,000 simulation runs. Tables S1-S2 give the relative frequencies of the various possible outcomes (e.g. extinction occurred in 28% of simulations involving W-shredders).

In Figure 1A, the release of 20 Z^*Z^* males at generation 50 was followed by the rapid invasion of the Z^* allele, which caused population extinction by reducing the number of females. Figure 1A assumes that the Z^* alleles causes perfect W-shredding ($p_{shred} = 1$), that Z^* has minimal fitness costs, and there is no resistance to W-shredding (Table S3).

In Figure 1B, Z^* invaded but failed to cause extinction, even though it was assumed that $p_{shred} = 1$ and there is no resistance to W-shredding. The simulation underlying Figure 1B assumed the presence of heavy fitness costs to individuals carrying at least one Z^* allele ($c_f = 0.5$ and $c_m = 0.2$), and the absence of any gene drive in males ($p_{conv} = 0$). The assumptions $p_{shred} = 1$ and $c_f = 0.5$ imply that the W-bearing eggs/offspring of Z^*W+ females are destroyed but not replaced, such that W-shredding increases the proportion but not the absolute number of offspring that inherit the Z^* allele. Essentially Z^* spreads via 'spite' [REF], in that it removes W chromosomes from the local population and thereby makes room for more Z^* alleles, providing indirect fitness benefits. However, the net fitness returns of the Z^* allele's 'strategy' (i.e. sacrificing 20% fitness in males in order to remove W chromosomes in females) decline as the W chromosome becomes rarer, allowing the cost in males to greatly slow the spread of Z^*.

Lastly, Figure 1C shows a case where the invasion of Z^* was halted and then reversed by the evolution of autosomal and Z-linked resistance alleles. Following the introduction of the Z^* allele, resistant Zr mutants were created via non-homologous end joining, and then Zr spread to fixation due to its immunity to gene conversion in males. The autosomal resistance allele A also spread; A confers resistance to W-shredding and was initially present in the population at 5% frequency. The spread of A caused the sex ratio to revert to normal, preventing extinction, and Z^* went extinct due to its direct fitness costs no longer being outweighed by the benefits of W-shredding and gene conversion. Incidentally, the reason the

resistant allele A was favoured over a is that the male-biased population sex ratio created by Z^* elevates the reproductive value of daughters, and AA and Aa females produce more daughters than aa females in populations where Z^* is present.

Effects of each parameter on a *W*-shredder

Figure 2 shows the main effects of each of model parameter for models of W-shredders, arranged in approximate order of their effect on extinction probability. By far the most important parameters were the strength of W-shredding in females (p_{shred}), the existence of resistance against W-shredding, and the fitness cost the Z* allele to females (c_f). These results were expected: the gene drive will not cause extinction unless Z*-carrying females produce a highly male-biased sex ratio (e.g. due to complete and unresistable W-shredding), and extinction is more likely when Z*-carrying females produce larger numbers of Z*-carrying sons.

The strength of gene conversion in ZZ males (p_{conv}) was a good predictor of extinction probability, though it was not nearly as salient as was the strength of W-shredding. Similarly, the existence of autosomal alleles conferring resistance to W-shredding (at locus A/a in the model) virtually guaranteed that extinction could not occur, while autosomal alleles preventing gene conversion in males (locus B/b) had a strong but much weaker effect on the probability of extinction. Thus, the model indicates that it is desirable but not essential to design W-shredders that also drive in males (e.g. via gene conversion), since a strong W-shredder may cause extinction on its own.

Effects of each parameter on a female-sterilising *Z* drive

The model was also used to examine another possible suppression drive: the evolution of a Z-linked allele that causes gene drive in males and also causes total sterility in females ($c_f = 1$). This alternative type of drive caused extinction in a few simulation runs, though it was far less effective than the W-shredder. Thus, gene drives which work by sterilising females may be less effective at suppressing populations than drives which instead 'hijack' these females to produce more drive-carrying males. Female-sterilising gene drives may still be useful for some appliations, but my model extends earlier results [REFS] by showing that they are liable to fail in the presence of Z-linked or autosomal alleles conferring resistance to male gene drive.

...

As expected, extinction was less likely to occur when some individuals show resistance to W-shredding, due to either W-linked or autosomal alleles. The presence of alleles conferring resistance to gene drive in males (either on the Z chromomsome or an autosome) also had an effect on the extinction rate. However, the gene drive was still perfectly capable of causing extinction even when resistance to the male-acting part of the gene drive was ubiquitous (e.g. when the gene drive created a resistant allele 10% of the time). This represents further

evidence that it may not be necessary to create W-shredding gene drives that also drive when carried by males.

Interestingly, the Z^* allele was still able to spread and to cause extinction when it had a strong negative effect on male reproductive success (e.g. $c_m = 0.2$, meaning that a drive male is only 80% as likely to father offspring as is a wild-type male in the same patch). This result makes sense in light of the finding, mentioned above, that a female-sterilising Z-linked drive can suppress populations. A female who produces an all-male clutch of low-fitness sons will have a similar negative effect on population growth as a sterile female, except that her sons will carry drive and will thus contribute to causing extinction when they manage to breed. Also, in patches where the Z^* allele is locally common, any fitness costs it imposes on males will be relatively unimportant, since there are fewer wild types males to compete with.

Several of the ecological variables examined also affected the extinction probability. Chief among these was the shape parameter of the Richards density-dependence function, α. $\alpha < 1$ means that female fecundity declines at a decelerating rate as density increases, such that most of the population needs to be depleted before per-female fecundity noticeably increases, making extinction more likely. Conversely for $\alpha > 1$, fecundity declines at an accelerating rate with increasing density, making extinction less likely due to the immediate increases in per-female fecundity once the population begins to shrink due to the spread of the gene drive. Unsurprisngly, I also found that populations in which females can reach high fecundity at low population densities (captured by the parameter r) are more difficult to drive extinct, though the difference in extinction probability was minimal for a ten-fold difference in r. Also, extinction was slightly more probable when female fecundity was more sensitive to local density than global density (ψ in Figure 2). This is because local density can remain high (and thus, per-female fecundity can remain low) even in populations that are declining due to the spread of the Z^* allele in other patches.

I also found increasing rates of extinction with δ, the parameter that scales how male density affects female fecundity. When δ is high, extinction is more probable because female fecundity does not increase as much once the drive has begun to spread, due to the ever-increasing proportion of males as the population shrinks. Conversely, lower values of δ mean that male numbers are relatively unimportant in determining female fecundity, making extinction less likely since the removal of females by the gene drive lessens competition on the remaining females. This result highlights that it is important to consider the ecology of the target species when designing gene drives that work by eliminating one sex.

Populations that are split into many semi-isolated patches were more difficult to drive extinct that those that comapratively free of spatial structure, though the effect on extinction rate was small (k in Figure 2). The likely reason is that a highly-structured population allows for refugia where the drive allele is absent, as well as for the possibility for the wild type allele to recolonise patches in which local extinction has occurred. The frequency and mode of migration was relatively unimportant, though there was a slight tendency for higher dispersal rates to stave off extinction, presumably because dispersal allows recolonisation. In a similar vein, extinction was slightly less likely to occur when dispersal could carry individuals to any patch, as opposed to only neighbouring patches. Finally, there was essentially no effect of the release strategy, suggesting that it may be unnecessary to release a W-shredding gene

drive across the species' entire range provided that there is gene flow between patches. An additional implication of this result is that *Z*-linked gene drives would be highly invasive, as previously shown for autosomal drives [6].

Discussion

- male drive not needed. Efficient, unresistable W-shredding is best
- schistos have large males, small females
- birds often have sex-biased dispersal
- females may be more demographically limiting leps, since it is them that lays the eggs
- dominant costs assumption

Acknowledgements

I thank the organisers, funding bodies (European Society for Evolutionary Biology; Swiss National Science Foundation), and attendees of the 2018 ESEB *Progress Meetings in Evolutionary Biology* on gene drives. I also thank Kevin Esvelt for promoting ongoing research on his website; the ideas there were crucial to this paper.

References

1. Gantz VM, Jasinskiene N, Tatarenkova O, Fazekas A, Macias VM, Bier E, James AA. 2015 Highly efficient Cas9-mediated gene drive for population modification of the malaria vector mosquito *Anopheles stephensi*. *PNAS* **112**, E6736–E6743.

2. Hammond A *et al.* 2016 A CRISPR-Cas9 gene drive system targeting female reproduction in the malaria mosquito vector *Anopheles gambiae*. *Nature Biotechnology* **34**, 78.

3. Wang Z, Pan Q, Gendron P, Zhu W, Guo F, Cen S, Wainberg MA, Liang C. 2016 CRISPR/Cas9-derived mutations both inhibit HIV-1 replication and accelerate viral escape. *Cell Reports* **15**, 481–489.

4. Prowse TA, Cassey P, Ross JV, Pfitzner C, Wittmann TA, Thomas P. 2017 Dodging silver bullets: good CRISPR gene-drive design is critical for eradicating exotic vertebrates. *Proc. Roy. Soc. B* **284**, 20170799.

5. Kyrou K, Hammond AM, Galizi R, Kranjc N, Burt A, Beaghton AK, Nolan T, Crisanti A. 2018 A CRISPR–Cas9 gene drive targeting *doublesex* causes complete population suppression in caged *Anopheles gambiae* mosquitoes. *Nature Biotechnology*

6. Noble C, Adlam B, Church GM, Esvelt KM, Nowak MA. 2018 Current CRISPR gene drive systems are likely to be highly invasive in wild populations. *eLife* **7**, e33423.

7. Lindholm AK *et al.* 2016 The ecology and evolutionary dynamics of meiotic drive. *Trends in Ecology & Evolution* **31**, 315–326.

8. Champer J, Buchman A, Akbari OS. 2016 Cheating evolution: Engineering gene drives to manipulate the fate of wild populations. *Nature Reviews Genetics* **17**, 146.

9. Marshall JM, Akbari OS. 2015 Gene drive strategies for population replacement. In *Genetic control of malaria and dengue*, pp. 169–200. Elsevier.

10. Beaghton A, Beaghton PJ, Burt A. 2017 Vector control with driving Y chromosomes: modelling the evolution of resistance. *Malaria Journal* **16**, 286.

11. Burt A, Deredec A. 2018 Self-limiting population genetic control with sex-linked genome editors. *Proc. Roy. Soc. B* **285**, 20180776.

12. Papathanos PA, Windbichler N. 2018 Redkmer: An assembly-free pipeline for the identification of abundant and specific X-chromosome target sequences for X-shredding by CRISPR endonucleases. *The CRISPR Journal* **1**, 88–98.

13. Drury DW, Dapper AL, Siniard DJ, Zentner GE, Wade MJ. 2017 CRISPR/Cas9 gene drives in genetically variable and nonrandomly mating wild populations. *Science Advances* **3**, e1601910.

14. Unckless RL, Clark AG, Messer PW. 2017 Evolution of resistance against CRISPR/Cas9 gene drive. *Genetics* **205**, 827–841.

15. Esvelt KM, Smidler AL, Catteruccia F, Church GM. 2014 Emerging technology: Concerning rna-guided gene drives for the alteration of wild populations. *Elife* **3**, e03401.

16. Gantz VM, Bier E. 2015 The mutagenic chain reaction: A method for converting heterozygous to homozygous mutations. *Science*, aaa5945.

17. Min J, Noble C, Najjar D, Esvelt KM. 2017 Daisyfield gene drive systems harness repeated genomic elements as a generational clock to limit spread. *BioRxiv*, 104877.

18. Maselko M, Heinsch SC, Das S, Smanski MJ. 2018 Genetic incompatibility combined with female-lethality is effective and robust in simulations of *Aedes aegypti* population control. *bioRxiv*, 316406.

19. Vella MR, Gunning CE, Lloyd AL, Gould F. 2017 Evaluating strategies for reversing CRISPR-Cas9 gene drives. *Scientific Reports* **7**, 11038.

20. North A, Burt A, Godfray HCJ. 2013 Modelling the spatial spread of a homing endonuclease gene in a mosquito population. *Journal of Applied Ecology* **50**, 1216–1225.

21. Galizi R, Doyle LA, Menichelli M, Bernardini F, Deredec A, Burt A, Stoddard BL, Windbichler N, Crisanti A. 2014 A synthetic sex ratio distortion system for the control of the human malaria mosquito. *Nature Communications* **5**, 3977.

22. Prowse TA, Adikusuma F, Cassey P, Thomas P, Ross JV. 2019 A Y-chromosome shredding gene drive for controlling pest vertebrate populations. *eLife* **8**, e41873.

412 23. Holman L, Price TA, Wedell N, Kokko H. 2015 Coevolutionary dynamics of polyandry
413 and sex-linked meiotic drive. *Evolution* **69**, 709–720.

414 24. Rankin DJ, Kokko H. 2007 Do males matter? The role of males in population dynamics.
415 *Oikos* **116**, 335–348.

416 25. Li X-Y, Kokko H. 2019 Sex-biased dispersal: A review of the theory. *Biological Reviews*
417 **in press**.

418 26. Fowler CW. 1981 Density dependence as related to life history strategy. *Ecology* **62**,
419 602–610.

Tables

Table 1: List of variables, and their corresponding parameter(s) in the model, which were varied in order to study their effects on the likelihood of population extinction.

Variable	Parameter(s)	Outcome
Strength of gene drive in females (e.g. W-shredding)	p_{shred}	1.00
Strength of gene drive in males (e.g. gene conversion)	p_{conv}	1.00
Cost of gene drive allele to female fecundity	c_f	1.00
Cost of gene drive allele to male mating success	c_m	1.00
Frequency of W-linked resistance mutations	μ_W	1.00
Frequency of Z-linked resistance mutations and NHEJ	μ_Z and p_{nhej}	1.00
Frequency of autosomal resistance alleles	μ_A and μ_B	1.00
Patchiness of the population	k	1.00
Dispersal rate of males and females	x_m and x_f	1.00
Global versus local density-dependence of female fecundity	ψ	1.00
Contribution of males relative to females in density-dependence	δ	1.00
Number of gene drive carrier males released	$n_{release}$	1.00
Release strategy: all in one patch, or global	-	1.00
Fecundity of females at low population densities	r	1.00
Shape of density dependence	α	1.00

Figures

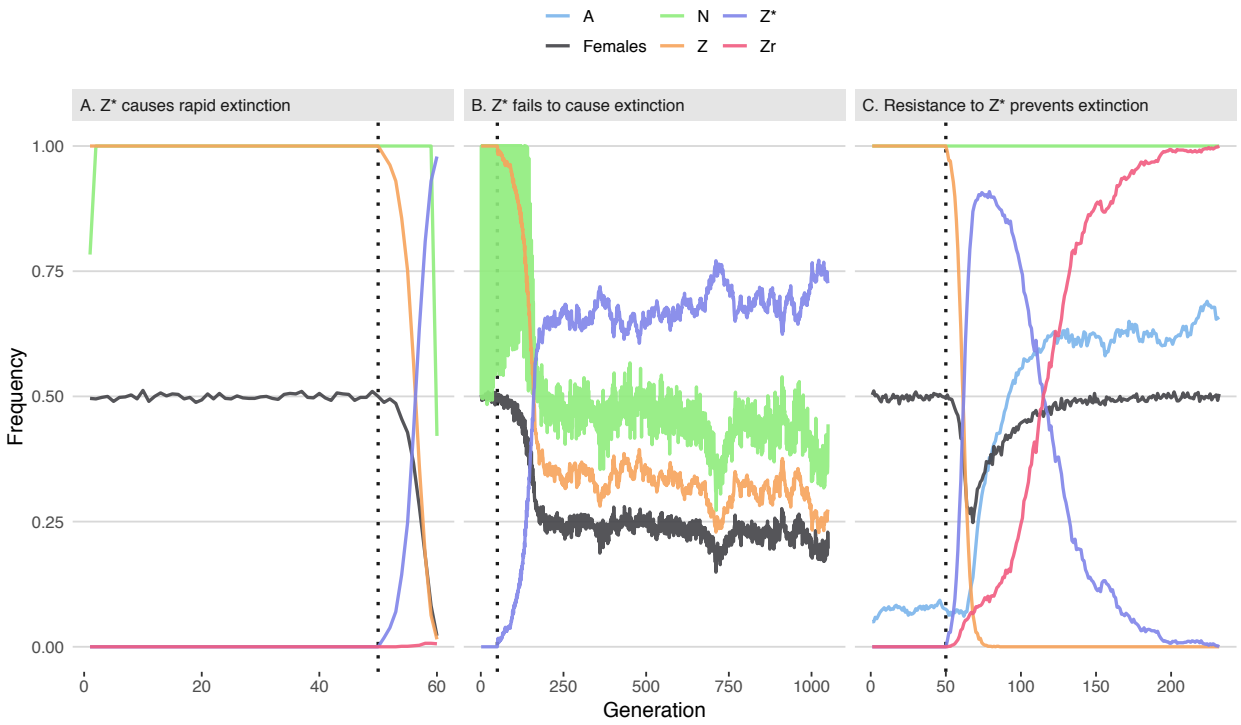

Figure 1: Three illustrative runs of the simulation, showing evolution in response to the introduction of 20 males carrying a W-shredder at Generation 50 (marked by the dotted line). In panel A, the driving Z^* allele fixed very quickly, causing population extinction as the number of females dropped to zero. In panel B, the Z^* allele spread up until the point that its fitness costs began to negate its transmission advantage, causing the population to halve in size but not to go extinct. In panel C, the Z^* allele invaded, selecting for the resistance alleles A and Z^r, and causing the Z^* allele to reverse course and go extinct. The population size N is shown as a fraction of its maximum value of 10,000. Table S3 gives the parameter spaces used for these three runs.

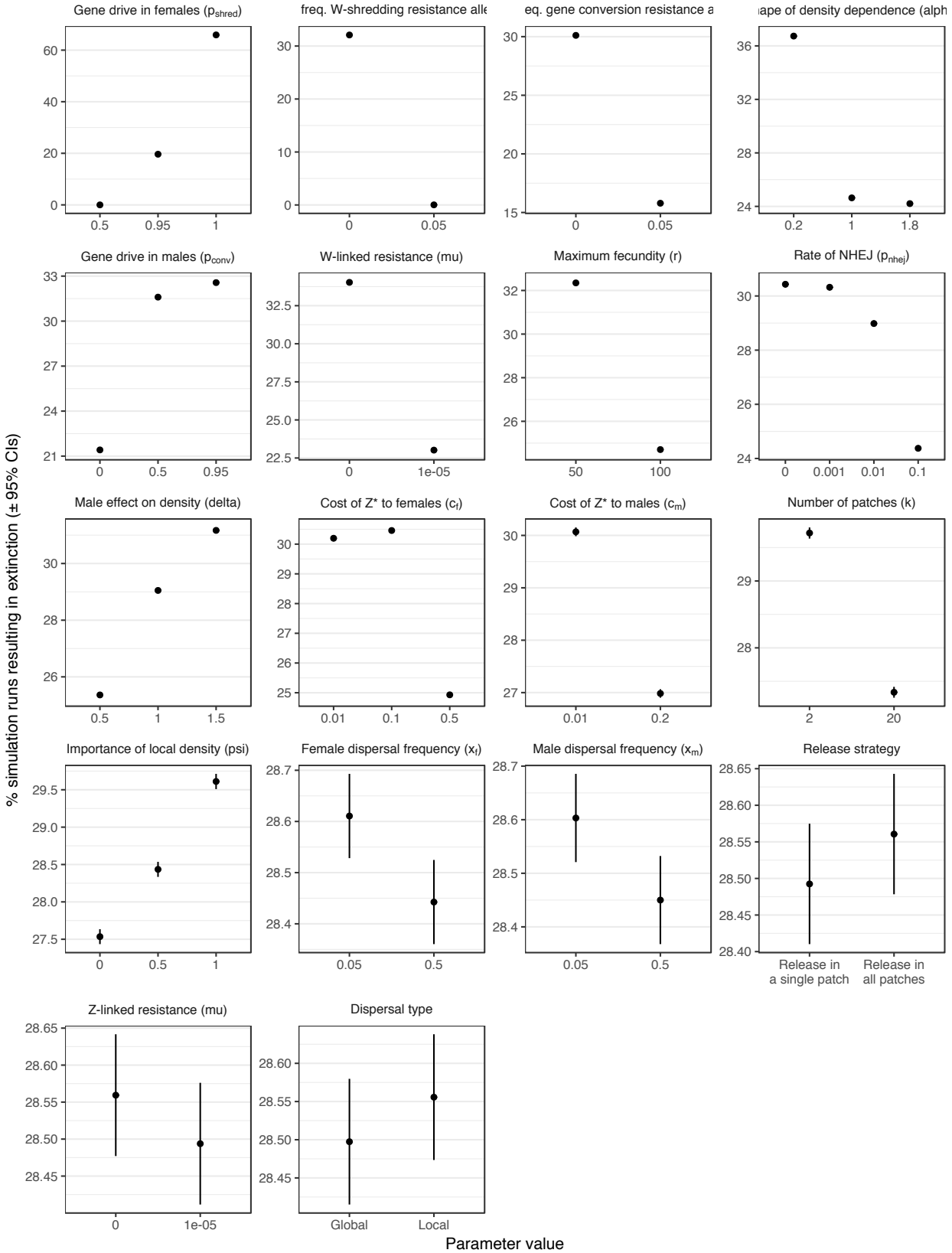

Figure 2: The percentage of simulations of a W-shredder that ended in extinction, for all runs with a particular value (shown on the x-axis) for a given parameter (shown in the panels). For example, there were no exrinctions in any of the thousands of runs for which I assumed $p_{shred} = 0.5$, while 60% of runs where $p_{shred} = 1$ resulted in extinction. The panels are ordered by the range of the x-axis, which gives some idea of the relative importance of the variables for the probability of extinction. Figure S1 gives a similar plot for simulations of a female-sterilising Z^* allele.

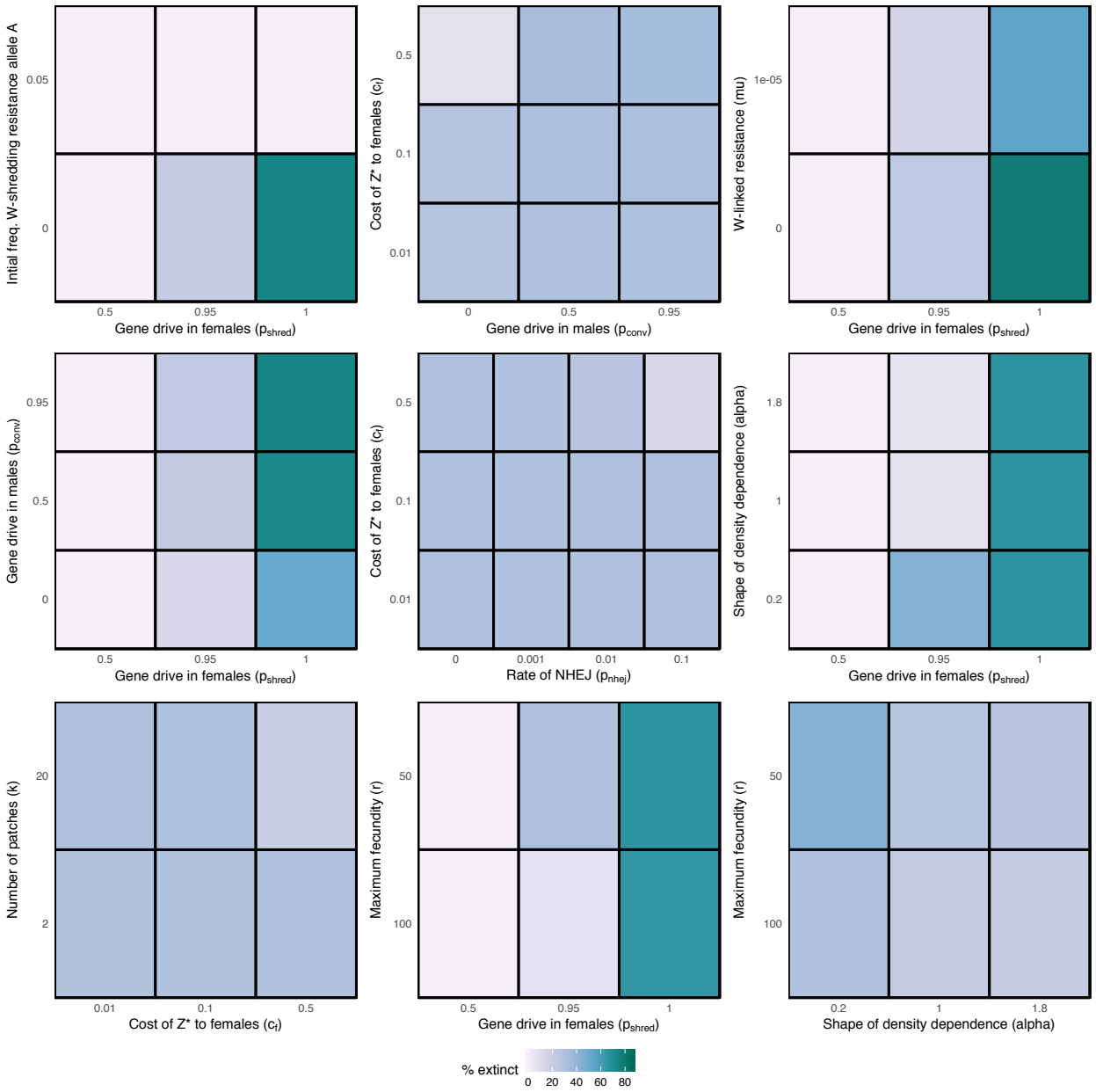

Figure 3: Heatmap showing the nine strongest interactions between pairs of parameters in the model, as determined by the GLM plotted in Figure SX.

Supporting information