The maintenance of polygenic sex determination depends on the dominance of fitness effects which are predictive of the role of sexual antagonism

Running title: Polygenic sex determination

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Abstract

In species with polygenic sex determination, multiple male- and/or female-determining loci on different proto-sex chromosomes segregate as polymorphisms within populations. The extent to which these polymorphisms are stable equilibria is not yet resolved. Previous work demonstrated that polygenic sex determination is most likely to be maintained as a stable polymorphism when the proto-sex chromosomes have opposite (sexually antagonistic) fitness effects in males and females. However, these models usually consider polygenic sex determination systems with only two proto-sex chromosomes, or they do not broadly consider the dominance of the variants under selection. To address these shortcomings, I used forward population genetic simulations to identify selection pressures that can maintain polygenic sex determination under different dominance scenarios in a system with more than two proto-sex chromosomes (modeled after the house fly). I found that overdominant fitness effects of male-determining proto-Y chromosomes in males are more likely to maintain polygenic sex determination than dominant, recessive, or additive fitness effects. I also found that additive fitness effects that maintain polygenic sex determination have the strongest signatures of sexually antagonistic selection, but there is also some evidence for sexually antagonism when fitness effects of proto-Y chromosomes are dominant or recessive. More generally, these results suggest that the expected effect of sexually antagonistic selection on the maintenance of genetic variation in natural populations will depend on whether the alleles are sex-linked and the dominance of their fitness effects.

Keywords

Sex determination; sex chromosomes; sexual antagonism; dominance

Introduction

Sex determination is the developmental process by which a genetic signal or environmental cue initiates sexually dimorphic gene regulatory pathways to produce phenotypically different males and females (Bachtrog et al. 2014). The master regulators of sex determination evolve fast, often differing between closely related species and sometimes even polymorphic within populations (Bull 1983; Wilkins 1995; Beukeboom and Perrin 2014). These master regulators can be found on sex chromosomes, and the evolutionary divergence of sex determiners can drive turnover of the sex chromosomes (Abbott et al. 2017). Species with polygenic sex determination have multiple master sex determining loci on different chromosomes segregating within natural populations (Moore and Roberts 2013). Understanding the population dynamics of polygenic sex determination systems is informative of the factors responsible for the evolutionary divergence of sex determination pathways and sex chromosomes.

Polygenic sex determination has been observed in many animal and plant species (Moore and Roberts 2013; Bachtrog et al. 2014; Beukeboom and Perrin 2014). Most population genetic models predict that polygenic sex determination will be an unstable intermediate between monogenic equilibria (Rice 1986; van Doorn and Kirkpatrick 2007). Models that do allow for stable polygenic sex determination often predict that opposing selection pressures in males and females (i.e., sexually antagonistic selection) may be important for maintaining the polymorphism (Bull 1983; Rice 1986; van Doorn and Kirkpatrick 2007). It is also possible that variable selection pressures across heterogenous environments could maintain polygenic sex determination (Bateman and Anholt 2017).

Most previous theoretical treatments of the selection pressures that maintain polygenic sex determination considered specific cases with a small number of sex chromosomes segregating within a population. For example, in the platyfish, *Xiphophorus maculatus*, a single chromosome can be an X, Y, or Z (Kallman 1973). Orzack et al. (1980) showed that this polymorphism is maintained because heterogametic females (WY or WX) and males (XY) have higher fitness than their homogametic counterparts (XX females or YY males). In addition, van Doorn and Kirkpatrick (2007, 2010) showed that sexually antagonistic selection could maintain a polymorphism in which one chromosome segregates as an XY pair and another chromosome is either an XY or ZW pair. However, these models do not capture the full diversity of polygenic sex determination, which can include, for example, more than two XY or ZW pairs segregating within a single population (Moore and Roberts 2013). Previous models also did not broadly consider how the maintenance of polygenic sex determination depends on the dominance of

60 fitness effects. Dominance is important for young proto-Y and proto-W chromosomes because
61 the proto-X/Y and proto-Z/W contain almost all of the the same genes, but they may differ in the
62 alleles they carry. This is different from heteromorphic sex chromsoomes, where dominance is
63 important for the X or Z chromosome (because it is diploid in one sex), and not the haploid Y or
64 W (Rice 1984; Charlesworth et al. 1987).

65 The house fly, *Musca domestica*, is a well-suited model for investigating the
66 maintenance of a complex polygenic sex determination because it has a highly polymorphic sex
67 determination system (Hamm et al. 2015). The *M. domestica male determiner* (*Mdmd*) can be
68 found on at least four of the six house fly chromosomes in natural populations (Sharma et al.
69 2017). *Mdmd* causes the house fly ortholog of *transformer* (*Md-tra*) to be spliced into a non-
70 functional isoform, which leads to male-specific splicing and expression of downstream target
71 genes, thereby producing fertile males (Hediger et al. 2010). In the absence of *Mdmd*, *Md-tra* is
72 spliced into a female-determining transcript that regulates the splicing of downstream targets,
73 which promote the development of female morphological and behavioral traits (Hediger et al.
74 2004; Meier et al. 2013).

75 There are two common male-determining proto-Y chromosomes and one female-
76 determining proto-W chromosome in house fly populations. *Mdmd* is most commonly found on
77 the Y (Y^M) and third (III^M) chromosomes in natural populations (Hamm et al. 2015). Both Y^M and
78 III^M are young proto-Y chromosomes that arose recently (Meisel et al. 2017). In addition, there is
79 a dominant allele of *Md-tra* that is found in many house fly populations (*Md-tra*^{D}), which is
80 resistant to *Mdmd* and causes female development regardless of whether there are copies of
81 *Mdmd* in the genotype (McDonald et al. 1978; Hediger et al. 2010). *Md-tra* is found on the fourth
82 chromosome, and thus fourth chromosomes carrying *Md-tra*^{D} (which I will refer to as IV^F) are W
83 chromosomes. All sequences of *Md-tra*^{D} from around the world are identical (Scott et al. 2014),
84 suggesting that *Md-tra*^{D} arose recently and IV^F is a young proto-W chromosome. In populations
85 where Y^M, III^M, and *Md-tra*^{D} all segregate, there are 18 possible sex chromosome genotypes
86 (Table 1).

87 There is evidence that natural selection maintains polygenic sex determination in house
88 fly populations. First, the proto-Y chromosomes have remained at stable frequencies within
89 house fly populations over decades (Kozielska et al. 2008; Meisel et al. 2016). Second, Y^M and
90 III^M are distributed along north-south clines on multiple continents (Denholm et al. 1986; Tomita
91 and Wada 1989; Hamm et al. 2005; Kozielska et al. 2008), and temperature is the best predictor
92 of their frequencies (Feldmeyer et al. 2008). This suggests that heterogenous (temperature-
93 dependent) selection pressures maintain the Y^M and III^M chromosomes across populations. The

stable frequencies within populations, yet divergent frequencies across populations, suggest that there are specific fitness effects associated with the house fly proto-Y chromosomes within each population. Third, males carrying multiple proto-Y chromosomes (e.g., both Y^M and III^M, or two copies of III^M) can be found in some populations (e.g., Hamm and Scott 2009). The frequency of *Md-tra*^{D} is positively correlated with the frequency of these multi-Y males across populations, suggesting that selection for balanced sex ratios may further maintain polygenic sex determination (Meisel et al. 2016).

The rich body of observational data in house fly makes it a well-suited system around which to develop population genetic models to determine how polygenic sex determination can be maintained in natural populations. For example, Kozielska et al. (2006) showed that, even in the complex house fly polygenic sex determination system, sex ratios are not expected to deviate substantially from 1:1 male:female. In addition, despite no experimental evidence for sexually antagonistic effects of any house fly proto-sex chromosomes, another population genetic model demonstrated that the frequencies of the proto-sex chromosomes in natural populations are consistent with sexual antagonism (Meisel et al. 2016). However, that model assumed that the proto-Y chromosomes (Y^M and III^M) have additive fitness effects, which may not be true if, for example, beneficial mutations are recessive or dominant (Orr 2010). In addition, Y chromosomes are expected to carry recessive deleterious alleles (Charlesworth and Charlesworth 2000), which can further violate the assumption of additive fitness effects. I used population genetic simulations to investigate whether sex chromosomes with non-additive fitness effects can maintain a complex polygenic sex determination system. I specifically tested if these selection pressures can produce proto-sex chromosome frequencies similar to those observed in natural populations of house fly, and I determined the conditions under which sexually antagonistic effects of those proto-sex chromsomes are expected.

122 **Table 1**. House fly sex chromosome genotypes.

	genotype			
	X or Y^M	III or III^M	IV or IV^F	sex
f_1	X/X	III/III	IV/IV	female
f_2	X/X	III/III	IV/IV^F	female
f_3	X/X	III/III^M	IV/IV^F	female
f_4	X/X	III^M/III^M	IV/IV^F	female
f_5	X/Y^M	III/III	IV/IV^F	female
f_6	X/Y^M	III/III^M	IV/IV^F	female
f_7	X/Y^M	III^M/III^M	IV/IV^F	female
f_8	Y^M/Y^M	III/III	IV/IV^F	female
f_9	Y^M/Y^M	III/III^M	IV/IV^F	female
f_{10}	Y^M/Y^M	III^M/III^M	IV/IV^F	female
m_1	X/X	III/III^M	IV/IV	male
m_2	X/X	III^M/III^M	IV/IV	male
m_3	X/Y^M	III/III	IV/IV	male
m_4	X/Y^M	III/III^M	IV/IV	male
m_5	X/Y^M	III^M/III^M	IV/IV	male
m_6	Y^M/Y^M	III/III	IV/IV	male
m_7	Y^M/Y^M	III/III^M	IV/IV	male
m_8	Y^M/Y^M	III^M/III^M	IV/IV	male

123

124 **Material and Methods**

125 ***General overview***

126 I used a simulation approach to identify fitness effects of two proto-Y chromosomes (Y^M
127 and III^M) and one proto-W chromosome (IV^F) that maintain polygenic sex determination under
128 four different dominance models. To do so, I first assigned random fitness effects to the Y^M, III^M,

129 and IV^F chromosomes (Figure 1A). I then calculated the fitness of each of the 18 genotypes
130 (Table 1) assuming one of four dominance models for the proto-Y chromosomes (Y^M and III^M):
131 additive effects, dominant effects, recessive effects, or overdominant effects (i.e., heterozygote
132 advantage). I next determined the frequency of each genotype and proto-sex chromosome after
133 1,000 generations of random mating with the assigned fitness effects (Figure 1A). I did this
134 using simulations for one million combinations of fitness effects using four different starting
135 genotype frequencies for each dominance model. I retained fitness effects in which all three
136 proto-sex chromosomes remained polymorphic after 1,000 generations, and from those I
137 selected the 1,000 for which the genotype frequencies were closest to those observed in natural
138 populations (Figure 1A). I also evaluated how well those fitness effects maintain polygenic sex
139 determination in a finite population (Figure 1B).
140 I used simulations to test if fitness effects maintain polygenic sex determination because
141 the model parameters were too complex to analytically solve for equilibrium conditions.
142 Specifically, I used dominance models to impose natural selection and recursion equations to
143 model random mating, and they could not be solved simultaneously. In my simulations, I tested
144 if polygenic sex determination persists for 1,000 generations, which may not reflect an
145 equilibrium state. In the Results, I show how this approach can be problematic if proto-Y
146 chromosomes are on a path to fixation or loss but have not yet reached fixation at 1,000
147 generations. However, most of my results are robust to this limitation because I specifically test
148 for proto-sex chromosome polymorphisms at frequencies resembling those observed in natural
149 populations. These observed frequencies are not near absorbing states involving fixation or
150 loss, and they are therefore more likely to be true equilibria.

151 ***Fitness effects and genotypic fitness***

152 To generate the fitness of the 18 possible sex chromosome genotypes (Table 1), I first
153 assigned fitness effects to Y^M, III^M, and IV^F by sampling from a uniform distribution between -1
154 and 1. Positive fitness effects mean that the proto-sex chromosome increases fitness (i.e., it is
155 beneficial), and negative values mean that it decreases fitness (i.e., it is deleterious). Fitness
156 effects of Y^M and III^M were assigned separately for males and females. The effect of IV^F was
157 only assigned for females because the female-determining proto-W chromosome (carrying *Md-*
158 *tra*^{D}) cannot be found in male genotypes (Table 1). Thus, there are five fitness effects: Y^M in
159 males (M_Y), Y^M in females (F_Y), III^M in males (M_{IIIM}), III^M in females (F_{IIIM}), and IV^F in
160 females (F_{IVF}). The fitness effects of the proto-sex chromosomes were then used to calculate
161 the fitness of the 18 possible sex chromosome genotypes (Table 1), using one of four

dominance scenarios. In each dominance scenario, the fitness of the 18 genotypes is represented by an array of 18 fitness values.

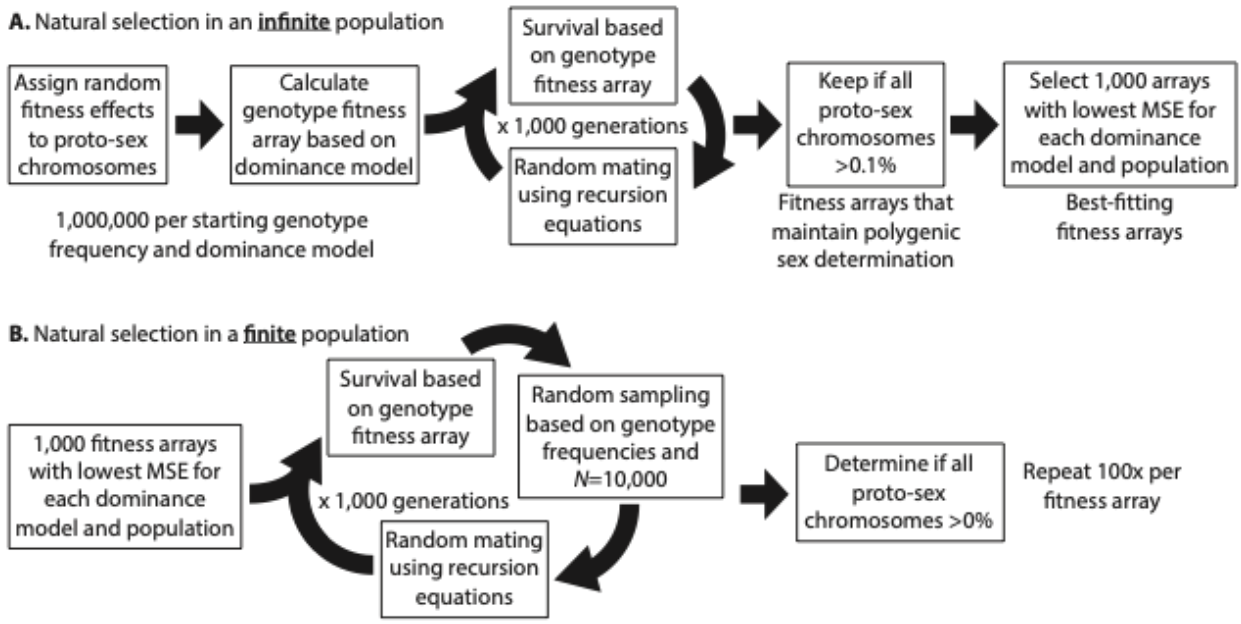


Figure 1. Steps in forward population genetic simulations with **(A)** infinite or **(B)** finite populations.

First, in the additive model, a baseline fitness of 1 was assigned to the f_1 genotype, which does not have any proto-Y or proto-W chromosomes (X/X; III/III; IV/IV). The fitnesses of the remaining genotypes were then calculated by adding the effects of Y^M, III^M, and IV^F to that baseline value according to their representation in each genotype (Supplementary Table 1). For example, the f_6 genotype (X/Y^M; III/III^M; IV/IV^F) has a fitness of $1 + F_Y + F_{IIIM} + F_{IVF}$ because it is female and there is one copy each of Y^M, III^M, and IV^F in the genotype. The m_2 genotype (X/X; III^M/III^M; IV/IV) has a fitness of $1 + 2M_{IIIM}$ because it is male and there are two copies of III^M.

In the second scenario, the fitness effects of the proto-Y chromosomes (Y^M and III^M) are dominant relative to their homologous proto-X chromosomes (X and III) in both males and females (Supplementary Table S2). As in the additive model, the fitness of the f_1 genotype was defined as 1 because it does not have any proto-Y or proto-W chromosomes. When the fitness effects of the proto-Y chromosomes are dominant, adding a second copy of Y^M or III^M to a genotype does not change the fitness of the genotype relative to an individual with one copy of that proto-Y chromosome. For example, genotypes m_1 (X/X; III/III^M; IV/IV) and m_2 (X/X; III^M/III^M; IV/IV) both have a fitness of $1 + M_{IIIM}$ because they have at least one III^M chromosome.

183 However, the inter-chromosomal effects are additive, such that the m_4, m_5, m_7, and m_8
184 genotypes all have a fitness of $1 + M_Y + M_{IIIM}$. In addition, IV^F has an additive effect on
185 female fitness, such that the f_5 (X/Y^M; III/III; IV/IV^F) and f_8 (Y^M/Y^M; III/III; IV/IV^F) genotypes both
186 have a fitness of $1 + F_Y + F_{IVF}$.

187 Third, when the effects of the proto-Y chromosomes are recessive, a single copy of Y^M
188 or III^M has no effect on genotype fitness in either males or females; genotype fitness is only
189 affected when there are two copies of either Y^M or III^M (Supplementary Table S3). For example,
190 genotype m_1 (X/X; III/III^M; IV/IV) has a fitness of 1, and m_2 (X/X; III^M/III^M; IV/IV) has a fitness of
191 $1 + M_{IIIM}$ because the fitness effect of III^M is only realized in the homozygous m_2 genotype.
192 Once again, inter-chromosome effects are additive, such that the fitness of the m_8 genotype
193 (Y^M/Y^M; III^M/III^M; IV/IV) is $1 + M_Y + M_{IIIM}$. As in all of the models, IV^F has an additive effect
194 on female fitness, such that the f_8 genotype (Y^M/Y^M; III/III; IV/IV^F) has a fitness of
195 $1 + F_Y + F_{IVF}$.

196 In the fourth scenario, the Y^M and III^M proto-Y chromosomes have overdominant effects
197 in males (Supplementary Table S4). In this scenario, a single-copy of either Y^M or III^M increases
198 male fitness because the chromosomes carry male-beneficial alleles with additive or dominant
199 effects. However, in the overdominant model, the proto-Y chromosomes also carry recessive
200 alleles that are deleterious in males (Charlesworth and Charlesworth 2000). Those recessive
201 deleterious alleles confer a fitness cost on male genotypes that are homozygous for either Y^M or
202 III^M (i.e., m_2, m_6, and m_8). To implement this, I determined the fitness effects of Y^M and III^M in
203 males (i.e., M_Y and M_{IIIM}) by taking the absolute value of the fitness sampled from the
204 uniform distribution between -1 and 1. I only applied those fitness effects to heterozygous male
205 genotypes (i.e., Y/Y^M and III/III^M). This ensures that heterozygous male genotypes have higher
206 fitness than either homozygote. For example, the m_1 genotype (X/X; III/III^M; IV/IV) has a fitness
207 of $1 + M_{IIIM}$, while the m_2 genotype (X/X; III^M/III^M; IV/IV) has a fitness of 1. Inter-chromosomal
208 fitness effects, on the other hand, are additive, as in the other three scenarios. Therefore, the
209 fitness of the m_4 genotype (X/Y^M; III^M/III; IV/IV) is $1 + M_Y + M_{IIIM}$. In contrast to males,
210 fitness effects in females are additive and drawn from a uniform distribution between -1 and 1
211 (as in the additive scenario). The female fitness effects are additive because the alleles that
212 increase fitness in heterozygous males are assumed to not confer a fitness benefit in females.
213 As in the other three scenarios, IV^F has an additive inter-locus effect on female fitness.

214 When calculating the fitness values of all 18 genotypes in a fitness array, the following
215 two normalizations were performed to ensure that the fitness of each genotype falls between 0

216 and 1. First, fitness effects of each proto-Y and proto-W chromosome can be negative because
217 they are drawn from a uniform distribution between -1 and 1. When the fitness effects of the
218 proto-sex chromosomes are negative, the fitness values of genotypes with multiple proto-Y and
219 proto-W chromosomes could be negative. To ensure that the fitness values of all 18 genotypes
220 are positive, the minimum fitness value across all 18 genotypes was subtracted from each
221 genotype fitness value. Second, genotype fitness values would be greater than 1 if the fitness
222 effect of a proto-Y or proto-W chromosome is positive. To ensure that all genotype fitness
223 values are less than 1, the adjusted fitness values were then divided by the maximum fitness
224 value across all 18 genotypes.

225 All four scenarios (additive, dominant, recessive, or overdominant) attribute fitness
226 effects to each proto-sex chromosome, whether it is Y^M, III^M, or IV^F. I am therefore assuming
227 that all copies of each proto-Y or proto-W chromosome in the population carry an identical suite
228 of beneficial and deleterious alleles (i.e., all copies of III^M are identical). In addition, all models
229 assume complete linkage between any allele(s) under selection and the *Mdmd* locus on the Y^M
230 and III^M chromosomes. Similarly, I assume complete linkage between the female-determining
231 *Md-tra*^{D} locus and any alleles on the IV^F chromosome. Complete linkage may be achieved if, for
232 example, chromosomal inversions suppress recombination between the proto-Y and proto-X (or
233 proto-Z and proto-W) chromosomes in heterozygotes (Bergero and Charlesworth 2009; Wright
234 et al. 2016). Free recombination in sex chromosome homozygotes would not affect genetic
235 variation on a sex chromosome because all copies of each chromosome are assumed to carry
236 the same alleles.

237 ***Simulations with infinite population size***

238 I used forward simulations to determine how the fitness effects of the proto-sex
239 chromosomes affect their frequencies in populations. These simulations were performed with
240 non-overlapping generations and random mating, assuming a population of infinite size
241 (simulations with finite population sizes are described later). Each generation of a simulation
242 consists of two discrete steps (Figure 1A). First, each of the 18 genotypes survives according to
243 its fitness value. This is accomplished by multiplying the frequency of each of the 18 genotypes
244 by its corresponding fitness value. Second, the 18 genotypes mate randomly and produce
245 progeny given by recursion equations, described in the Supplementary Table S5 and elsewhere
246 (Hamm 2008; Meisel et al. 2016). After 1,000 generations of selection and random mating, the
247 frequency of each genotype and proto-sex chromosome was calculated. This process was

repeated for 1,000,000 fitness arrays for each dominance scenario (additive, dominant, recessive, and overdominant).

I considered four possible initial genotype frequencies for each simulation. The first three initial frequencies are based on the observed frequencies of Y^M, III^M, and *Md-tra*^{D} from three North American populations (Meisel et al. 2016). These populations were sampled in Chino, CA (Meisel et al. 2016), Wake County, NC (Hamm and Scott 2008), and Chemung County, NY (Scott et al. 2013). Initial genotype frequencies were calculated based on the observed frequencies of Y^M, III^M, and *Md-tra*^{D}, assuming random mating (Meisel et al. 2016). These three populations were chosen because Y^M, III^M, and *Md-tra*^{D} have all remained at a frequency >1% across multiple years of sampling (Hamm et al. 2005; Hamm and Scott 2008; Meisel et al. 2016). I used frequencies from actual populations as the initial frequencies because I am specifically interested in how selection can maintain polygenic sex determination at the frequencies observed in natural populations. However, I also performed simulations in which all 18 genotypes started at the same initial frequency (i.e., 1/18 for each genotype). This allows me to evaluate how selection pressures can drive the proto-sex chromosomes to the frequencies observed in natural populations if they start at arbitrary values that deviate from the observed frequencies.

To determine whether the fitness effects maintain polygenic sex determination, I first selected fitness arrays that produced frequencies of each proto-sex chromosome (Y^M, III^M, IV^F, and their homologous chromosomes, X, III, and IV) that are all >0.1% after the 1,000 generation simulations. This criterion ensures that all three chromosomes are polymorphic with both alleles at a frequency that is measurable (i.e., 1/1,000) given the sampling schemes used in previous studies of natural house fly populations (Hamm et al. 2005; Hamm and Scott 2008; Meisel et al. 2016). I refer to genotype fitness arrays with all proto-sex chromosomes present at a frequency >0.1% as maintaining polygenic sex determination, although not necessarily at the frequencies observed in natural populations.

From the genotype fitness arrays that maintain polygenic sex determination (i.e., all proto-sex chromosomes >0.1%), I selected those that produce proto-sex chromosome frequencies most similar to the frequencies observed in natural populations. To do so, I calculated the mean squared error (MSE) between the frequencies of Y^M, III^M, and IV^F after 1,000 generations in a simulation (S) and the observed frequencies (O) in a natural population (CA, NC, or NY):

$$MSE = \frac{1}{3}[(O_{Y^M} - S_{Y^M})^2 + (O_{III^M} - S_{III^M})^2 + (O_{tra^D} - S_{tra^D})^2].$$

When using simulations that started with proto-sex chromosome frequencies observed in a population (e.g., CA), I only used MSE to compare with observed frequencies from that same population. When simulations started with equal frequencies of all 18 genotypes, I compared simulated chromosomes frequencies with the observed frequencies in each of the three populations (CA, NC, and NY). I retained the 1,000 fitness arrays that produce proto-sex chromsome frequencies with the lowest MSE for a given dominance model, population, and starting genotype frequency. I refer to each of these 1,000 fitness arrays as the best-fitting arrays for each population.

Simulations with finite population sizes

I assessed how well the 1,000 best-fitting fitness arrays for each population, starting frequency, and dominance model maintain polygenic sex determination when population sizes are finite. Finite population sizes introduce stochasticity to the change in allele frequencies across generations (i.e., genetic drift), in contrast to the deterministic effects of constant fitness values in a population of infinite size (Hartl and Clark 2007). It is not possible to calculate an expected probability of fixation or time to fixation for alleles when there is complex sex-linked inheritance, as in the house fly polygenic sex determination system (Meisel et al. 2016). To overcome this limitation, I used simulations that modeled finite populations by including binomial sampling each generation.

I simulated finite populations by adding one additional step to the simulations described above in order to model a population size (N) of 10,000 individuals (Figure 1B). This population size was chosen because it is small enough to capture the effects of drift within the time scale of my simulations. Each simulation was started with the frequencies of the proto-sex chromosomes observed in the population where the fitness array was identified as best-fitting. In each generation, following multiplication by the fitness array (i.e., natural selection), I used a binomial sampling process to calculate a new frequency for each genotype. Binomial sampling for each genotype was performed with 10,000 trials (i.e., $N = 10^4$) and a probability of success equal to the frequency of the genotype (genotype frequencies were those after selection). The resulting array of 18 genotype frequencies was then divided by the sum of all values to ensure that the genotype frequencies sum to 1. This array of genotype frequencies (after selection and binomial sampling) was next used as input into the same recursion equations described above to calculate genotype frequencies after random mating (Supplementary Table S5). The process (selection, binomial sampling, and random mating) was repeated for 1,000 generations. I simulated 100 replicate finite populations for each fitness array. I then used the frequency with

314 which fixation and loss occur in those 100 replicates as an estimate of the probability of fixation
315 and loss of proto-sex chromosomes within 1,000 generations.
316 I determined a null expectation for fixation or loss of proto-sex chromosomes in finite
317 populations by using simulations without selection (i.e., genetic drift and no fitness differences
318 across genotypes). To those ends, I performed simulations with 10,000 individuals for 1,000
319 generations. I performed 1,000 replicate simulations with starting values at the observed
320 frequencies of the proto-sex chromosomes in each population (CA, NC, and NY). These drift-
321 only simulations included the same steps as the simulations with both selection and drift, except
322 that there is no differential survival (i.e., natural selection) step in the drift-only simulation. After
323 1,000 generations, I calculated the frequency of each proto-Y and proto-W chromosome (Y^M,
324 III^M, and IV^F), as well as the frequency with which each chromosome was lost, across the 1,000
325 simulations.

326 **Results**

327 I used forward population genetic simulations to assess how natural selection can
328 maintain polygenic sex determination in a species with two proto-Y chromosomes (Y^M and III^M)
329 and one proto-W chromosome (IV^F), modeled after the house fly system (Hamm et al. 2015).
330 Simulations were performed assuming either additive, dominant, recessive, or overdominant
331 fitness effects of the Y^M and III^M proto-Y chromosomes. I first identified fitness arrays that
332 produced frequencies of each proto-sex chromosome (Y^M, III^M, IV^F, and their corresponding
333 homologs) that are all >0.1% after 1,000 generations. I refer to these as fitness arrays that
334 maintain polygenic sex determination, but not necessarily at the frequencies observed in natural
335 populations. I next identified the 1,000 fitness arrays that produce frequencies of each proto-Y
336 and proto-W chromosome (Y^M, III^M, and IV^F) that are closest to those observed in each of three
337 natural populations (CA, NC, and NY). These are referred to as the 1,000 best-fitting arrays. To
338 determine null expectations, I considered 1,000 randomly chosen fitness arrays, regardless of
339 the frequencies of the proto-sex chromosomes after 1,000 generations.
340 Simulations were started with four possible arrays of genotype frequencies. The first
341 three arrays of starting genotype frequencies are based on the frequencies of Y^M, III^M, and IV^F
342 observed in each of the three populations (CA, NC, or NY). In each of these populations, Y^M is
343 at a higher frequency than III^M (Hamm and Scott 2008; Scott et al. 2013; Meisel et al. 2016).
344 The fourth starting genotype array has equal frequencies of all 18 genotypes (i.e., each at a
345 frequency of 1/18). Simulations with starting frequencies based on those observed in NC or NY
346 and recessive fitness effects of Y^M and III^M caused populations to go extinct because of the loss

of one sex. Therefore, results for recessive fitness effects starting with NC or NY genotype frequencies were not obtained. For that reason, I report results from simulations starting with equal genotype frequencies in the main text, and those with starting frequencies based on the natural populations are given in the Supplementary Materials. For the most part, results are largely the same regardless of the starting genotype frequencies, but I highlight when the results differ meaningfully. It can be assumed that the results are the same if I do not explicitly say that they differ.

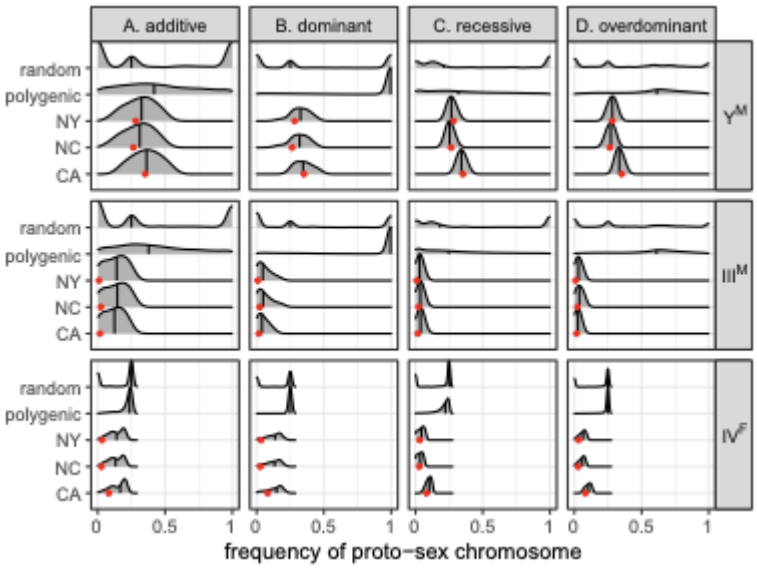


Figure 2. Smoothed histograms show the frequency of each proto-sex chromosome (Y^M, III^M, and IV^F) after 1,000 generations in simulations using either 1,000 random fitness arrays (random), fitness arrays that maintain polygenic sex determination (polygenic), or the 1,000 best-fitting fitness arrays for each population (CA, NC, or NY). Simulations were started with equal frequencies of all genotypes. The vertical line within each histogram shows the median. Red dots show the observed proto-sex chromosome frequencies in each natural population. Fitness arrays were calculated assuming either (**A)** additive, **(B)** dominant, **(C)** recessive, or **(D)** overdominant fitness effects of the proto-Y chromosomes

Dominance of fitness effects and the maintenance of polygenic sex determination

The probability with which fitness arrays maintain polygenic sex determination is affected by the dominance of the effects of the Y^M and III^M proto-Y chromosomes (Supplementary Figures S1A, S2A). For example, when fitness effects of proto-Y chromosomes are additive, less than 1% of all fitness arrays maintain polygenic sex determination with a frequency of all proto-sex chromosomes >0.1%. In contrast, when fitness effects of the proto-Y chromosomes are dominant or recessive, ~20% of fitness arrays maintain polygenic sex determination. Overdominant fitness effects are the most likely (>40%) to maintain polygenic sex determination

371 with proto-sex chromosomes at a frequency >0.1%. These results are consistent when
372 simulations are started with equal genotype frequencies or frequencies found in natural
373 populations (Supplementary Figures S1A, S2A).
374 The dominance of fitness effects of the proto-Y chromosomes also affects how similar
375 the proto-sex chromosomes frequencies are to those observed in natural populations. Additive
376 and recessive fitness effects that maintain polygenic sex determination result in frequencies of
377 Y^M, III^M, and IV^F that are more similar to those observed in natural populations (i.e., lower MSE
378) than proto-sex chromosome frequencies produced by overdominant fitness effects; the proto-
379 sex chromosome frequencies produced by dominant fitness effects are the most divergent from
380 those in natural populations (Figure 2). However, when I select the 1,000 best-fitting genotypic
381 fitness arrays (i.e., those with the lowest MSE), the relative distributions of MSE for different
382 dominance models are dramatically shifted (Supplementary Figures S1A, S2A). Recessive and
383 overdominant fitness effects that produce the best-fitting genotypic fitness values generally
384 produce proto-sex chromosome frequencies more similar to those observed in natural
385 populations than additive or dominant fitness effects (Figure 2). These results are not affected
386 by the starting genotype frequencies used in the simulations (Figure 2; Supplementary Figures
387 S1, S2, S3).
388 Fitness arrays that maintain polygenic sex determination produce different frequencies
389 of proto-sex chromosomes depending on the dominance of fitness effects. For example, when
390 additive, recessive, or overdominant fitness effects maintain polygenic sex determination, there
391 is a broad range of frequencies at which the proto-Y chromosomes (Y^M and III^M) are maintained
392 (Figure 2). In contrast, most dominant fitness effects that maintain polygenic sex determination
393 result very high frequencies of the proto-Y chromosomes (Figure 2B). Fitness arrays based on
394 dominant effects that produce high frequency proto-Y chromosomes correspond to a large
395 number of fitness arrays that maintain polygenic sex determination and have a high MSE
396 (Supplementary Figures S1, S2). Randomly chosen fitness arrays, in comparison, produce
397 three distinct proto-Y chromosome frequency classes: 0 (loss of the proto-Y, because a different
398 XY or ZW system has taken over), 1 (fixation of the proto-Y, corresponding to fixation of the ZW
399 system), and 0.25 (the expectation for a monogenic XY system). These results are consistent
400 regardless of the starting genotype frequencies (Supplementary Figure S3).
401 The high frequency of proto-Y chromosomes when fitness effects are dominant likely
402 reflects a path to fixation rather than a stable polymorphism. A proto-Y chromosome with a
403 dominant fitness effect segregating at very high frequency is consistent with a beneficial effect
404 that masks the recessive allele, allowing the relatively deleterious recessive allele to persist at

405 very low frequency (Hartl and Clark 2007). Ultimately, the proto-Y chromosome with a dominant
406 beneficial effect will reach fixation, although it appears to take longer than the 1,000 generations
407 considered in my simulations. Therefore, most of the cases where dominant fitness effects
408 maintain polygenic sex determination, under the criteria I have defined, are not in fact conditions
409 where polygenic sex determination is maintained at equilibrium. However, there is a small
410 number of fitness arrays based on dominant effects of proto-Y chromosomes that maintain
411 polygenic sex determination at frequencies far from fixation, including those that most closely
412 resemble the frequencies observed in natural populations (Figure 2B).
413 In the majority of fitness arrays that maintain polygenic sex determination, regardless of
414 the dominance of the proto-Y chromosome, IV^F is almost exclusively at a frequency of 25%
415 (Figure 2). This is the expected frequency of a W chromosome in a randomly mating population
416 with ZW sex determination. This pattern holds for all starting genotype frequencies used in the
417 simulations (Supplementary Figure S3). Therefore, in a system with 2 proto-Y chromosomes
418 and one proto-W, polygenic sex determination is expected to be maintained by the (near)
419 fixation of the ZW female genotype. In contrast, *Md-tra*^{D} (the female-determining allele on the
420 IV^F chromosome) is at a frequency <25% in the three natural populations under consideration
421 (Hamm and Scott 2008; Scott et al. 2013; Meisel et al. 2016). A subset of fitness arrays in my
422 simulations maintain IV^F at <25% for all dominance models, including those that produce proto-
423 sex chromosome frequencies most similar to those observed in natural populations (Figure 2;
424 Supplementary Figure S3).

425 ***Sexual antagonism and the maintenance of polygenic sex determination***

426 I next inspected if there is a relationship between the fitness effects of proto-sex
427 chromosomes in males and females. Each of the eight male proto-sex chromosome genotypes
428 has a corresponding female genotype that differs only because females carry a copy of the IV^F
429 chromosome and males do not (Table 1). There are two additional female genotypes (XX; III/III;
430 IV/IV and X/X; III/III; IV^F/IV) that do not have a corresponding male genotype because they do
431 not have a proto-Y chromosome. I calculated Spearman's rank order correlation of fitness
432 between males and females (ρ_{MF}) for the eight pairs of male and female genotypes in each
433 fitness array. I specifically tested for a negative intersexual fitness correlation ($\rho_{MF} < 0$), which
434 is a hallmark of sexually antagonistic genetic variation (Chippindale et al. 2001; Rice and
435 Chippindale 2001; Bonduriansky and Chenoweth 2009).
436 I confirmed a previous finding that additive fitness effects of Y^M and III^M that produce
437 proto-sex chromosome frequencies most similar to those observed in natural house fly

populations have $\rho_{MF} < 0$ (Meisel et al. 2016). For example, the single best-fitting additive fitness effects for the CA population result in genotype fitness values that are negatively correlated between males and females (Figure 3A). In other words, the genotype that maximizes male fitness results in the lowest female fitness (Y^M/Y^M; III^M/III^M), and as female fitness increases across genotypes, male fitness decreases. The genotype that produces the highest female fitness (X/X; III/III; IV/IV^F) has no corresponding male genotype because that genotype does not have any proto-Y chromosomes.

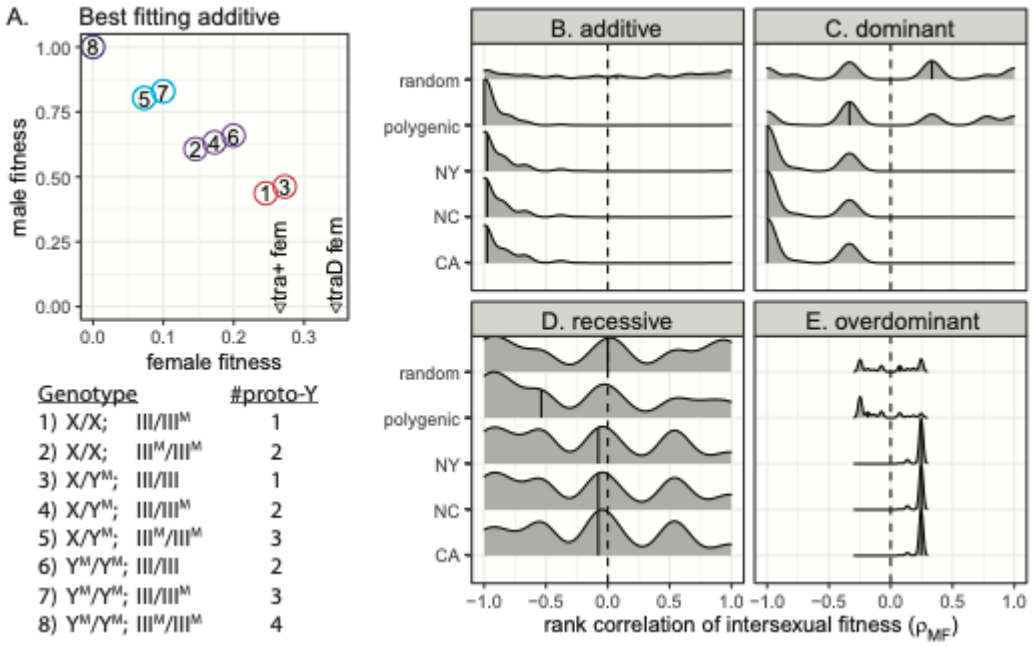


Figure 3. Intersexual fitness correlations (ρ_{MF}) are plotted for genotype fitness arrays used in simulations started with equal frequencies of all genotypes. **(A)** Male and female fitness values are plotted for the 8 proto-Y chromosome genotypes found in both males and females for the additive fitness effects that produce proto-sex chromosome frequencies most similar to those observed in the CA population. Numbers in the graph correspond to genotypes shown below the graph. Genotypes with the same color circle have the same number of proto-Y chromosomes (#proto-Y). The fitness of two genotypes only found in females (X/X; III/III) are shown on the X-axis, with their genotype at *Md-tra* indicated by whether they carry IV^F (traD) or two wild-type copies of *Md-tra* (tra+). **(B–E)** Smoothed histograms show ρ_{MF} for the 1,000 best-fitting genotypic fitness arrays for the CA, NC, and NY populations. Distributions of ρ_{mf} are also shown for all fitness arrays that maintain polygenic sex determination, as well as 1,000 random fitness arrays. Dashed vertical lines show $\rho_{MF} = 0$, and solid vertical lines within histograms show the median.

460 Negative ρ_{MF} is a feature of nearly all additive fitness effects that maintain polygenic
461 sex determination. Almost all of the 1,000 best-fitting genotypic fitness arrays resulting from
462 additive effects of the proto-Y chromosomes in all three populations (CA, NC, and NY) have
463 have $\rho_{MF} < 0$ (Figure 3B). In addition, nearly all additive fitness effects that maintain polygenic
464 sex determination, regardless how similar the proto-sex chromosome frequencies are to those
465 in natural populations, have $\rho_{MF} < 0$ (Figure 3B). There is a negative ρ_{MF} for additive fitness
466 effects that maintain polygenic sex determination regardless of the genotype frequencies used
467 to start the simulations (Supplementary Figure S4A). In contrast, when I selected 1,000 random
468 additive fitness effects of proto-Y chromosomes (i.e., fitness values that do not necessarily
469 maintain polygenic sex determination), ρ_{MF} is uniformly distributed between -1 and 1
470 (Figure 3B). This demonstrates that $\rho_{MF} < 0$ is not an intrinsic property of additive fitness
471 effects. Instead, $\rho_{MF} < 0$ is a unique feature of additive fitness effects that maintain polygenic
472 sex determination.
473 Dominant fitness effects of proto-Y chromosomes that produce the best-fitting genotype
474 frequencies also tend to have $\rho_{MF} < 0$ (Figure 3C). I observe $\rho_{MF} < 0$ for the best-fitting
475 fitness arrays based on dominant effects regardless of the genotype frequencies used to start
476 the simulations (Supplementary Figure S4B). In contrast, when I consider dominant fitness
477 effects that maintain polygenic sex determination without producing proto-sex chromosome
478 frequencies similar to those in natural populations, ρ_{MF} is not as negative as in the best-fitting
479 fitness arrays (Figure 3C). However, most of these dominant fitness effects that appear to
480 maintain polygenic sex determination after 1,000 generiations are more likely to be in the
481 process of driving the proto-Y chromosomes to fixation (see above). Random fitness arrays
482 produced with dominant fitness effects do not tend to have negative ρ_{MF}, demonstrating that
483 $\rho_{MF} < 0$ is not an intrinsic property of dominant fitness effects. Therefore, $\rho_{MF} < 0$ is only
484 expected for dominant fitness effects of proto-Y chromosomes that maintain polygenic sex
485 determination with genotype frequencies similar to those observed in natural populations.
486 Recessive and overdominant fitness effects that maintain polygenic sex determination,
487 on the other hand, do not tend to have $\rho_{MF} < 0$. There is no clear relationship between male
488 and female fitness (i.e., the average $\rho_{MF} \approx 0$) for recessive fitness effects that produce the
489 best-fitting proto-sex chromosome frequencies in each natural population (Figure 3D;
490 Supplementary Figure S4C). However, recessive fitness effects that maintain polygenic sex
491 determination, regardless of the fit of proto-sex chromosome frequencies, tend to have
492 $\rho_{MF} < 0$ (Figure 3D), although this depends on the starting genotype frequencies used in the

simulations (Supplementary Figure S4C). Overdominant fitness effects that produce the best-fitting genotype fitness arrays nearly all have $\rho_{MF} > 0$ (Figure 3E; Supplementary Figure S4D), but this result is difficult to interpret because overdominant effects produce a concave relationship between male genotype and fitness (i.e., heterozygotes are defined as the most fit genotype). Therefore, a simple rank order correlation (i.e., ρ_{MF}) does not accurately capture the relationship between male and female fitness when the proto-Y chromosomes have overdominant effects in males.

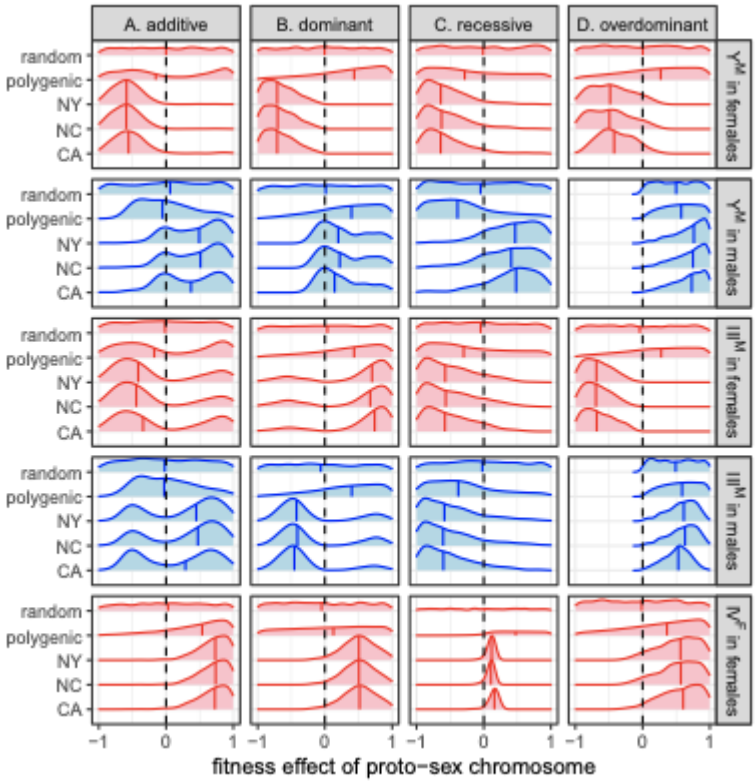


Figure 4. Smoothed histograms show the distributions of fitness effects of each proto-sex chromosome in each sex for 1,000 random fitness arrays (random), fitness arrays that maintain polygenic sex determination (polygenic), or the 1,000 best-fitting fitness arrays for each population (CA, NC, or NY). Simulations were started with equal frequencies of all genotypes. The vertical line within each histogram shows the median, and dashed lines in each panel are at fitness value of 0. Fitness arrays were calculated assuming either (**A**) additive, (**B**) dominant, (**C**) recessive, or (**D**) overdominant fitness effects of the proto-Y chromosomes.

Negative intersexual fitness correlations can be caused by opposing effects of proto-Y chromosomes in males and females

The negative ρ_{MF} that I observe when additive or dominant fitness effects maintain polygenic sex determination (Figure 3B–C) are suggestive of sexually antagonistic selection. A

513 potential cause of $\rho_{MF} < 0$ is sexually antagonistic effects of the proto-Y chromosomes if, for
514 example, Y^M and III^M carry male-beneficial and female-detrimental alleles (Fisher 1931; Rice
515 1987). Indeed, the best-fitting additive effects in the CA population are consistent with such a
516 sexually antagonistic model. In the best-fitting array, male fitness is highest in the genotype
517 carrying 4 proto-Y chromosomes (Y^M/Y^M; III^M/III^M), and male fitness decreases as the number of
518 proto-Y chromosomes in the genotype decreases (Figure 3A). In contrast, female fitness
519 increases as the number of proto-Y chromosomes in the genotype decreases. This opposing
520 correlation between the number of proto-Y chromosomes and fitness across males and females
521 is consistent with male-beneficial and female-detrimental effects of the proto-Y chromosomes.
522 I find that opposing effects of proto-Y chromosomes on male and female fitness are a
523 common feature when additive fitness effects produce proto-sex chromosome frequencies
524 similar to those observed in house fly populations. The additive fitness effects that produce the
525 best-fitting proto-sex chromosome frequencies for all three natural populations (CA, NC, and
526 NY) tend to have proto-Y chromosomes that decrease female fitness and increase male
527 fitness—i.e., the average fitness effects of Y^M and III^M are negative in females and positive in
528 males (Figure 4A). To test if these fitness effects result in opposing intersexual fitness, I
529 calculated Spearman's rank order correlation between the number of proto-Y chromosomes
530 across genotypes and their sex-specific fitness within each array of 8 genotypes with at least
531 one proto-Y (ρ_Y). The best-fitting additive fitness effects for all three natural populations tend to
532 have a positive ρ_Y for male fitness and negative ρ_Y for female fitness (Figure 5A), consistent
533 with sexually antagonistic (male-beneficial and female detrimental) effects of the proto-Y
534 chromosomes. With 8 genotypes per sex and 4 possible values for the number of proto-Y
535 chromosomes, ρ_Y can take 8 possible values (4 positive and 4 negative). I counted the number
536 of fitness arrays with ρ_Y greater than or equal to one of the 4 positive values and the number of
537 fitness arrays with a ρ_Y equal to or less than the equivalent negative value. I then compared
538 those counts between the best-fitting arrays and either the random fitness arrays or all arrays
539 that maintain polygenic sex determination. In nearly all cases, there is a significant excess of
540 positive ρ_Y for the best-fitting male fitness values and negative ρ_Y for the best-fitting female
541 fitness values (Supplementary Table S6). I observe these opposing correlations regardless of
542 the genotype frequencies used to start the simulations (Supplementary Figures S5A, S6A;
543 Supplementary Table S7).
544

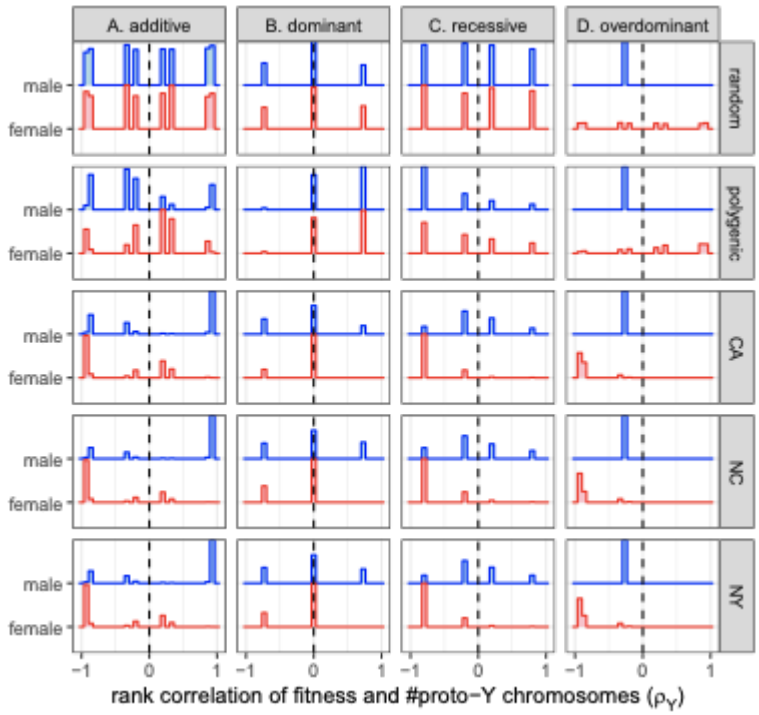


Figure 5. Histograms show the distributions of Spearman's rank order correlation (ρ_Y) between the number of proto-Y chromosomes (Y^M or III^M) and fitness in either males (blue) or females (red). Correlations are calculated for the 1,000 best-fitting fitness arrays that produce proto-sex chromosomes most similar to those observed in natural populations (CA, NC, or NY), fitness arrays that maintain polygenic sex determination (polygenic), or 1,000 random fitness arrays (random). Simulations were started with equal frequencies of all genotypes. Dashed vertical lines indicate $\rho_Y = 0$. Genotype fitness was determined assuming either **(A)** additive, **(B)** dominant, **(C)** recessive, or **(D)** overdominant fitness effects of the proto-Y chromosomes.

When I consider all additive effects that maintain polygenic sex determination, regardless of how well they fit natural populations, there does not tend to be opposing fitness effects of proto-Y chromosomes in males and females. For example, the average fitness effects of Y^M and III^M trend toward 0 in both females and males in fitness arrays that maintain polygenic sex determination (Figure 4A). In addition, there is not a consistent positive or negative ρ_Y for either male or female fitness (Figure 5A). The lack of a relationship between proto-Y chromosomes and sex-specific fitness effects for all additive models that maintain polygenic sex determination holds regardless of the starting genotype frequencies used in the simulations (Supplementary Figures S4A, S5A). Similarly, there is not a relationship between proto-Y chromosomes and sex-specific fitness for 1,000 random fitness arrays produced by additive fitness effects (Figure 4A, Figure 5A; Supplementary Figures S5A, S6A). Therefore, proto-Y chromosomes with additive fitness effects are only predicted to be sexually antagonistic if polygenic sex determination is maintained with proto-sex chromosome frequencies observed in

567 natural house fly populations. This is notable because $\rho_{MF} < 0$ when additive effects maintain
568 polygenic sex determination regardless of how well proto-sex chromosome frequencies fit
569 natural populations (Figure 3B). This result suggests that genotypes can be sexually
570 antagonistic when polygenic sex determination is maintained by additive fitness effects without
571 sexually antagonistic effects of the proto-Y chromosomes themselves. The lack of sexually
572 antagonistic effects of the proto-Y chromosomes themselves suggests that combinations of
573 proto-Y chromosomes create genotypes with opposing fitness in males and females.
574 When dominant fitness effects of proto-Y chromosomes maintain polygenic sex
575 determination at proto-sex chromosomes frequencies observed in natural populations, ρ_{MF}
576 tends to be negative (Figure 3C). The negative ρ_{MF} suggests that sexually antagonistic
577 selection is involved in maintaining polygenic sex determination when the proto-Y chromosomes
578 have dominant fitness effects. Consistent with this hypothesis, Y^M tends to be deleterious in
579 females when dominant fitness effects maintain polygenic sex determination at proto-sex
580 chromosome frequencies observed in natural populations (Figure 4B). However, III^M does not
581 tend to have a negative fitness effect in females (Figure 4B). Moreover, ρ_Y is usually 0 in both
582 males and females when dominant fitness effects maintain polygenic sex determination
583 (Figure 5B). This is true regardless of the starting genotype frequencies used in the simulations
584 (Supplementary Figures S5B, S6B). This provides further evidence that polygenic sex
585 determination can be maintained with sexually antagonistic genotypes when the underlying
586 fitness effects of the proto-sex chromosomes are not sexually antagonistic.
587 Recessive fitness effects of proto-Y chromosomes that maintain polygenic sex
588 determination do not tend to have $\rho_{MF} < 0$, although there are some exceptions (Figure 3D).
589 This suggests that sexually antagonistic selection is not involved in maintaining polygenic sex
590 determination when proto-Y chromosomes have recessive fitness effects. It is therefore
591 surprising that both Y^M and III^M tend to be deleterious in females when polygenic sex
592 determination is maintained by recessive fitness effects, regardless of how well the proto-sex
593 chromosome frequencies fit those observed in natural populations (Figure 4C). Moreover, Y^M
594 tends to be beneficial to males when proto-sex chromosome frequencies are similar to those
595 observed in natural populations (Figure 4C). As expected based on the deleterious effects in
596 females, recessive fitness effects that maintain polygenic sex determination tend to have
597 $\rho_Y < 0$ for females (Figure 5C). In contrast, random recessive fitness effects do not tend to be
598 female-deleterious and do not produce $\rho_Y < 0$ (Figure 4C, Figure 5C), demonstrating that
599 female-deleterious effects and $\rho_Y < 0$ are not intrinsic properties of recessive fitness effects.
600 These patterns are the same regardless of the starting genotype frequencies used in the

601 simulations (Supplementary Figures S5C, S6C). Therefore, while genotypes are not sexual
602 antagonistic when polygenic sex determination is maintained by recessive fitness effects, the
603 proto-Y chromosomes are expected to have female-detrimental effects. The lack of sexually
604 antagonistic genotypes occurs because the proto-Y chromosomes do not have consistent male-
605 beneficial effects (Figure 4C, Figure 5C).

606 When overdominant fitness effects in males maintain polygenic sex determination with
607 proto-sex chromosome frequencies most similar to those observed in natural house fly
608 populations, the proto-Y chromosomes tend to have deleterious effects in females (Figure 4D).
609 This results in $\rho_Y < 0$ for females when there are overdominant effects of the proto-Y
610 chromosomes in males (Figure 5D). There is also a significant excess of $\rho_Y < 0$ in females for
611 the best fitting fitness arrays relative to both all fitness arrays that maintain polygenic sex
612 determination and random arrays (Supplementary Table S8). In contrast, when I consider all
613 fitness arrays that can maintain polygenic sex determination with overdominant effects
614 (regardless of how well they fit natural populations), the proto-Y chromosomes do not
615 necessarily have deleterious effects in females (Figure 4D) and ρ_Y is usually not negative in
616 females (Figure 5D). The same patterns are observed for all starting genotype frequencies used
617 in the simulations (Supplementary Figures S5D, S6D; Supplementary Table S9).

618 ***Female-beneficial effects of the proto-W chromosome***

619 The IV^F proto-W chromosome is nearly always female-beneficial in fitness arrays that
620 maintain polygenic sex determination (Figure 4). I observe the female-beneficial effect of IV^F in
621 all cases when polygenic sex determination is maintained, regardless of the the dominance of
622 fitness effects of the proto-Y chromosomes, whether or not proto-sex chromosomes frequencies
623 are similar to those in natural populations, and independently of the genotype frequencies used
624 to start the simulations (Figure 4; Supplementary Figure S5). There is also a positive correlation
625 between the fitness effect and frequency of IV^F when proto-Y chromosomes have dominant,
626 recessive, or overdominant effects (Figure 6), regardless of the starting genotype frequencies
627 used in the simulations (Supplementary Figure S7). A positive correlation indicates that the
628 more female-beneficial the IV^F chromosome, the higher its frequency.

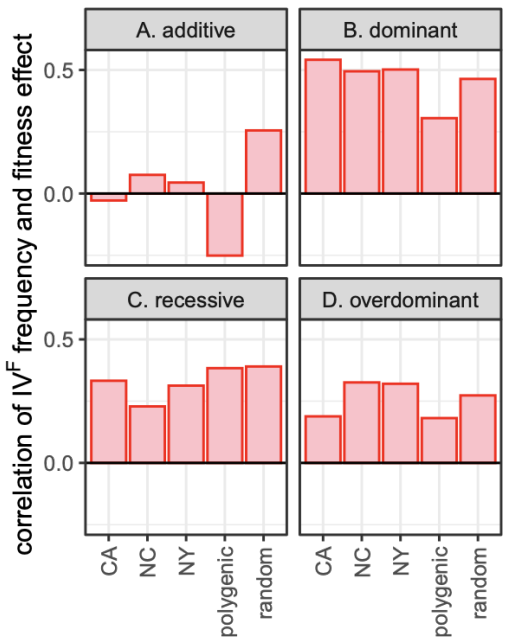


Figure 6. Bar plots show Spearman's rank order correlation between the frequency the IV^F proto-W chromosome across simulated populations and its fitness effect in the populations. Simulated populations include those that best-fit proto-sex chromosome frequencies in natural populations (CA, NC, or NY), those that maintain polygenic sex determination (polygenic), and 1,000 random populations (random). Simulations were started with equal frequencies of all genotypes. Genotype fitness in populations was calculated assuming **(A)** additive, **(B)** dominant, **(C)** recessive, or **(D)** overdominant effects of the proto-Y chromosomes.

When additive fitness effects maintain polygenic sex determination, there is not necessarily a positive correlation between the fitness effect and frequency of IV^F (Figure 6A) or the correlation is less than in other dominance scenarios (Supplementary Figure S7A). The reduced correlation is somewhat surprising because IV^F is female-beneficial when additive fitness effects maintain polygenic sex determination (Figure 4A). The negative or reduced correlation between IV^F fitness and frequency may be caused by the female-deleterious effects of proto-Y chromosomes when additive effects maintain polygenic sex determination (Figure 4A, Figure 5A). When the proto-Y chromosomes have female-deleterious effects, there will be selection against females carrying IV^F and at least one proto-Y. Selection against females that carry a female-detrimental proto-Y could explain why the correlation between IV^F frequency is negative or low when fitness effects are additive.

Selection pressures can maintain polygenic sex determination in finite populations

I next examined how well selection pressures can maintain polygenic sex determination when population sizes are finite. I used the best-fitting selection pressures that I had identified as producing proto-sex chromosome frequencies most similar to those observed in the three natural house fly populations (CA, NC, and NY) with each of the four starting genotype frequencies (CA, NC, NY, and equal). I performed 100 replicate forward simulations for each of the best-fitting selection pressures, with each simulation consisting of 1,000 generations and 10,000 individuals. Simulations using finite populations were started with genotype frequencies observed in natural populations (regardless of how the best-fitting array was identified) because

665 I am specifically interested in the maintenance of proto-sex chromosome polymorphisms in finite
666 populations, and not on the approach to the observed frequencies from arbitrary values.
667 I used the simulations of finite populations to estimate the probability that proto-sex
668 chromsomes are lost (or fix) via genetic drift in spite of selection pressures that are expected to
669 maintain polygenic sex determination. To those ends, I calculated the proportion of simulations
670 (out of 100) in which a given proto-sex chromosome reached fixation for each fitness array. I
671 used this proportion as an estimate of the probability of fixation (P_{fix}). The P_{fix} value for a
672 given proto-X or proto-Z chromosome (i.e., X, III, or IV) is equal to the probability of loss (P_{loss})
673 of the homologous proto-Y or proto-W (Y^M, III^M, or IV^F), and vice versa. For example, P_{fix} of Y^M
674 is equal to P_{loss} of the X chromosome. I used Fisher's exact test to determine if the number of
675 fixations of a proto-sex chromosome is in excess or deficiency when compared to a finite
676 population of the same size and without selection (i.e., genetic drift only).
677 Selection pressures that produce proto-sex chromsome frequencies similar to those
678 observed in natural populations (i.e., the best-fitting arrays) promote the maintenance of the III^M
679 and IV^F polymorphism relative to the expectation under genetic drift alone. The standard third
680 chromosome without *Mdmd* (III) tends to have a lower P_{fix} with the best-fitting selection
681 pressures than under drift alone, and the III^M chromosome does not reach fixation at rates that
682 deviate from neutrality (Figure 7). Similarly, the standard fourth chromosome without *Md-tra*^{D}
683 (IV) also tends to have a lower P_{fix} with the best-fitting selection pressures than under drift
684 alone (Figure 7). The IV^F proto-W chromosome cannot reach fixation because it is not possible
685 to create ZW males given the house fly sex determination system. The maintenance of the III^M
686 and IV^F polymorphisms in finite populations is observed for all four dominance models and
687 regardless of how the best-fitting arrays are identified (Supplementary Figure S8).
688 The frequency of a sex chromosome in natural populations affects its P_{fix}. For
689 example, the X and Y^M chromosomes rarely reach fixation, regardless of the selection
690 pressures or population (Supplementary Figures S9, S10). Y^M is at a higher frequency than III^M
691 and IV^F across all three populations (Meisel et al. 2016), and the high frequency of Y^M is likely
692 responsible for its low P_{loss} within 1,000 generations. However, for a small fraction of the best-
693 fitting arrays (<10%), P_{fix} for both the X and Y^M chromosomes is greater than under drift alone
694 (Figure 7). The slightly elevated P_{fix} of the X and Y^M chromosomes in finite populations is
695 observed for all four dominance models and regardless of how the best-fitting arrays are
696 identified (Supplementary Figure S8). Therefore, when fixation or loss is unlikely via drift
697 (because the proto-Y chromosome is at high frequency), selection pressures that maintain

polygenic sex determination do not decrease P_{fix} or P_{loss} of a proto-sex chromosome in a finite population.

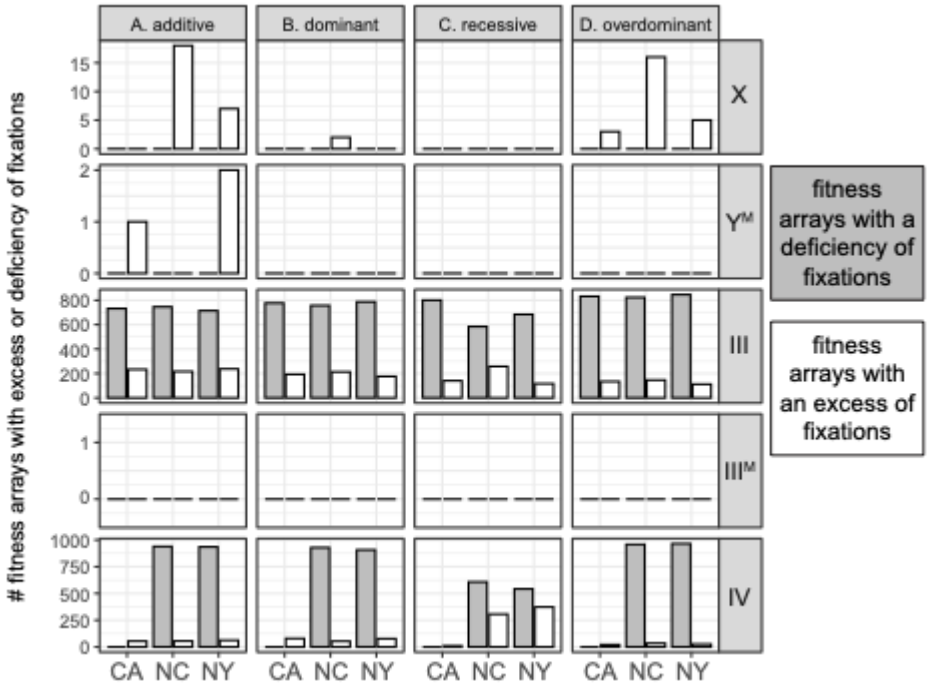


Figure 7. Bar plots show the number of fitness arrays that result in a deficiency (gray) or excess (white) of fixations of each proto-sex chromosome (X, Y^M, III, III^M, or IV) relative to simulations in which there are no fitness differences across proto-sex chromosomes (i.e., drift only). The fitness arrays are the 1,000 best-fitting fitness values for each population (CA, NC, or NY) when simulations were started with equal frequencies of all genotypes. Fitness effects of the proto-Y chromosomes are either **(A)** additive, **(B)** dominant, **(C)** recessive, or **(D)** overdominant.

When a proto-sex chromosome is at low frequency, selection pressures that maintain polygenic sex determination decrease P_{fix} and P_{loss} in finite populations. For example, III^M is rare in all three populations, and it has the largest P_{loss} of any proto-sex chromosome (Supplementary Figures S9, S10). In turn, selection pressures that maintain polygenic sex determination decrease P_{fix} of chromosome III, thereby decreasing P_{loss} if III^M (Figure 7). Furthermore, selection pressures that push III^M and IV^F to higher frequencies when population sizes are infinite also decrease P_{fix} and P_{loss} in finite populations (Supplementary Figures S11, S12). Therefore, when fixation is likely via drift because a proto-sex chromosome is at low frequency (such as III^M and IV^F), the selection pressures that maintain polygenic sex determination with a higher frequency of the rare proto-sex chromosome decrease P_{loss} in a finite population.

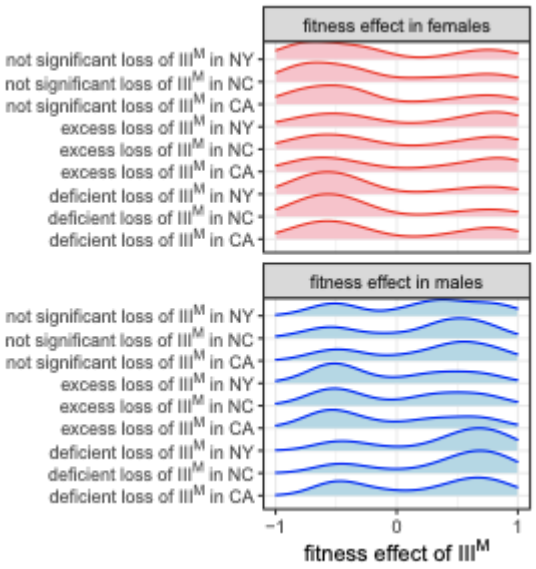


Figure 8. Smoothed histograms show the distributions of the additive effect of III^M in females and males for fitness arrays that have a lower P_{loss} of III^M than drift alone (deficient), greater P_{loss} (excess), or not significantly different from drift. Fitness effects are from the best-fitting arrays in CA, NC, or NY determined using simulations started with equal genotype frequencies.

I finally tested if sexually antagonistic selection affects P_{loss} of III^M in a finite population. I focus on III^M because it has the highest P_{loss} of any proto-sex chromosome, allowing for comparisons across fitness effects with high and low rates of loss. I compared additive effects across fitness arrays with differing rates of P_{loss} (excess, deficient, and not significantly different from drift alone) because additive effects have the strongest signatures of sexually antagonistic selection (Figure 3, Figure 4, Figure 5). When additive effects decrease P_{loss} of III^M relative to drift, III^M tends to confer a fitness cost to females (i.e., the fitness effect is negative) and benefit to males (Figure 8). The fitness cost to females and benefit to males when P_{loss} of III^M is low are significantly greater than when P_{loss} is high or not significantly different from drift alone (P<0.05 in a Mann-Whitney test for all populations). These opposing fitness effects in males and females are consistent with sexually antagonistic selection. Therefore, III^M is more likely to be maintained in a finite population when additive fitness effects are sexually antagonistic. I observe these sexually antagonistic effects regardless of how the best-fitting fitness arrays are identified (Supplementary Figure S13).

Discussion

I used forward simulations of a complex polygenic sex determination system—with two proto-X/Y pairs and one proto-Z/W, modeled after the house fly—to investigate how the dominance of alleles under selection affects the maintenance of polygenic sex determination. I found that the ability of natural selection to maintain polygenic sex determination does indeed

751 depend on the dominance of fitness effects. For example, polygenic sex determination is more
752 likely to be maintained if proto-Y chromosomes have overdominant fitness effects, and least
753 likely if the fitness effects are additive (Supplementary Figures S1, S2). Moreover, the role of
754 sexually antagonistic selection in the maintenance of polygenic sex determination also depends
755 on the dominance of fitness effects (Figure 3, Figure 4, Figure 5). Despite these differences
756 across dominance models, selection pressures that are predicted to maintain polygenic sex
757 determination are equally effective at preventing the fixation or loss of proto-sex chromosomes
758 via genetic drift regardless of the dominance of fitness effects (Figure 7).

759 ***Additive sexually antagonistic fitness effects maintain polygenic sex determination***

760 Additive fitness effects are unlikely (<1%) to maintain polygenic sex determination, and,
761 when they do, the proto-sex chromosome frequencies are often divergent from those observed
762 in natural populations (Figure 2A). When additive fitness effects maintain polygenic sex
763 determination, they do so with signatures of sexual antagonism. For example, additive effects
764 that maintain polygenic sex determination have negative intersexual correlations of genotype
765 fitness, regardless of whether proto-sex chromosome frequencies are similar to those observed
766 in natural house fly populations (Figure 3B). Furthermore, when fitness effect are additive and
767 proto-sex chromosomes frequencies are similar to natural populations, proto-Y chromosomes
768 tend to be male-beneficial and female-detrimental (Figure 4A, Figure 5A). Moreover, when I
769 model overdominant effects in males, I assume that the proto-Y chromosomes have additive
770 effects in females. When polygenic sex determination is maintained by overdominant effects in
771 males, the additive effects in females are deleterious (Figure 4D, Figure 5D). Enhanced sexual
772 antagonism is also associated with additive effects that maintain polygenic sex determination
773 when population sizes are finite. Specifically, I find that when the house fly III^M proto-Y
774 chromosome has male-beneficial and female-detrimental effects, it is more likely to be retained
775 as a polymorphism in a finite population (Figure 8). Therefore, additive fitness effects maintain
776 polygenic sex determination via sexually antagonistic selection, even if the additive effects are
777 sex-specific.

778 The role of sexually antagonistic additive fitness effects in maintaining a complex
779 polygenic sex determination system is consistent with previous work demonstrating that simpler
780 polygenic sex determination systems could also be maintained by sexually antagonistic
781 selection (Bull 1983; Rice 1986; van Doorn and Kirkpatrick 2007). These sexually antagonistic
782 effects are notable in the house fly polygenic sex determintion system because females can
783 carry one or more copies of a female-deleterious proto-Y chromosome if they also have an IV^F

784 proto-W chromosome (Table 1). The reduced fitness of these females with a proto-Y
785 chromosome likely explains the weak relationship between the fitness benefit of IV^F and its
786 frequency when fitness effects of the proto-Y chromosomes are additive (Figure 6A). These
787 hypotheses could be tested in house fly by measuring fitness-related phenotypes in females
788 carrying proto-Y chromosomes.

789 ***Dominant and recessive fitness effects that maintain polygenic sex determination have***
790 ***contrasting signatures of sexual antagonism***

791 Dominant fitness effects are unlikely to maintain polygenic sex determination, but there
792 is evidence for sexual antagonism when they do. While dominant fitness effects can maintain a
793 three proto-sex chromosome polymorphism after 1,000 generations (Supplementary Figures
794 S1, S2), most dominant fitness effects that appear to maintain polygenic sex determination are
795 likely to be paths to fixation. Nonetheless, there is small subset of dominant fitness effects that
796 maintain polygenic sex determination with proto-sex chromosomes at frequencies close to those
797 observed in natural populations (Figure 2B). In that subset, the dominant effects create
798 genotypes with opposing fitness effects in males and females (Figure 3C), consistent with
799 sexual antagonism. Despite this evidence for sexual antagonism, the proto-Y chromosomes
800 themselves are not male-beneficial and female-detrimental (Figure 4B, Figure 5B). Therefore,
801 sexual antagonism is an emergent property of genotypes that maintain polygenic sex
802 determination when fitness effects are dominant, and not an intrinsic feature of the fitness
803 effects of the proto-sex chromosomes.

804 Recessive fitness effects of proto-Y chromosomes often maintain polygenic sex
805 determination (Supplementary Figures S1, S2), and are capable of producing proto-sex
806 chromosome frequencies very similar to those observed in natural populations (Figure 2C).
807 When recessive fitness effects maintain polygenic sex determination, the genotypes are not
808 sexually antagonistic (Figure 3D). Despite no evidence for sexually antagonistic genotypes, the
809 proto-Y chromosomes are female-deleterious (Figure 4C, Figure 5C). This is a notable contrast
810 with dominant effects that maintain polygenic sex determination, where the genotypes are
811 sexually antagonistic but the proto-Y chromosomes are not. These results suggest that there
812 can be substantial decoupling of the sex-specific effects individual proto-sex chromosomes and
813 the sexually antagonistic effects of genotypes depending on the dominance of fitness effects.

814 ***Balancing selection and the maintenance of polygenic sex determination***

815 The factors that contribute to the long-term maintenance of genetic variation in natural
816 populations include recurrent mutation, population structure, and selection (Charlesworth et al.

817 2003; Mitchell-Olds et al. 2007). Natural selection that maintains genetic variation is often
818 referred to as balancing selection (Delph and Kelly 2014; Fijarczyk and Babik 2015). Balancing
819 selection can take many different forms, including overdominance (also called heterozygote
820 advantage or heterosis) and context-dependent effects. My model explicitly considers two of
821 these types of balancing selection: overdominance and sex-specific fitness effects (a specific
822 case of context-dependence).

823 I find that overdominant fitness effects of proto-Y chromosome are more likely to
824 maintain polygenic sex determination than any other dominance scenario (Supplementary
825 Figures S1, S2). Most overdominant fitness effects produce proto-sex chromosome frequencies
826 that deviate from those observed in natural populations, but a subset are very similar to those
827 seen in the three house fly populations (Figure 2D). There is conflicting evidence for the amount
828 of genetic variation with overdominant fitness effects in natural populations, including some
829 work that suggests overdominance is rare (Andrés et al. 2009; Sellis et al. 2011, 2016; Hedrick
830 2012; Bitarello et al. 2018). It therefore remains to be investigated whether there are sufficient
831 alleles with overdominant effects to explain the maintenance of polygenic sex determination in
832 natural populations.

833 Sex-specific selection, including sexual antagonism, is a type of context-dependent
834 selection that is hypothesized to maintain genetic variation (e.g., Owen 1953; Kidwell et al.
835 1977; Connallon and Clark 2014). I observe multiple different signatures of sexually antagonistic
836 selection when polygenic sex determination is maintained under a variety of dominance regimes
837 (Figure 3, Figure 4, Figure 5, Figure 8), consistent with previous theory implicating sexual
838 antagonism in the maintenance of polygenic sex determination (Bull 1983; Rice 1986; van
839 Doorn and Kirkpatrick 2007; Meisel et al. 2016). There appears to be a substantial amount of
840 sexually antagonistic genetic variation segregating within a wide range of species (Rice 1992;
841 Chippindale et al. 2001; Calsbeek and Sinervo 2004; Fedorka and Mousseau 2004; Brommer et
842 al. 2007; Foerster et al. 2007; Innocenti and Morrow 2010). However, these sexually
843 antagonistic alleles are not necessarily maintained by balancing selection—they could be
844 transient variation on the path to fixation or maintained by mutation-selection balance (Mitchell-
845 Olds et al. 2007). Indeed, the evidence that sexually antagonistic genetic variation is maintained
846 by balancing selection in natural populations is limited and conflicting (Cheng and Kirkpatrick
847 2016; Dutoit et al. 2018; Meisel 2018; Kasimatis et al. 2019; Ruzicka et al. 2019).

848 There are also theoretical predictions that X chromosomes should be enriched for
849 sexually antagonistic genetic variation (Rice 1984; Patten and Haig 2009; Patten 2019), and
850 there is some evidence in support of this theory (Gibson et al. 2002; Pischedda and Chippindale

851 2006; Foerster et al. 2007; Innocenti and Morrow 2010). The enrichment of X-linked sexually
852 antagonistic variants may depend on the dominance of their fitness effects (Fry 2010), which
853 can further affect whether male- or female-beneficial variants are favored on the X chromosome
854 (Frank and Crespi 2011; Frank and Patten 2020). The theory relating sex chromsomes and
855 sexually antagonistic alles was developed for differentiated X and Y (or Z and W)
856 chromosomes. In systems with young, homomorphic sex chromosomes, such as house fly
857 (Meisel et al. 2017), different dynamics may be at play because the X and Y carry nearly all the
858 same genes (albeit with different alleles). My results suggest that young sex chromosomes are
859 also likely to harbor sexually antagonistic alleles, and the extent of this sexually antagonistic
860 variation depends on the dominance of fitness effects. This is consistent with theory that
861 suggests sexually antagonistic selection is important for the early evolution of sex chromosomes
862 and empirical studies that identified sexually antagonistic variants on young sex chromosomes
863 (Lindholm and Breden 2002; van Doorn and Kirkpatrick 2007, 2010; Roberts et al. 2009). I
864 remains to be determined, however, whether young sex chromosomes are enriched for sexually
865 antagonistic variants, if those variants are maintained by balancing selection, and if the sexually
866 antagonistic alleles contribute to the maintenance of sex chromosome polymorphisms.
867 Investigating these problems could follow the approaches previously developed to test if
868 balancing selection maintains sexually antagonistic alleles throughout the genome (e.g., Dutoit
869 et al. 2018; Ruzicka et al. 2019).
870 Additional context-dependent effects may also be important for the maintenance of
871 polygenic sex determination. Spatially variable, temporally fluctuating, and other heterogeneous
872 selection pressures can contribute to the maintenance of genetic variation (Hedrick 2006),
873 including maintaining polygenic sex determination (Bateman and Anholt 2017). Geographical
874 variation may be important for maintaining polygenic sex determination in house fly because the
875 Y^M and III^M proto-Y chromosomes are distributed along north-south clines on multiple continents
876 (Denholm et al. 1986; Tomita and Wada 1989; Hamm et al. 2005; Kozielska et al. 2008).
877 Intriguingly, sex-specific selection, such as sexual antagonism, is predicted to vary across
878 environments under certain conditions (Connallon 2015; Connallon et al. 2019), although the
879 empirical evidence for variation in sexual antagonism across heterogenous environments is
880 mixed (Delcourt et al. 2009; Delph et al. 2011; Lasne et al. 2018). In addition, migration across a
881 species range can prevent local adaptation (Lenormand 2002; Tigano and Friesen 2016), which
882 may also affect the maintenance of alleles with sex-specific effects. Sex chromosome
883 polymorphisms, and specifically the house fly polygenic sex determination system, may be

884 promising models for investigating how sexually antagonistic variation across heterogenous
885 environments can maintain genetic variation in natural populations.

886 ***Recombination on the sex chromosomes and Y-linked alleles***

887 My model makes assumptions about recombination on the sex chromosomes that may
888 affect my conclusions about the maintenance of polygenic sex determination. Specifically, I
889 assume that there is no recombination between the male-determining locus (*Mdmd*) on each
890 proto-Y chromosome and the allele(s) under selection. I similarly assume that there is no
891 recombination between the female-determining allele (*Md-tra*^{D}) and alleles under selection on
892 the proto-W chromosome (IV^{F}). X-Y and Z-W recombination may be suppressed by
893 chromosomal inversions that create tight genetic linkage between the sex-determining locus
894 and sexually antagonistic alleles (Bergero and Charlesworth 2009; Wright et al. 2016). While
895 there is no direct evidence for inversions or other recombination suppressors on the house fly
896 sex chromosomes, male meiosis in flies is thought to occur without crossing over (Gethmann
897 1988). A lack of crossing over in males would prevent X-Y recombination in males without the
898 need for sex-chromosome-specific modifiers. However, there is evidence for male
899 recombination in house fly (Feldmeyer et al. 2010), which opens up the possibility for X-Y
900 recombination. Furthermore, in the absence of Y-linked suppressors, X-Y recombination could
901 also occur in the multiple female genotypes that carry a proto-Y chromosome (Table 1).

902 There are at least two other consequences of recombination on the sex chromosomes
903 that could affect how sex-specific selection maintains polygenic sex determination. First, my
904 model assumes that all copies of each proto-Y and proto-W chromosome have identical fitness
905 effects. This assumption is consistent with population genetics theory that predicts Y-linked
906 variants cannot segregate as protected polymorphisms when there is no X-Y recombination
907 (Clark 1987). In contrast to this prediction, there are numerous examples of Y-linked
908 polymorphisms with phenotypic effects in *Drosophila*, where the X and Y do not recombine
909 (Clark 1990; Zhang et al. 2000; Lemos et al. 2008, 2010; Griffin et al. 2015; Brown et al. 2020),
910 including Y-linked variation across heterogeneous environments over the species' geographic
911 range (Rohmer et al. 2004). Future work could incorporate variation in fitness effects of each
912 proto-Y and proto-W chromosome into my model to assess how segregating polymorphisms on
913 the proto-sex chromosomes affect the maintenance of polygenic sex determination.

914 A second consequence of sex chromosome recombination could affect how
915 overdominant fitness effects maintain polygenic sex determination. I assume that
916 overdominance arises from proto-Y chromosomes that carry recessive deleterious alleles. This

917 assumption is based on the prediction that suppressed X-Y recombination leads to the
918 accumulation of (recessive) deleterious alleles on the Y chromosome because of Muller's
919 ratchet and hitchhiking (Charlesworth and Charlesworth 2000; Bachtrog 2013). X-Y
920 recombination in males and/or females could prevent the accumulation of recessive deleterious
921 mutations on the Y^M and III^M chromosomes (Muller 1964; Felsenstein 1974). Moreover, there
922 are multiple genotypes that are homozygous for a proto-Y chromosome (Table 1), which would
923 expose recessive deleterious mutations to selection. Selection in proto-Y chromosome
924 homozygotes may purge recessive deleterious alleles from populations (Charlesworth et al.
925 1990; Barrett and Charlesworth 1991). Therefore, while overdominant fitness effects are likely to
926 maintain polygenic sex determination, proto-Y chromosomes may not posses the necessary
927 alleles for such selection pressures to act upon.

928 ***Generalizing to even more complex polygenic sex determination systems***

929 I anticipate that many of the general patterns I observe here would be similar in other
930 complex polygenic sex determination systems with at least three different proto-sex
931 chromosomes. For example, in a system with >2 proto-Y chromosomes, I expect to observe
932 similar effects of dominance on the maintenance of polygenic sex determination and sexual
933 antagonism as in the system I modeled with 2 proto-Y chromosomes. In addition, if there are
934 multiple proto-W chromosomes and a single (epistatic) proto-Y (i.e., the opposite of the house
935 fly system), I would expect similar patterns as I observe in the house fly system, but with the
936 sexes reversed. These expectations could be tested in future work. It is not clear, however,
937 what would be expected if there were multiple proto-W and multiple proto-Y chromosomes in a
938 population. It is worth noting that polygenic systems with many sex-determining loci are
939 expected to be evolutionarily unstable (Rice 1986). Zebrafish may represent a promising model
940 for combining empirical and theoretical approaches to make additional progress in the
941 maintenance of many sex determining loci (Liew et al. 2012; Wilson et al. 2014).

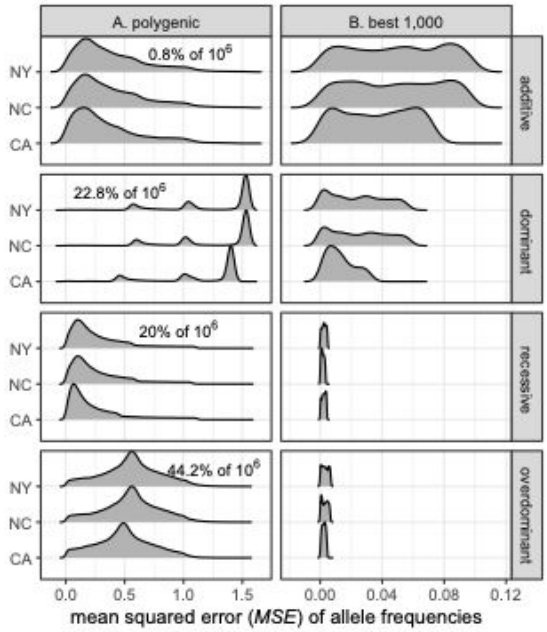
942 ***Conclusions***

943 My results demonstrate that the maintenance of polygenic sex determination depends
944 on the dominance of fitness effects, which are further predictive of the sexually antagonistic
945 effects of proto-sex chromosomes and genotypes. Notably, overdominant fitness effects are
946 most likely to maintain polygenic sex determination, but that conclusion requires at least one of
947 two important assumptions. First, genetic variation with overdominant effects must exist in
948 natural populations, or, second, proto-Y chromosomes must carry recessive deleterious alleles
949 (in addition to additive or dominant beneficial alleles). My results also suggest that sexual

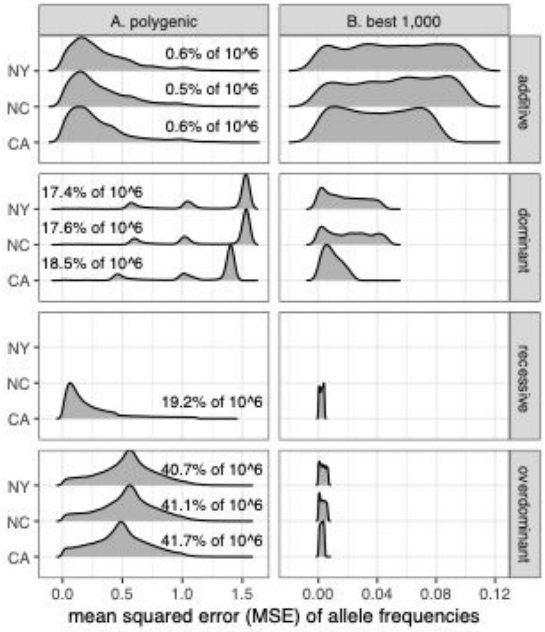
950 antagonism should be most prominent when fitness effects of proto-Y chromosomes are
951 additive, although sexually antagonistic effects are expected to a lesser degree under other
952 dominance scenarios. Lastly, future work is needed to evaluate how X-Y (and Z-W)
953 recombination and migration across demes affect the ability of sex-specific selection pressures
954 to maintain complex polygenic sex determination, including systems with other combinations of
955 proto-sex chromosomes.
956

957

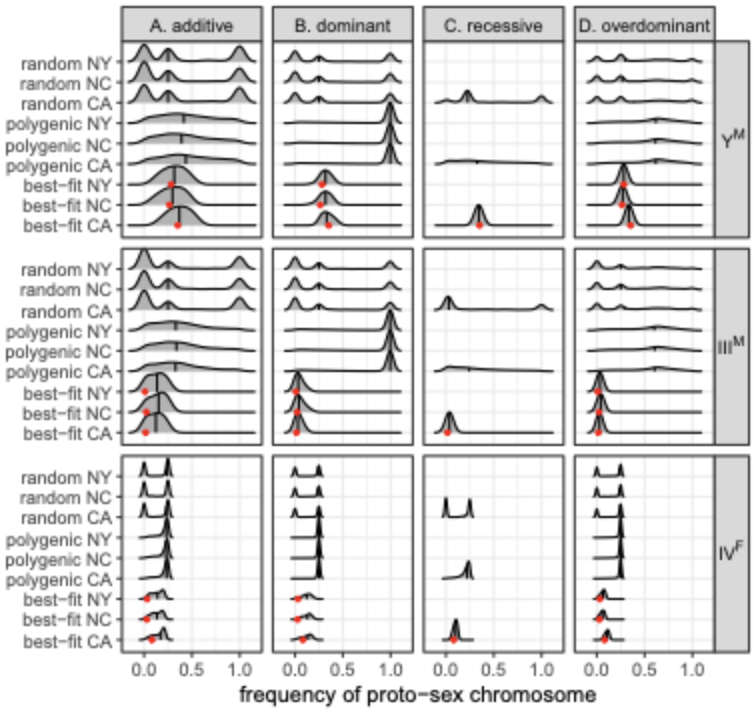
Supplementary Material



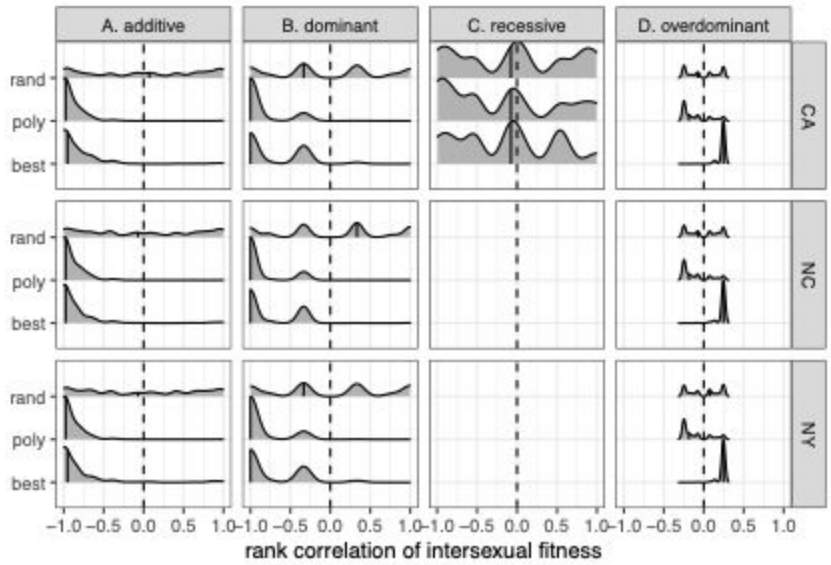
Supplementary Figure S1. Smoothed histograms show the distributions of mean squared error (MSE) values for fitness arrays that maintain polygenic sex determination (i.e., all proto-sex chromosomes at a frequency >0.1%). MSE is calculated based on the frequencies of Y^M, III^M, and IV^F found in either CA, NC, or NY. Simulations were started with equal frequencies of all genotypes. Fitness arrays are based on additive, dominant, recessive, or overdominant fitness effects of Y^M and III^M. (A) Distributions of MSE are shown for all fitness arrays that maintain polygenic sex determination, with the percent of those arrays that maintain polygenic sex determination shown in each panel. (B) Distributions of MSE are shown for the 1,000 best-fitting arrays with the lowest MSE.



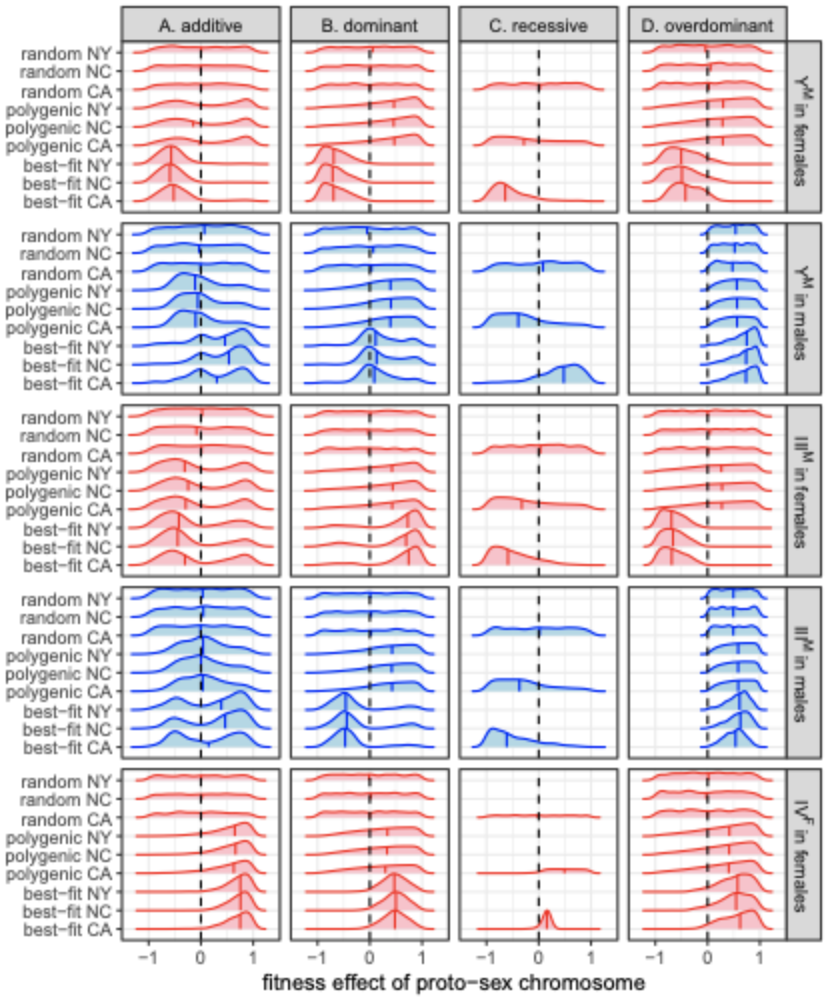
Supplementary Figure S2. Smoothed histograms show the distributions of mean squared error (MSE) values for fitness arrays that maintain polygenic sex determination (i.e., all proto-sex chromosomes at a frequency >0.1%). MSE is calculated based on the frequencies of Y^M, III^M, and IV^F found in either CA, NC, or NY. Simulations were started with genotype frequencies observed in natural populations. Fitness arrays are based on additive, dominant, recessive, or overdominant fitness effects of Y^M and III^M. (A) Distributions of MSE are shown for all fitness arrays that maintain polygenic sex determination, with the percent of those arrays that maintain polygenic sex determination shown in each panel. (B) Distributions of MSE are shown for the 1,000 best-fitting arrays with the lowest MSE.



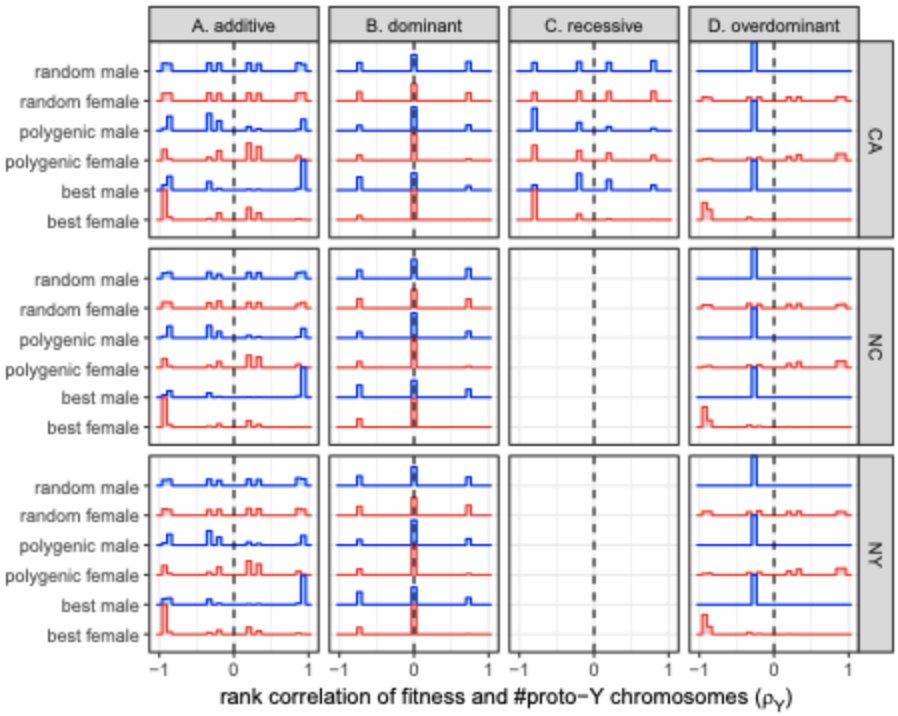
Supplementary Figure S3. Smoothed histograms show the frequency of each proto-sex chromosome (Y^M, III^M, and IV^F) after 1,000 generations in simulations using either 1,000 random fitness arrays (random), fitness arrays that maintain polygenic sex determination (polygenic), or the 1,000 best-fitting fitness arrays for each population (CA, NC, or NY). Simulations were started with genotype frequencies observed in each population. The vertical line within each histogram shows the median. Red dots show the observed proto-sex chromosome frequencies in each natural population. Fitness arrays were calculated assuming either (**A)** additive, **(B)** dominant, **(C)** recessive, or **(D)** overdominant fitness effects of the proto-Y chromosomes.



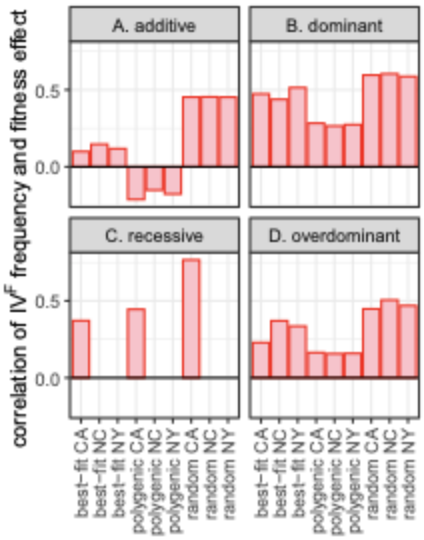
Supplementary Figure S4. Smoothed histograms show intersexual fitness correlations (ρ_{MF}) for the 1,000 best-fitting genotypic fitness arrays for the CA, NC, and NY populations, all fitness arrays that maintain polygenic sex determination (poly), and 1,000 random fitness arrays. Simulations were started with genotype frequencies observed in natural populations. Dashed vertical lines show $\rho_{MF} = 0$, and solid vertical lines within histograms show the median.



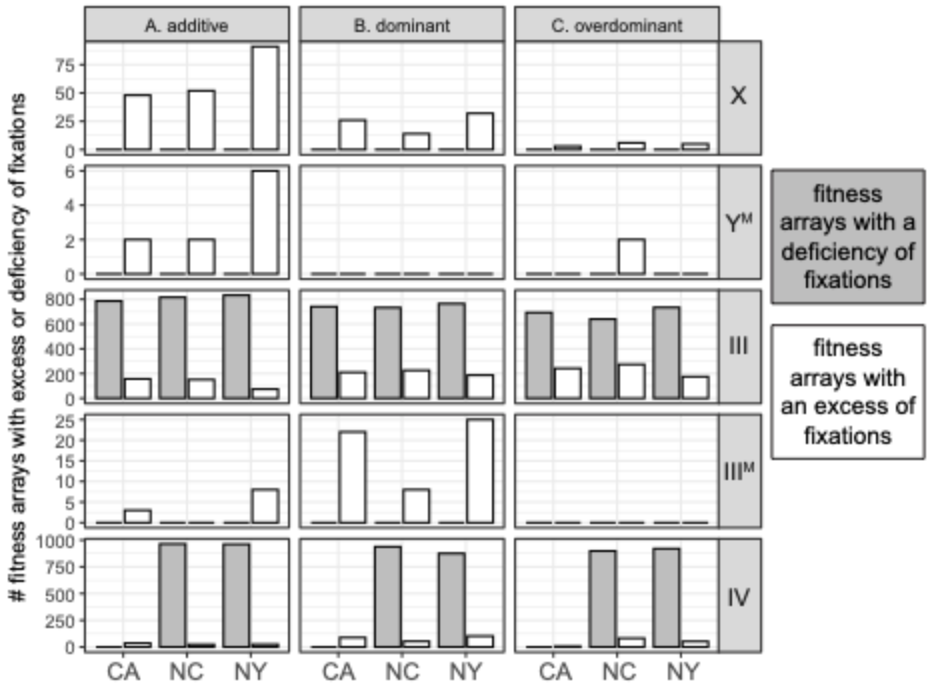
Supplementary Figure S5. Smoothed histograms show the distributions of fitness effects of each proto-sex chromosome in each sex for 1,000 random fitness arrays (random), fitness arrays that maintain polygenic sex determination (polygenic), or the 1,000 best-fitting fitness arrays for each population (CA, NC, or NY). Simulations were started with genotype frequencies observed in natural populations. The vertical line within each histogram shows the median, and dashed lines in each panel are at fitness value of 0. Fitness arrays were calculated assuming either (**A)** additive, **(B)** dominant, **(C)** recessive, or **(D)** overdominant fitness effects of the proto-Y chromosomes.



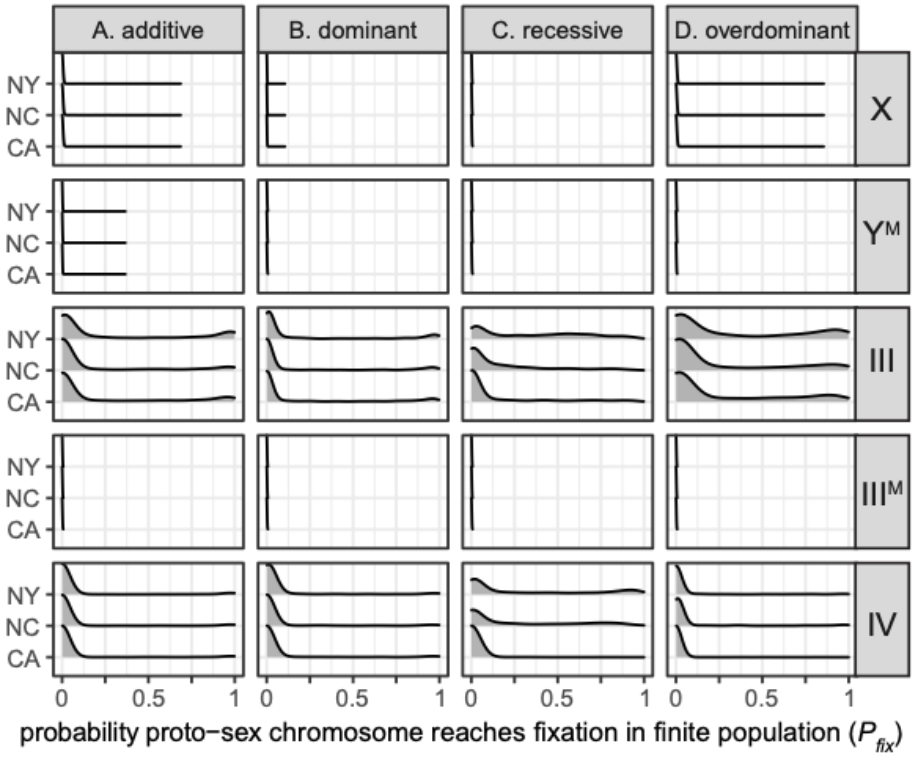
Supplementary Figure S6. Histograms show the distributions of Spearman's rank order correlation (ρ_Y) between the number of proto-Y chromosomes (Y^M or III^M) and fitness in either males (blue) or females (red). Correlations are calculated for the 1,000 best-fitting fitness arrays that produce proto-sex chromosomes most similar to those observed in natural populations (CA, NC, or NY), fitness arrays that maintain polygenic sex determination (polygenic), or 1,000 random fitness arrays (random). Simulations were started with genotype frequencies observed in natural populations. Dashed vertical lines indicate $\rho_Y = 0$. Genotype fitness was determined assuming either **(A)** additive, **(B)** dominant, **(C)** recessive, or **(D)** overdominant fitness effects of the proto-Y chromosomes.



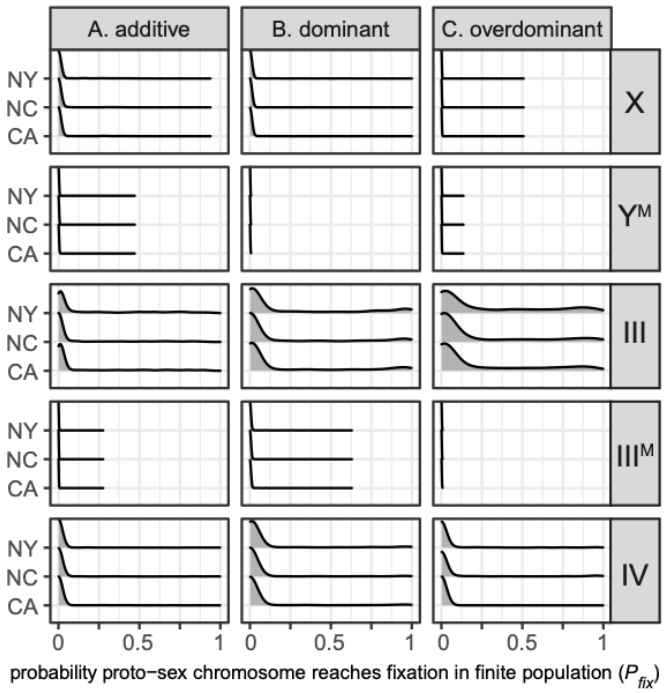
Supplementary Figure S7. Bar plots show Spearman's rank order correlation between the frequency the IV^F proto-W chromosome across simulated populations and its fitness effect in the populations. Simulated populations include those that best-fit proto-sex chromosome frequencies in natural populations (CA, NC, or NY), those that maintain polygenic sex determination (polygenic), and 1,000 random populations (random). Simulations were started with equal frequencies of all genotypes. Genotype fitness in populations was calculated assuming **(A)** additive, **(B)** dominant, **(C)** recessive, or **(D)** overdominant effects of the proto-Y chromosomes.



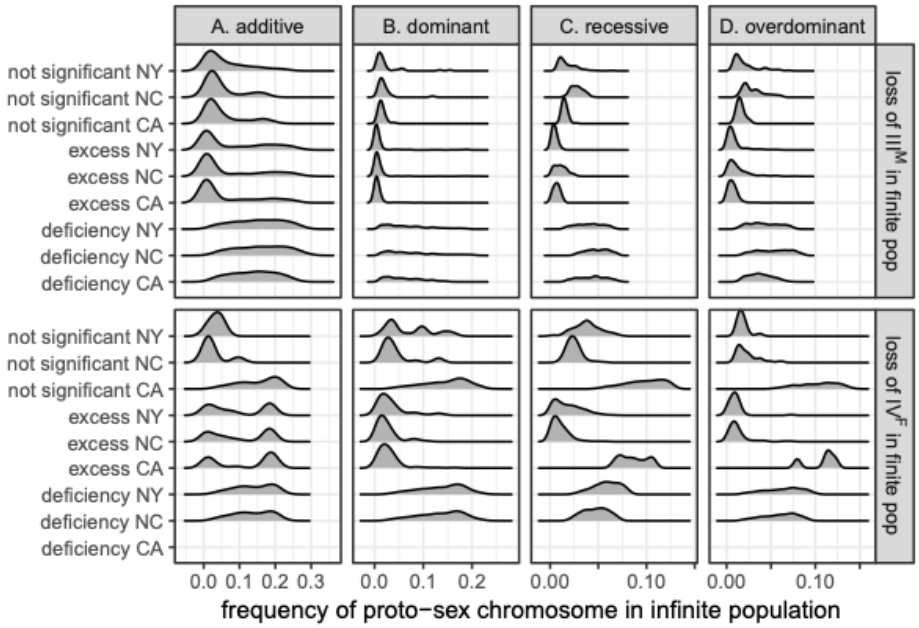
Supplementary Figure S8. Bar plots show the number of fitness arrays that result in a deficiency (gray) or excess (white) of fixations of each proto-sex chromosome (X, Y^M, III, III^M, or IV) relative to simulations in which there are no fitness differences across proto-sex chromosomes (i.e., drift only). The fitness arrays are the 1,000 best-fitting fitness values for each population (CA, NC, or NY) when simulations were started with genotype frequencies observed in natural populations. Fitness effects of the proto-Y chromosomes are either **(A)** additive, **(B)** dominant, or **(C)** overdominant. Recessive fitness effects are not shown because simulations could not be run for NC and NY.



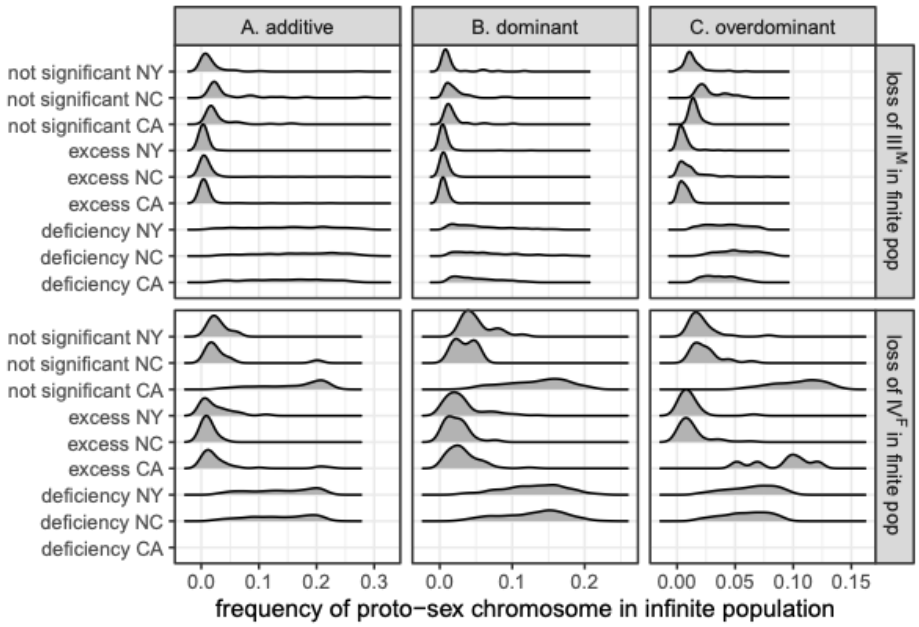
Supplementary Figure S9. Smoothed histograms show the distributions of probability of fixation for each proto-sex chromosome in each pouplation when $N = 10^4$. Simulations were performed with the 1,000 best-fitting selection pressures for each population (CA, NC, or NY) that were identified from simulations started with equal frequencies of all genotypes, using either **(A)** additive, **(B)** dominant, **(C)** recessive, or **(D)** overdominant fitness effects of the proto-Y chromosomes.



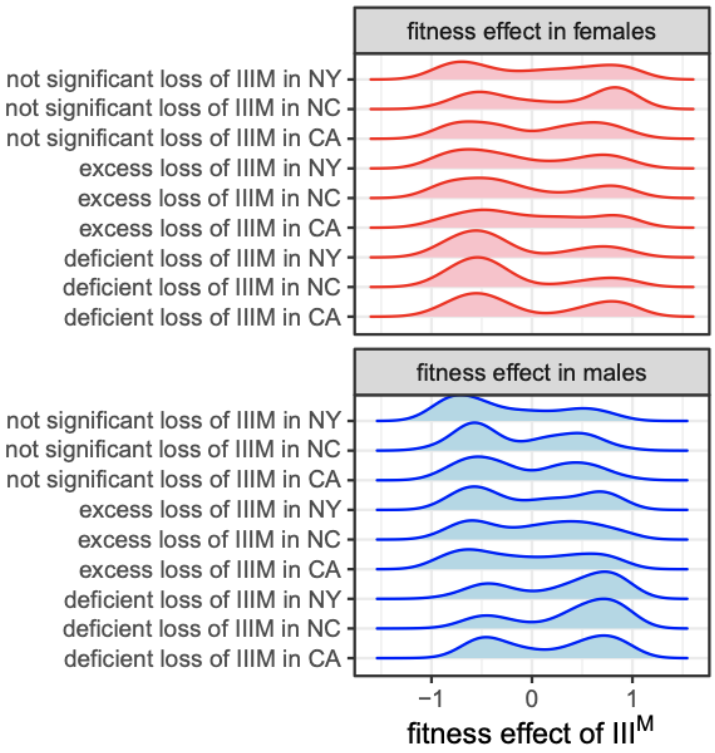
Supplementary Figure S10. Smoothed histograms show the distributions of probability of fixation for each proto-sex chromosome in each pouplation when $N = 10^4$. Simulations were performed with the 1,000 best-fitting selection pressures for each population (CA, NC, or NY) that were identified from simulations started with genotype frequencies observed in each population using either **(A)** additive, **(B)** dominant, or **(C)** overdominant fitness effects of the proto-Y chromosomes. Recessive fitness effects are not shown because simulations could not be run for NC and NY.



Supplementary Figure S11. Smoothed histograms show the distributions of the frequency of the III^M and IV^F chromosomes after 1,000 generations in an infinite population for fitness effects that result in a deficiency, excess, or not significant loss of III^M and IV^F relative to drift alone. Fitness effects are from the 1,000 best-fitting selection pressures in each population (CA, NC, or NY) that were identified from simulations started with equal frequencies of all genotypes, using either **(A)** additive, **(B)** dominant, **(C)** recessive, or **(D)** overdominant fitness effects of the proto-Y chromosomes.



Supplementary Figure S12. Smoothed histograms show the distributions of the frequency of the III^M and IV^F chromosomes after 1,000 generations in an infinite population for fitness effects that result in a deficiency, excess, or not significant loss of III^M and IV^F relative to drift alone. Fitness effects are from the 1,000 best-fitting selection pressures in each population (CA, NC, or NY) that were identified from simulations started with genotype frequencies observed in each population, using either **(A)** additive, **(B)** dominant, or **(C)** overdominant fitness effects of the proto-Y chromosomes. Recessive fitness effects are not shown because simulations could not be run for NC and NY.



Supplementary Figure S13. Smoothed histograms show the distributions of the additive effect of III^M in females and males for fitness arrays that have a lower P_{loss} of III^M than drift alone (deficient), greater P_{loss} (excess), or not significantly different from drift. Fitness effects are from the best-fitting arrays in CA, NC, or NY determined using simulations started with genotype frequencies observed in each population.

Supplementary Table S1. Genotype fitness with additive effects of proto-Y chromosomes.

	genotype	additive fitness effects
f_1	X/X; III/III; IV/IV	1
f_2	X/X; III/III; IV/IV^F	$1 + F_{IVF}$
f_3	X/X; III/III^M; IV/IV^F	$1 + F_{IIIM} + F_{IVF}$
f_4	X/X; III^M/III^M; IV/IV^F	$1 + 2F_{IIIM} + F_{IVF}$
f_5	X/Y^M; III/III; IV/IV^F	$1 + F_Y + F_{IVF}$
f_6	X/Y^M; III/III^M; IV/IV^F	$1 + F_Y + F_{IIIM} + F_{IVF}$
f_7	X/Y^M; III^M/III^M; IV/IV^F	$1 + F_Y + 2F_{IIIM} + F_{IVF}$
f_8	Y^M/Y^M; III/III; IV/IV^F	$1 + 2F_Y + F_{IVF}$
f_9	Y^M/Y^M; III/III^M; IV/IV^F	$1 + 2F_Y + F_{IIIM} + F_{IVF}$
f_{10}	Y^M/Y^M; III^M/III^M; IV/IV^F	$1 + 2F_Y + 2F_{IIIM} + F_{IVF}$
m_1	X/X; III/III^M; IV/IV	$1 + M_{IIIM}$
m_2	X/X; III^M/III^M; IV/IV	$1 + 2M_{IIIM}$
m_3	X/Y^M; III/III; IV/IV	$1 + M_Y$
m_4	X/Y^M; III/III^M; IV/IV	$1 + M_Y + M_{IIIM}$
m_5	X/Y^M; III^M/III^M; IV/IV	$1 + M_Y + 2M_{IIIM}$
m_6	Y^M/Y^M; III/III; IV/IV	$1 + 2M_Y$
m_7	Y^M/Y^M; III/III^M; IV/IV	$1 + 2M_Y + M_{IIIM}$
m_8	Y^M/Y^M; III^M/III^M; IV/IV	$1 + 2M_Y + 2M_{IIIM}$

Supplementary Table S2. Genotype fitness with dominant effects of proto-Y chromosomes.

	genotype	dominant fitness effects
f_1	X/X; III/III; IV/IV	1
f_2	X/X; III/III; IV/IV^F	$1 + F_{IVF}$
f_3	X/X; III/III^M; IV/IV^F	$1 + F_{IIIM} + F_{IVF}$
f_4	X/X; III^M/III^M; IV/IV^F	$1 + F_{IIIM} + F_{IVF}$
f_5	X/Y^M; III/III; IV/IV^F	$1 + F_Y + F_{IVF}$
f_6	X/Y^M; III/III^M; IV/IV^F	$1 + F_Y + F_{IIIM} + F_{IVF}$
f_7	X/Y^M; III^M/III^M; IV/IV^F	$1 + F_Y + F_{IIIM} + F_{IVF}$
f_8	Y^M/Y^M; III/III; IV/IV^F	$1 + F_Y + F_{IVF}$
f_9	Y^M/Y^M; III/III^M; IV/IV^F	$1 + F_Y + F_{IIIM} + F_{IVF}$
f_{10}	Y^M/Y^M; III^M/III^M; IV/IV^F	$1 + F_Y + F_{IIIM} + F_{IVF}$
m_1	X/X; III/III^M; IV/IV	$1 + M_{IIIM}$
m_2	X/X; III^M/III^M; IV/IV	$1 + M_{IIIM}$
m_3	X/Y^M; III/III; IV/IV	$1 + M_Y$
m_4	X/Y^M; III/III^M; IV/IV	$1 + M_Y + M_{IIIM}$
m_5	X/Y^M; III^M/III^M; IV/IV	$1 + M_Y + M_{IIIM}$
m_6	Y^M/Y^M; III/III; IV/IV	$1 + M_Y$
m_7	Y^M/Y^M; III/III^M; IV/IV	$1 + M_Y + M_{IIIM}$
m_8	Y^M/Y^M; III^M/III^M; IV/IV	$1 + M_Y + M_{IIIM}$

Supplementary Table S3. Genotype fitness with recessive effects of proto-Y chromosomes.

	genotype	recessive fitness effects
f_1	X/X; III/III; IV/IV	1
f_2	X/X; III/III; IV/IV^F	$1 + F_{IVF}$
f_3	X/X; III/III^M; IV/IV^F	$1 + F_{IVF}$
f_4	X/X; III^M/III^M; IV/IV^F	$1 + F_{IIIM} + F_{IVF}$
f_5	X/Y^M; III/III; IV/IV^F	$1 + F_{IVF}$
f_6	X/Y^M; III/III^M; IV/IV^F	$1 + F_{IVF}$
f_7	X/Y^M; III^M/III^M; IV/IV^F	$1 + F_{IIIM} + F_{IVF}$
f_8	Y^M/Y^M; III/III; IV/IV^F	$1 + F_Y + F_{IVF}$
f_9	Y^M/Y^M; III/III^M; IV/IV^F	$1 + F_Y + F_{IVF}$
f_{10}	Y^M/Y^M; III^M/III^M; IV/IV^F	$1 + F_Y + F_{IIIM} + F_{IVF}$
m_1	X/X; III/III^M; IV/IV	1
m_2	X/X; III^M/III^M; IV/IV	$1 + M_{IIIM}$
m_3	X/Y^M; III/III; IV/IV	1
m_4	X/Y^M; III/III^M; IV/IV	1
m_5	X/Y^M; III^M/III^M; IV/IV	$1 + M_{IIIM}$
m_6	Y^M/Y^M; III/III; IV/IV	$1 + M_Y$
m_7	Y^M/Y^M; III/III^M; IV/IV	$1 + M_Y$
m_8	Y^M/Y^M; III^M/III^M; IV/IV	$1 + M_Y + M_{IIIM}$

Supplementary Table S4. Genotype fitness with overdominant effects of proto-Y chromosomes in males (additive effects in females).

	genotype	overdominant fitness effects
f_1	X/X; III/III; IV/IV	1
f_2	X/X; III/III; IV/IV^F	$1 + F_{IVF}$
f_3	X/X; III/III^M; IV/IV^F	$1 + F_{IIIM} + F_{IVF}$
f_4	X/X; III^M/III^M; IV/IV^F	$1 + 2F_{IIIM} + F_{IVF}$
f_5	X/Y^M; III/III; IV/IV^F	$1 + F_Y + F_{IVF}$
f_6	X/Y^M; III/III^M; IV/IV^F	$1 + F_Y + F_{IIIM} + F_{IVF}$
f_7	X/Y^M; III^M/III^M; IV/IV^F	$1 + F_Y + 2F_{IIIM} + F_{IVF}$
f_8	Y^M/Y^M; III/III; IV/IV^F	$1 + 2F_Y + F_{IVF}$
f_9	Y^M/Y^M; III/III^M; IV/IV^F	$1 + 2F_Y + F_{IIIM} + F_{IVF}$
f_{10}	Y^M/Y^M; III^M/III^M; IV/IV^F	$1 + 2F_Y + 2F_{IIIM} + F_{IVF}$
m_1	X/X; III/III^M; IV/IV	$1 + M_{IIIM}$
m_2	X/X; III^M/III^M; IV/IV	1
m_3	X/Y^M; III/III; IV/IV	$1 + M_Y$
m_4	X/Y^M; III/III^M; IV/IV	$1 + M_Y + M_{IIIM}$
m_5	X/Y^M; III^M/III^M; IV/IV	$1 + M_Y$
m_6	Y^M/Y^M; III/III; IV/IV	1
m_7	Y^M/Y^M; III/III^M; IV/IV	$1 + M_{IIIM}$
m_8	Y^M/Y^M; III^M/III^M; IV/IV	1

Supplementary Table S5. Recursion equations to simulate random mating.

genotype	frequency	frequency after random mating
X/X; III/III; IV/IV	f_1	$f_1m_1/2 + f_1m_3/2 + f_1m_4/4$ $+ f_2m_1/4 + f_2m_3/4 + f_2m_4/8$ $+ f_3m_1/4 + f_3m_3/8 + f_3m_4/16$ $+ f_5m_1/8 + f_5m_3/8 + f_5m_4/16$ $+ f_6m_1/16 + f_6m_3/16 + f_6m_4/32$
X/X; III/III; IV/IV^F	f_2	$f_2m_1/4 + f_2m_3/4 + f_2m_4/8$ $+ f_3m_1/8 + f_3m_3/8 + f_3m_4/16$ $+ f_5m_1/8 + f_5m_3/8 + f_5m_4/16$ $+ f_6m_1/16 + f_6m_3/16 + f_6m_4/32$
X/X; III/III^M; IV/IV^F	f_3	$f_2m_1/4 + f_2m_2/2 + f_2m_4/8 + f_2m_5/4$ $+ f_3m_1/4 + f_3m_2/4 + f_3m_3/8$ $+ f_3m_4/8 + f_3m_5/8$ $+ f_4m_1/4 + f_4m_3/4 + f_4m_4/8$ $+ f_5m_1/8 + f_5m_2/4 + f_5m_4/16 + f_5m_5/8$ $+ f_6m_1/8 + f_6m_2/8 + f_6m_3/16$ $+ f_6m_4/16 + f_6m_5/16$ $+ f_7m_1/8 + f_7m_3/8 + f_7m_4/16$
X/X; III^M/III^M; IV/IV^F	f_4	$f_3m_1/8 + f_3m_2/4 + f_3m_4/16 + f_3m_5/8$ $+ f_4m_1/4 + f_4m_2/2 + f_4m_4/8 + f_4m_5/4$ $+ f_6m_1/16 + f_6m_2/8 + f_6m_4/32 + f_6m_5/16$ $+ f_7m_1/8 + f_7m_2/4 + f_7m_4/16 + f_7m_5/8$
X/Y^M; III/III; IV/IV^F	f_5	$f_2m_3/4 + f_2m_4/8 + f_2m_6/2 + f_2m_7/4$ $+ f_3m_3/8 + f_3m_4/16 + f_3m_6/4 + f_3m_7/8$ $+ f_5m_1/8 + f_5m_3/4 + f_5m_4/8$ $+ f_5m_6/4 + f_5m_7/8$ $+ f_6m_1/16 + f_6m_3/8 + f_6m_4/16$ $+ f_6m_6/8 + f_6m_7/16$ $+ f_8m_1/4 + f_8m_3/4 + f_8m_4/8$ $+ f_9m_1/8 + f_9m_3/8 + f_9m_4/16$

X/Y^{M}; III/III^{M}; IV/IV^{F}	f_6	$f_2m_4/8 + f_2m_5/4 + f_2m_7/4 + f_2m_8/2$ $+ f_3m_3/8 + f_3m_4/8 + f_3m_5/8$ $+ f_3m_6/4 + f_3m_7/4 + f_3m_8/4$ $+ f_4m_3/4 + f_4m_4/8 + f_4m_6/2 + f_4m_7/4$ $+ f_5m_1/8 + f_5m_2/4 + f_5m_4/8$ $+ f_5m_5/4 + f_5m_7/8 + f_5m_8/4$ $+ f_6m_1/8 + f_6m_2/8 + f_6m_3/8 + f_6m_4/8$ $+ f_6m_5/8 + f_6m_6/8 + f_6m_7/8 + f_6m_8/8$ $+ f_7m_1/8 + f_7m_3/4 + f_7m_4/8$ $+ f_7m_6/4 + f_7m_7/8$ $+ f_8m_1/4 + f_8m_2/2 + f_8m_4/8 + f_8m_5/4$ $+ f_9m_1/4 + f_9m_2/4 + f_9m_3/8$ $+ f_9m_4/8 + f_9m_5/8$ $+ f_{10}m_1/4 + f_{10}m_3/4 + f_{10}m_4/8$
X/Y^{M}; III^{M}/III^{M}; IV/IV^{F}	f_7	$f_3m_4/16 + f_3m_5/8 + f_3m_7/8 + f_3m_8/4$ $+ f_4m_4/8 + f_4m_5/4 + f_4m_7/4 + f_4m_8/2$ $+ f_6m_1/16 + f_6m_2/8 + f_6m_4/16$ $+ f_6m_5/8 + f_6m_7/16 + f_6m_8/8$ $+ f_7m_1/8 + f_7m_2/4 + f_7m_4/8$ $+ f_7m_5/4 + f_7m_7/8 + f_7m_8/4$ $+ f_9m_1/8 + f_9m_2/4 + f_9m_4/16 + f_9m_5/8$ $+ f_{10}m_1/4 + f_{10}m_2/2 + f_{10}m_4/8 + f_{10}m_5/4$
Y^{M}/Y^{M}; III/III; IV/IV^{F}	f_8	$f_5m_3/8 + f_5m_4/16 + f_5m_6/4 + f_5m_7/8$ $+ f_6m_3/16 + f_6m_4/32 + f_6m_6/8 + f_6m_7/16$ $+ f_8m_3/4 + f_8m_4/8 + f_8m_6/2 + f_8m_7/4$ $+ f_9m_3/8 + f_9m_4/16 + f_9m_6/4 + f_9m_7/8$
Y^{M}/Y^{M}; III/III^{M}; IV/IV^{F}	f_9	$f_5m_4/16 + f_5m_5/8 + f_5m_7/8 + f_5m_8/4$ $+ f_6m_3/16 + f_6m_4/16 + f_6m_5/16$ $+ f_6m_6/8 + f_6m_7/8 + f_6m_8/8$ $+ f_7m_3/8 + f_7m_4/16 + f_7m_6/4 + f_7m_7/8$ $+ f_8m_4/8 + f_8m_5/4 + f_8m_7/4 + f_8m_8/2$ $+ f_9m_3/8 + f_9m_4/8 + f_9m_5/8$ $+ f_9m_6/4 + f_9m_7/4 + f_9m_8/4$ $+ f_{10}m_3/4 + f_{10}m_4/8 + f_{10}m_6/2 + f_{10}m_7/4$

Y^M/Y^M; III^M/III^M; IV/IV^F	f_{10}	$f_6m_4/32 + f_6m_5/16 + f_6m_7/16 + f_6m_8/8$ $+ f_7m_4/16 + f_7m_5/8 + f_7m_7/8 + f_7m_8/4$ $+ f_9m_4/16 + f_9m_5/8 + f_9m_7/8 + f_9m_8/4$ $+ f_{10}m_4/8 + f_{10}m_5/4 + f_{10}m_7/4 + f_{10}m_8/2$
X/X; III/III^M; IV/IV	m_1	$f_1m_1/2 + f_1m_2/1 + f_1m_4/4 + f_1m_5/2$ $+ f_2m_1/4 + f_2m_2/2 + f_2m_4/8 + f_2m_5/4$ $+ f_3m_1/8 + f_3m_2/4 + f_3m_3/8$ $+ f_3m_4/8 + f_3m_5/8$ $+ f_4m_1/4 + f_4m_3/4 + f_4m_4/8$ $+ f_5m_1/8 + f_5m_2/4 + f_5m_4/16 + f_5m_5/8$ $+ f_6m_1/8 + f_6m_2/8 + f_6m_3/16$ $+ f_6m_4/16 + f_6m_5/16$ $+ f_7m_1/8 + f_7m_3/8 + f_7m_4/16$
X/X; III^M/III^M; IV/IV	m_2	$f_3m_1/8 + f_3m_2/4 + f_3m_4/16 + f_3m_5/8$ $+ f_4m_1/4 + f_4m_2/2 + f_4m_4/8 + f_4m_5/4$ $+ f_6m_1/16 + f_6m_2/8 + f_6m_4/32 + f_6m_5/16$ $+ f_7m_1/8 + f_7m_2/4 + f_7m_4/16 + f_7m_5/8$
X/Y^M; III/III; IV/IV	m_3	$f_1m_3/2 + f_1m_4/4 + f_1m_6/1 + f_1m_7/2$ $+ f_2m_3/4 + f_2m_4/8 + f_2m_6/2 + f_2m_7/4$ $+ f_3m_3/8 + f_3m_4/16 + f_3m_6/4 + f_3m_7/8$ $+ f_5m_1/8 + f_5m_3/4 + f_5m_4/8$ $+ f_5m_6/4 + f_5m_7/8$ $+ f_6m_1/16 + f_6m_3/8 + f_6m_4/16$ $+ f_6m_6/8 + f_6m_7/16$ $+ f_8m_1/4 + f_8m_3/4 + f_8m_4/8$ $+ f_9m_1/8 + f_9m_3/8 + f_9m_4/16$ $+ f_5m_6/4 + f_5m_7/8$ $+ f_6m_1/16 + f_6m_3/8 + f_6m_4/16$ $+ f_6m_6/8 + f_6m_7/16$ $+ f_8m_1/4 + f_8m_3/4 + f_8m_4/8$ $+ f_9m_1/8 + f_9m_3/8 + f_9m_4/16$

X/Y^{M}; III/III^{M}; IV/IV	m_4	$f_1m_4/4 + f_1m_5/2 + f_1m_7/2 + f_1m_8/1$ $+f_2m_4/8 + f_2m_5/4 + f_2m_7/4 + f_2m_8/2$ $+f_3m_3/8 + f_3m_4/8 + f_3m_5/8$ $+f_3m_6/4 + f_3m_7/4 + f_3m_8/4$ $+f_4m_3/4 + f_4m_4/8 + f_4m_6/2 + f_4m_7/4$ $+f_5m_1/8 + f_5m_2/4 + f_5m_4/8$ $+f_5m_5/4 + f_5m_7/8 + f_5m_8/4$ $+f_6m_1/8 + f_6m_2/8 + f_6m_3/8 + f_6m_4/8$ $+f_6m_5/8 + f_6m_6/8 + f_6m_7/8 + f_6m_8/8$ $+f_7m_1/8 + f_7m_3/4 + f_7m_4/8$ $+f_7m_6/4 + f_7m_7/8$ $+f_8m_1/4 + f_8m_2/2 + f_8m_4/8 + f_8m_5/4$ $+f_9m_1/4 + f_9m_2/4 + f_9m_3/8$ $+f_9m_4/8 + f_9m_5/8$ $+f_{10}m_1/4 + f_{10}m_3/4 + f_{10}m_4/8$
X/Y^{M}; III^{M}/III^{M}; IV/IV	m_5	$f_3m_4/16 + f_3m_5/8 + f_3m_7/8 + f_3m_8/4$ $+f_4m_4/8 + f_4m_5/4 + f_4m_7/4 + f_4m_8/2$ $+f_6m_1/16 + f_6m_2/8 + f_6m_4/16$ $+f_6m_5/8 + f_6m_7/16 + f_6m_8/8$ $+f_7m_1/8 + f_7m_2/4 + f_7m_4/8$ $+f_7m_5/4 + f_7m_7/8 + f_7m_8/4$ $+f_9m_1/8 + f_9m_2/4 + f_9m_4/16 + f_9m_5/8$ $+f_{10}m_1/4 + f_{10}m_2/2 + f_{10}m_4/8 + f_{10}m_5/4$
Y^{M}/Y^{M}; III/III; IV/IV	m_6	$f_5m_3/8 + f_5m_4/16 + f_5m_6/4 + f_5m_7/8$ $+ f_6m_3/16 + f_6m_4/32 + f_6m_6/8 + f_6m_7/16$ $+ f_8m_3/4 + f_8m_4/8 + f_8m_6/2 + f_8m_7/4$ $+ f_9m_3/8 + f_9m_4/16 + f_9m_6/4 + f_9m_7/8$

Y^M/Y^M; III/III^M; IV/IV	m_7	$f_5m_4/16 + f_5m_5/8 + f_5m_7/8 + f_5m_8/4$ $+f_6m_3/16 + f_6m_4/16 + f_6m_5/16$ $+f_6m_6/8 + f_6m_7/8 + f_6m_8/8$ $+f_7m_3/8 + f_7m_4/16 + f_7m_6/4 + f_7m_7/8$ $+f_8m_4/8 + f_8m_5/4 + f_8m_7/4 + f_8m_8/2$ $+f_9m_3/8 + f_9m_4/8 + f_9m_5/8$ $+f_9m_6/4 + f_9m_7/4 + f_9m_8/4$ $+f_{10}m_3/4 + f_{10}m_4/8 + f_{10}m_6/2 + f_{10}m_7/4$
Y^M/Y^M; III^M/III^M; IV/IV	m_8	$f_6m_4/32 + f_6m_5/16 + f_6m_7/16 + f_6m_8/8$ $+f_7m_4/16 + f_7m_5/8 + f_7m_7/8 + f_7m_8/4$ $+f_9m_4/16 + f_9m_5/8 + f_9m_7/8 + f_9m_8/4$ $+f_{10}m_4/8 + f_{10}m_5/4 + f_{10}m_7/4 + f_{10}m_8/2$

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