Abstract

The heterogametic sex chromosomes (i.e. mammalian Y and avian W) do not usually recombine with the homogametic sex chromosomes which is known to lead into rapid degeneration of Y and W due to accumulation of deleterious mutations. On the other hand, some 96% of amphibian species have homomorphic, i.e. non-degenerate Y chromosomes. Nicolas Perrin’s fountain-of-youth hypothesis states that this is a result of recombination between X and Y chromosomes in sex-reversed individuals. In this study, I model the consequences of such recombination for the dynamics of a deleterious mutation occurring in Y chromosomes. As expected, even relatively low levels of sex reversal help to purge deleterious mutations. However, the population-dynamic consequences of this depend on the type of selection that operates on the population undergoing sex reversal. Under fecundity selection, sex reversal can be beneficial for some parameter values, whereas under survival selection, it seems to be always harmful.

Introduction

Some recombination needs to occur between homologous chromosomes to enable proper pairing during meiosis. In heterogametic sex chromosomes (Y or W for male or female heterogamy, respectively), this recombination occurs only in so-called pseudoautosomal regions. Typically, the pseudoautosomal regions are relatively small, and as a result of this, sex chromosomes missegregate more frequently than autosomes [1]. It is thought that large non-recombining regions have evolved because of the need to maintain linkage between sex-determining loci and other loci which are subject to sexual-antagonistic selection [2]. Presumably as a result of this, the heterogametic sex chromosomes are often ‘degenerate’, i.e. they have fewer functional genes than the homogametic sex chromosomes [3,4,5].

Charlesworth and Charlesworth [3] provide an excellent review of how the Y chromosomes degenerate. To start with, deleterious mutations occurring in the non-recombining regions of Y chromosomes are always heterozygous, and selection against them is weaker, accordingly. Secondly, the effective population size of Y chromosomes is only one third of that of X chromosomes, and one fourth of that of autosomes, and thus drift effects are more important in case of Y chromosomes. The consequences of a weakened selection and a reduced effective population size are manifold. Charlesworth and Charlesworth [3] discuss four main effects: Firstly, Muller’s ratchet [6] implies that deleterious mutations may reach fixation purely as a result of random drift, turning off one gene after another. In Y chromosomes, this fixation process is much faster than in freely recombining chromosomes. Secondly, mildly deleterious mutations may hitchhake into fixation when they are linked with beneficial mutations [7]. Thirdly, the need to purify the genome of strongly deleterious mutations may drive mildly deleterious ones into fixation, if they are in negative linkage with the more deleterious ones [8]. Finally, Hill-Robertson effects [9] may cause that new, functional variants are unable to recombine and thus drift effects are more important in case of Y chromosomes. Nicolas Perrin’s fountain-of-youth hypothesis [10] states that sex reversal can be beneficial for some parameter values, whereas under survival selection, it seems to be always harmful.
Sexes (males) occur in considerable quantities in many species of fish, amphibians and reptiles [17,18,19,20,21,22]. In fact, a pure genetic sex determination system where XY individuals are bound to be males, is a special case in the animal kingdom, and no more ‘natural’ than an environmental sex determination system [23]. In addition, the rate of recombination, both in autosomes and sex chromosomes, typically depends on the sex of the individual. This pattern is known as ‘heterochiasmy’ [24]. Specifically, there is a growing body of evidence indicating that the recombination rate depends on the phenotypic (instead of the genotypic) sex [25,26,27,28,29]. Why this is the case, is at present unknown, but differences in the physiological process of male and female meiosis have been offered as an explanation [30]. Secondly, sex-specific differences in gene regulation may play a role [10].

In this study, I provide a mathematical demonstration of Perrin’s fountain-of-youth hypothesis. To this end, I develop analytical theory by writing a dynamical system for allele frequencies in sex chromosomes of populations that are subject to sex reversal. Secondly, I test the predictions of this theory by using individual-based simulations.

**Methods**

**Modeling framework**

I consider three loci in sex chromosomes, denoted by X/Y, K/k and +/−. Locus X/Y determines the genotypic sex which can be environmentally reversed. Locus K/k is subject to sexual-antagonistic selection such that K alleles (k alleles) are subject to a selection differential 0 ≤ s ≤ 0.5 in phenotypic males (females), i.e. it is harmful to inherit alleles that are adapted for the other sex. This represents the fitness cost associated with sex reversal. Locus +/− is subject to directional selection so that − alleles are deleterious mutations having a selection differential 0 ≤ s ≤ 0.5 in both sexes. Mutation occurs by probability μ per locus per generation. Recombination occurs between X/Y and either of the other two loci by probability r per meiosis. (Here it is assumed that K/k and +/− segregate independently of each other on condition of X/Y, but see material S1.)

I assume that gene action is additive within loci, but the fitness consequences are multiplicative, so that the relative fitness of individual i is

\[
    w_i = (1 - s_i \cdot \mu_i \cdot \rho_i)(1 - \delta_i \cdot n_i)(1 - \delta_m \cdot n_m) \in [0,1].
\]

(1)

In the individual-based simulations, this is the expected relative number of offspring. Above, \(n_i\), \(n_k\) and \(n_l\) are the numbers of the respective alleles in the sex chromosomes of i, whereas \(\delta_i\) and \(\delta_m\) are the indicators of the phenotypic sexes (denoted by \(f\) and \(m\)).

Biologically, the proportions of phenotypic females in different genetic sexes (denoted by \(p_{Xf}, p_{Yf}\) and \(p_{Yf}\)) depend on each other [31]. Unless otherwise noted, it is assumed in the rest of this study that \(p = (1 - p_{Xf}) = p_{Yf}\), i.e. sex reversal is equally likely for both genotypic sexes. According to analysis of Grossen et al. [31], it is likely that \(p_{Yf} = 0\) in this case, which is assumed. This makes it possible to specify the amount of sex reversal as a scalar rate \(\rho\). \(\rho = 0\) produces a pure genetic sex-determination system, and \(\rho = 0.5\) produces a pure environmental sex-determination system. In many species, values of \(\rho\) differing significantly from zero are observed [20,23,32,33,34].

Initially, all Y chromosomes are Y\(^k\) haplotypes, and all X chromosomes are X\(^s\) haplotypes, and the genetic sex ratio is even (i.e. 25% of all sex chromosomes are Y chromosomes). Locus K/k has been subject to sexual-antagonistic differentiation, whereas the fixation of the deleterious mutation represents the degeneration of the Y chromosomes. In this study, I investigate how undergoing sex reversal helps a population to rejuvenate its Y chromosomes, i.e. to purge deleterious mutations. Except for Equation 1, this framework has been directly adopted from Perrin [10].

**Dynamics of allele frequencies**

I now consider how the allele frequencies evolve in time, given the framework stated above. To this end, I compose a system of difference equations. In this section, I give the equations, whereas discussion on their coefficient matrices is included in the material S1, and the matrices themselves are given in material S3. The key idea is that it is possible to model this biological system by using matrix algebra, because there are a finite number of haplotypes in the modeling framework. However, selection operates on the ‘phenotype’ of the individual which in this case comprises the phenotypic sex (two options, \(m\) and \(f\)), in addition to the diploid haplotype (64 options, e.g. \(X^s + Y^k\)). Therefore, it is necessary to consider the distribution of these phenotypes in each generation \(t\), denoted by \(p_t\). Two of the phenotypes are always biologically equivalent (e.g. \(mX^s + Y^k\) and \(mY^k - X^s\)), but an artificial distinction has to be made between them to model the union of gametes at a later stage (Equation 2). Out of \(p_t\), the frequencies of all alleles and the sex ratio can be calculated.

First, selection and recombination change the frequencies of the phenotypes by matrix multiplication:

\[
    p_S = W p_t,
\]

\[
    p_R = \Gamma p_S.
\]

Matrix \(W\) has values corresponding to selective pressure, and matrix \(\Gamma\) has values corresponding to probability of recombination. Then, gametes are produced. For clarity, I consider the allele frequencies of eggs and sperm separately.

\[
    p_f = X_f p_R\quad \text{for eggs, and}
\]

\[
    p_m = X_m p_R\quad \text{for sperm.}
\]

Mutation operates on \(p_f\) and \(p_m\). Matrix \(\Lambda\) has values corresponding to probability of mutation.

\[
    p_{fM} = \Lambda p_f
\]

\[
    p_{mM} = \Lambda p_m.
\]

Then, union of gametes occurs. Assuming random mating, the distribution of diploid haplotypes is given by the matrix

\[
    P = p_f M p_m^T
\]

(2)

Note that Equation 2 implies non-linearity. If this was not the case, it would be a trivial task to solve the equilibrium allele frequencies.
explicitly. The frequencies of the phenotypes on generation $t+1$ are obtained by sexing the diploid haplotypes and renormalizing the system:

$$p_{t+1} = \frac{X_{mf}(P)}{\sum_{i=1}^{128} p_{ti}}$$

As a technical convenience, I use a mapping $c$ that transforms $P$ into a vector, column by column. In the material S2, there is an R code to calculate $p_t$ given an arbitrary initial state $p_0$.

### Individual-based simulations

The above system of difference equations is essentially a model of an infinite population. Individual-based simulations are needed for three things: to test the predictions of this theory, to investigate drift effects in the allele frequencies of small populations, and to study the population-dynamic consequences of sex reversal. To this end, I consider organisms with discrete, non-overlapping generations. The life history of these organisms is simulated by the following algorithm:

1. In each generation, the phenotypic sex of each individual is randomized. Depending on the respective genotype, an...
individual is a phenotypic female by probability $p_{XXf}$, $p_{XYf}$ or $p_{YYf}$, i.e. sex reversal happens at this stage.

2. The relative fitness of each individual is calculated by using Equation 1.

3. Natural selection and breeding occur. Here, I consider two options.

a. Survival selection: individual $i$ dies with probability $1 - w_i$ before producing offspring, but each surviving female gets Poisson$(2r_0)$ offspring. A random mate is assigned for each female from the surviving males. In this case, the expected number of offspring for individual $i$ is $2r_0w_i$.

b. Fecundity selection: the total number of offspring produced depends on all $w_i$ values, but no individual dies before getting chance to reproduce. Namely, each phenotypic female $i$ is first assigned a random mate $j$ among the phenotypic males. They get Poisson$(2r_0w_iw_j)$ offspring, depending on their relative fitness values. In this case, the expected number of offspring for individual $i$ is $2r_0w_i\bar{w}_j$ (denoting the average of $w_j$ over the other sex), which is proportional to $2r_0w_i$.

4. Gametes are produced. In phenotypic females, recombination occurs by probability $r$ between each pair of loci. Mutations occur by probability $\mu$ per locus per generation in both sexes. Each offspring gets one random gamete from the sire, and one random gamete from the dam. I assume that $K$ and $k$ alleles may mutate into one other, but $-$ alleles cannot change into $+$ alleles.

5. The reproducing individuals are removed, and the procedure is carried out for a new generation. If population exceeds a carrying capacity $K = 2n_0$ after reproduction, individuals are deleted by random. This is merely a technical convenience to avoid simulating very large populations; natural selection occurs on step 3.

The R codes used for these simulations are given in the online material S2.

Parameter values and sensitivity analysis

Table 1 lists the relevant parameters. Their values are more or less arbitrary, as they are only meant to illustrate the range of factors that affect the dynamics of the $+/-$ locus. A sensitivity analysis is performed by giving each parameter higher and lower values. For $r$, a number of values from 0 to 0.50 are tried in each scenario. Apart from changing the parameter values, a few structural assumptions are tested, namely: the effect of the carrying capacity $K$, symmetric sex reversal (i.e. assumption that $p_{XYf} = 1 - p_{XYf}$), and the pattern of recombination (i.e. that segregation of $+/-$ and $K/k$ is independent on condition of $X/Y$). The results of sensitivity analysis are discussed in detail in material S1 and the accompanying Figures S1, S2, S3, S4, S5.

Shortly, as is to be expected, selective pressures $s_+$ and $s_-$ determine the advantage related to sex reversal (see below, ‘Individual-based simulations’). Secondly, the advantage related to sex reversal is lesser, if the $+/-$ locus is in a closer linkage with the sex-determining locus, yet it is still observable.
Results

Dynamics of allele frequencies

The system of equations laid out above is nonlinear, because the union of gametes (Eq. 2) follows a mass-action principle. Because of this, it is possible that multiple equilibria exist. I investigated that possibility by initializing the system with random initial values (i.e. initial frequencies $p_0$), but only one equilibrium was observed for each set of parameter values, having properties discussed below.

Initially, sex reversal creates a burst of polymorphism, rapidly generating new phenotypes (such as $fX^k + Y^k$) and haplotypes (such as $X^k$). Eventually, frequencies of some phenotypes (i.e. those with the deleterious mutation) decline, as natural selection wipes them out while the system slowly approaches the equilibrium. However, all phenotypes are maintained in populations of sufficient size, because mutation and recombination always produce them.

Figure 1 illustrates the dynamics of a few key statistics, namely the sex ratio, proportion of $Y$ chromosomes, and prevalence of the deleterious mutation in $X$ and $Y$ chromosomes. As can be seen from Figure 1, the deleterious mutation is purged so that the prevalence in $X$ chromosomes first catches up with that of $Y$ chromosomes, and then both prevalences start to approach their equilibrium value. The end result of this process is the usual mutation-selection balance, i.e. roughly $m/s$, and it does not depend on $\rho$. After an initial disturbance (barely visible in Fig. 1B), proportion of $Y$ chromosomes and the sex ratio remain constant.

This was to be expected on basis of earlier research: using difference equations, Grossen et al. [10] have shown that this is the equilibrium state for $\rho<0.5$, which they interpret as sex-ratio selection. However, this concerns the sex ratio after birth, given by $p_t$, not that of adult life stage which is given by $p_S$; as long as there are more — in $Y$ chromosomes, there are less breeding males than females in $p_S$.

Higher values of $\rho$ always imply that the mutation can be purged more rapidly. Using the parameter values in Table 1 and $\rho = 0.01 - 0.50$, it takes 167 to 452 generations for the deleterious mutation to reach the mutation-selection balance by 10^{-8} accuracy, except of the case of neutral mutation ($s_\perp = 0$) where more than 3,000 generations are required to break the linkage between — and Y. Most of the advantage is produced by relatively low values of $\rho$ (see Fig. 2). As a rule of thumb, it could be said that $\rho \approx 0.1$ purges mutations almost as effectively as a pure environmental sex-determination system ($r \approx 0.5$).

Finally, linkage disequilibrium is a necessary condition for the fountain-of-youth hypothesis to work in this modeling framework. If the system is initialized at Hardy-Weinberg equilibrium, or by even phenotypic frequencies, sex reversal does not affect the dynamics of $z = f$ locus in any way. Because no epistasis has been assumed, recombination only helps to break the linkage disequilibrium between — and $Y$. If this disequilibrium does not exist initially, recombination is not relevant, and hence sex reversal is not relevant.

**Figure 3.** Drift effects under survival selection. Drift effects are studied in populations undergoing survival selection (Option a. in the main matter), using prevalence of the deleterious mutation in $Y$ chromosomes as a summary statistic. The black line represents the theoretical value of this statistic, calculated by applying Equation 3 iteratively. The colored lines illustrate the distribution of this statistic in small populations ($n_0 = 200$ blue, $n_0 = 1,000$ green, and $n_0 = 5,000$ red). The dashed lines indicate the 0.05 and 0.95 quantiles of these distributions. The solid blue line indicates the sample mean for $n_0 = 200$. (For $n_0 = 1,000$ and $n_0 = 5,000$, the sample mean has been omitted, because it is so near to the expectation.) For each $n_0$, 150 Monte Carlo replicates have been generated. The rate of sex reversal is $\rho = 0.1$, and the other parameter values are as in ‘Baseline’, Table 1. doi:10.1371/journal.pone.0025362.g003
Individual-based simulations

The drift effects are illustrated by Figures 3 and 4, using the prevalence of the deleterious mutation in the \(Y\) chromosomes as a summary statistic. Regardless of the drift effects, even populations of the modest initial size \(n_0 = 200\) behave in great consistency with the theory developed above in ‘Dynamics of allele frequencies’. However, the drift effects are more visible for any initial population size if fecundity selection is assumed. Under survival selection, the fate of individual’s genetic material depends on its own fitness, only. Under fecundity selection, the expected number of offspring depends also on the fitness of a random mate. This creates a hitch-hike type opportunity for the deleterious mutations. In some simulation runs, it was observed that the random drift drove — back to fixation in the \(Y\) chromosomes under fecundity selection, which was never observed under survival selection.

Under fecundity selection, sex reversal has a non-uniform effect on the expected population size. This is illustrated by Figure 5. If the \(\gamma\) allele is neutral (\(\gamma = 0\)), sex reversal always decreases the expected population size, because it only breaks down the co-adaptation of the \(X/Y\) and \(K/k\) loci. For \(\gamma > 0\), there is a trade-off between this effect and the need to purify deleterious mutations. This is shown in Figure 4B by using ‘Baseline’ parameter values. In this case, an intermediate value of \(\rho\) is optimal, whereas pure genetic (\(\rho = 0\)) and pure environmental (\(\rho = 0.5\)) sex-determination systems decrease the expected population size. However, it should be added that due to demographic stochasticity, even populations with a nearly optimal \(\rho\) will sometimes drift into extinction.

It should be stressed that the advantage related to sex reversal concerns only fecundity selection (Option b.) and a suitable range of parameter values. I was unable to find parameter values for which sex reversal would have a significant positive effect on the expected population size under survival selection (Option a.). This is understandable by recalling the mechanism of survival selection: in Option a., the number of offspring depends only on the number of surviving females, as long as there is at least one male to breed with. In this case, it is presumably optimal to keep the \(\gamma\) and \(K\) alleles out of the \(X\) chromosomes, even if it means that the deleterious mutation cannot be purged.

Discussion

I have used Perrin’s framework [10] to study his fountain-of-youth hypothesis. According to theoretical considerations, environmental sex reversal helps to purge deleterious mutations out of the \(Y\) chromosomes. This was also observed in individual-based simulations for a wide range of parameters. The fitness consequences of this effect depend on the type of natural selection used in the simulation model: For populations undergoing pure survival selection, sex reversal seems to be always harmful, but for populations undergoing fecundity selection, intermediate rates of sex reversal could be optimal. Doubtless, other selection mechanisms than these two could be implemented, but the key seems to be that in survival selection, the number of males is not important, whereas in fecundity selection, male and female fitness
are equally important. In short, it is only desirable to rejuvenate the \( Y \) chromosomes, if the males matter.

The concept of sex reversal deserves to be discussed in some length. As far as this study is concerned, it comprises both environmental sex determination and sex changes before reproductive age. Bull [23] distinguishes between these two, so that in environmental sex determination, sex is permanently fixed as a result of environmental conditions, e.g. temperature, during ontogeny. Ah-King and Nylin [21] have proposed that phenotypic sex could be seen as a reaction norm. In that context, sex reversal can occur in all species, but some species are extremely insensitive to environmental conditions (genetic sex-determination system), whereas some other species are infinitely sensitive to them (environmental sex-determination system). Pure environmental and pure genetic sex-determination systems can be seen as opposite ends of a spectrum, with many species falling somewhere in between [21,31,35]. The intermediate species could still be said to have sex chromosomes, whereas pure environmental sex determination implies that no such chromosomes exist. Such is the case of many crocodilian species [22].

From the biological side, viability of the population does not imply that a trait such as sex reversal is an evolutionary advantage for the individual. This relates to the discussion about group selection [36]. In our context, turning sex reversal on in an individual-based simulation does not imply that it would evolve in nature out of the need to rejuvenate \( Y \) chromosomes. The next step would be to endogenize the evolution of the sex-determination system. This could be done by including a fourth locus in the present framework, one that determines liability for sex reversal, and studying its allele frequencies. Perrin also hints at this [10].

Finally, this study is insufficient to judge the truth of Perrin’s hypothesis. It is almost self-evident that sex reversal helps to purify deleterious mutations out of \( Y \) chromosomes, if recombination of sex chromosomes occurs only in phenotypic females. I have only verified the quantitative consequences of this notion. The evolutionary consequences are more complicated, as they depend on the type of selection that the species undergoes. Judging the importance of fountain-of-youth hypothesis (or more appropriately, ‘fountain-of-youth effects’), calls for studying sex reversals and recombination rates, but also the selective pressures acting in the wild.

Supporting Information

Material S1 Further details of the model. In this section, further details of the model are given. Firstly, the coefficient matrices of ‘Dynamics of allele frequencies’ are discussed. Secondly, results of the sensitivity analysis are discussed. (DOC)

Material S2 The \( R \) programs. In this zip file, the \( R \) programs used for this study are given. Further information is available in a readme file within the zip file. (ZIP)

Material S3 The coefficient matrices. In this Excel file, the coefficient matrices from ‘Dynamics of allele frequencies’ are given in their full extent, in a symbolic form. (XLS)
Figure S1 The effect of uneven sex reversal. The black lines represent the proportion of phenotypic females, the blue lines the proportion of Y chromosomes out of all sex chromosomes, the dashed blue lines prevalence of the deleterious mutation in Y chromosomes, and the dashed red lines prevalence of the deleterious mutation in X chromosomes. (Note the log_10 scale of the y axis.) In panel A, it is assumed that \( p_{XY} = 1, p_{YY} = 0.1, p_{YX} = 0.05 \), i.e. the female phenotype is strongly favored so that even some of the YY individuals are female. In panel B, the male phenotype is favored so that \( p_{XY} = 0.7, p_{YX} = 0.1, p_{YY} = 0 \). (TIF)

Figure S2 Less favorable sex reversal under fecundity selection. This figure illustrates the sensitivity of the results towards the rates of directional selection (\( s_z \)) and recombination (\( r \)). In panel A, \( s_z = 0.02 \). In panel B, \( r = 0.10 \). The other parameters are as in 'Baseline', Table 1. Fecundity selection has been assumed. The black data are for population with no sex reversal, the green data for populations with environmental sex determination, and the blue data for \( p = 0.10 \). The dots and the continuous lines represent the sample mean, and the error bars represent 95% confidence intervals. Both of these figures should look like Figure 5B, if the results were not sensitive to \( (s_z, r) \). (TIF)

Figure S3 The effect of recombination pattern. In Figure 1, it is implicitly assumed that the chromosomal organization is \((+/-)X/Y,K/k)\), i.e. that loci \(+/-\) and \(K/k\) are conditionally independent. In this figure, that assumption is relaxed by modifying the recombination matrix \( \Gamma \). In panel A, the chromosomal organization is \((X/Y,+/-,K/k)\), and in panel B, it is \((X/Y,K,k,+/-)\). In both panels, the black lines represent the proportion of phenotypic females, the blue lines the proportion of Y chromosomes out of all sex chromosomes, the dashed blue lines prevalence of the deleterious mutation in Y chromosomes, and the dashed red lines prevalence of the deleterious mutation in X chromosomes. (TIF)

Figure S4 The recombination pattern matters for phenotypes. In this figure, the effect of chromosomal organization is illustrated by investigating the dynamics of phenotypic frequencies. In panel A, the chromosomal organization is \((X/Y,+/-,K/k)\), and in panel B, it is \((X/Y,K/k,+/-)\). The blue lines are the frequencies of the 64 male phenotypes, and the red lines are the frequencies of the 64 female phenotypes. The parameter values are as in the 'Baseline' scenario, Table 1. (TIF)

Figure S5 The recombination pattern and population dynamics. This figure illustrates the sensitivity of the population-dynamic consequences of sex reversal towards the chromosomal organization. In panel A, the chromosomal organization is \((X/Y,+/-,K/k)\), and in panel B, it is \((X/Y,K,k,+/-)\). Fecundity selection has been assumed, and the parameter values are as in 'Baseline', Table 1. If the chromosomal organization did not matter, both of these figures should look like Figure 5B. (TIF)

Acknowledgments

I thank Prof. Otso Ovaskainen and Prof. Juha Merilä for commenting on this manuscript.

Author Contributions

Conceived and designed the experiments: MK. Performed the experiments: MK. Analyzed the data: MK. Contributed reagents/materials/analysis tools: MK. Wrote the paper: MK.

References

1. Keeney S, Kauppi L, Barchi M, Basadat F, Romanienko PJ, et al. (2011) Distinct Properties of the XY Pseudoautosomal Region Crucial for Male Meiosis. Science 331: 916–920.
2. Rice WR (1996) Evolution of the Y sex chromosome in animals. Bioessays 27: 1076–1083.
3. Steinemann S, Steinemann M (2005) Y chromosomes: born to be destroyed. Science 313: 916–920.
4. Steinemann M, Steinemann S, Lottspeich F (1993) How Y chromosomes become genetically inert. Proc Natl Acad Sci U S A 90: 5737–5741.
5. Perrin N (2009) Sex Reversal: A Fountain of Youth for Sex Chromosomes? Genome Biology and Evolution 2: 347–357.
6. Rice WR (1987) Genetic hitchhiking and the evolution of reduced genetic activity of the Y sex chromosome. Genetics 116: 161–167.
7. Rice WR (1996) The Effect of Background Selection against Deleterious Mutation in Weakly Selected, Linked Variants. Genetic Research 63: 213–227.
8. Hill WG, Robertson A (1966) Effect of Linkage on Limits to Artificial Selection. Genetic Research 8: 269–294.
9. Ospina-Alvarez N, Piferrer F (2008) Temperature-Dependent Sex Determination in Fish Revisited: Prevalence, a Single Sex Ratio Response Pattern, and Possible Effects of Climate Change. Plos One 3: -.
10. Ah-King M, Nylin S (2010) Sex in an Evolutionary Perspective: Just Another Evolutionary Solution? Trends in Ecology & Evolution 25: 331–337.
11. Schartl M (2004) Sex chromosome evolution in non-mammalian vertebrates. Genetical Research 8: 269–294.
12. Bachtrog D (2006) A dynamic view of sex chromosome evolution. Current Opinion in Genetics & Development 16: 634–641.
13. Engelstädter J (2008) Muller’s ratchet and the degeneration of Y chromosomes: a simulation study. Genetics 180: 957–967.
14. Marais GAB, Campos PRA, Gordo I (2010) Can Intra-Y Gene Conversion Oppose the Degeneration of the Human Y Chromosome? A Simulation Study. Genome Biology and Evolution 2: 537–557.
15. Ospina-Alvarez N, Piferrer F (2008) Temperature-Dependent Sex Determination in Fish Revisited: Prevalence, a Single Sex Ratio Response Pattern, and Possible Effects of Climate Change. Plos One 3: -.
16. Alho JS, Matsuura C, Merila J (2010) Sex reversal and primary sex ratios in the common frog (Rana temporaria). Molecular Ecology 19: 1763–1773.
17. Stellken RB, Wedekind C (2010) Environmental sex reversal, Trojan sex genes, and sex ratio adjustment: conditions and population consequences. Molecular Ecology 19: 627–646.
18. Oegema-Alvarez N, Piferrer F (2008) Temperature-Dependent Sex Determination in Fish Revisited: Prevalence, a Single Sex Ratio Response Pattern, and Possible Effects of Climate Change. Plos One 3: -.
19. Ah-King M, Nylin S (2010) Sex in an Evolutionary Perspective: Just Another Reaction Norm. Evolutionary Biology 37: 234–246.
20. Janzen FJ, Phillips PC (2006) Exploring the evolution of environmental sex determination, especially in reptiles. Journal of Evolutionary Biology 19: 1775–1784.
21. Bull JJ (1983) Evolution of sex determining mechanisms. Merlo Park, USA: The Benjamin/Cummings Publishing Company.
22. Burt A, Bell G, Harvey PH (1991) Sex-Differences in Recombination. Journal of Evolutionary Biology 4: 239–277.
23. Alho JS, Matsuura C, Merila J (2010) Recombination Rate between Sex Chromosomes Depends on Phenotypic Sex in the Common Frog. Evolution 64: 3634–3637.
24. Matsuura C, Alho JS, Merila J (2010) Recombination Rate between Sex Chromosomes Depends on Phenotypic Sex in the Common Frog. Evolution 64: 3634–3637.
25. Kondo M, Nagao E, Mitani H, Shima A (2001) Differences in recombination lengths largely determine the different recombination rates in male and female gametes. Chromosome Research 9: 555–562.
26. Merila J, Matsuura C (2009) Isolation and characterization of 145 polymorphic microsatellite loci for the common frog (Rana temporaria). Molecular Ecology Resources 9: 533–562.
27. Teixeira C, Huben MA (2004) Inter-sex variation in synaptonemal complex lengths largely determine the different recombination rates in male and female germ cells. Cytogenetic and Genome Research 107: 208–215.
28. Groese C, Neuenwander S, Perrin N (2011) Temperature-Dependent Turnovers in Sex-Determination Mechanisms. A Quantitative Model. Evolution 65: 64–78.
32. Matsuba C, Miura I, Merila J (2008) Disentangling genetic vs. environmental causes of sex determination in the common frog, Rana temporaria. BMC Genetics 9: -.

33. Shine R, Elpick MJ, Donnellan S (2002) Co-occurence of multiple, supposedly incompatible models of sex determination in a lizard population. Ecological Letters 5: 486–489.

34. Aida T (1936) On the inheritance of color in a fresh-water fish Aplocheilus latipes Temnack and Schlegel, with special reference to the sex-linked inheritance. Genetics 6: 554–573.

35. Sarre ND, Georges A, Quinn A (2004) The ends of a continuum: genetic and temperature-dependent sex determination in reptiles. Bioessays 26: 639–645.

36. West SA, Griffin AS, Gardner A (2007) Social semantics: altruism, cooperation, mutualism, strong reciprocity and group selection. Journal of Evolutionary Biology 20: 415–432.