The evolution of chromosomal sex determination and dosage compensation

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In many species, sex is determined by a system based on X and Y chromosomes, the latter having lost much of their genetic activity. Y chromosomes have evolved independently many times, and the associated change in gene dosage in the heterogametic (XY) sex is often compensated for by regulatory mechanisms which ensure equal amounts of gene products of X-linked loci in males and females. There have recently been substantial advances in our knowledge of the molecular biology and genetics of sex chromosomes and dosage compensation, and in our understanding of the population genetic processes which are involved in their evolution.

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Introduction

Systems of chromosomal sex determination have evolved independently in many groups of animals and plants. These systems have several striking characteristics (Box 1), found in a variety of taxonomic groups. This suggests that similar evolutionary forces have operated in different lineages. Sex chromosome evolution often produces morphologically and genetically distinct X and Y chromosomes (with male heterogamety), or Z and W chromosomes (with female heterogamety) [1–3]. For convenience, I shall mostly consider male heterogametic systems from now on; similar considerations apply to female heterogamety, with the appropriate change of notation. It is generally believed that the X and Y chromosome were originally homologous, and have only gradually diverged, with the end stage being the loss of active genes from most or all of the Y (genetic erosion) [3–7].

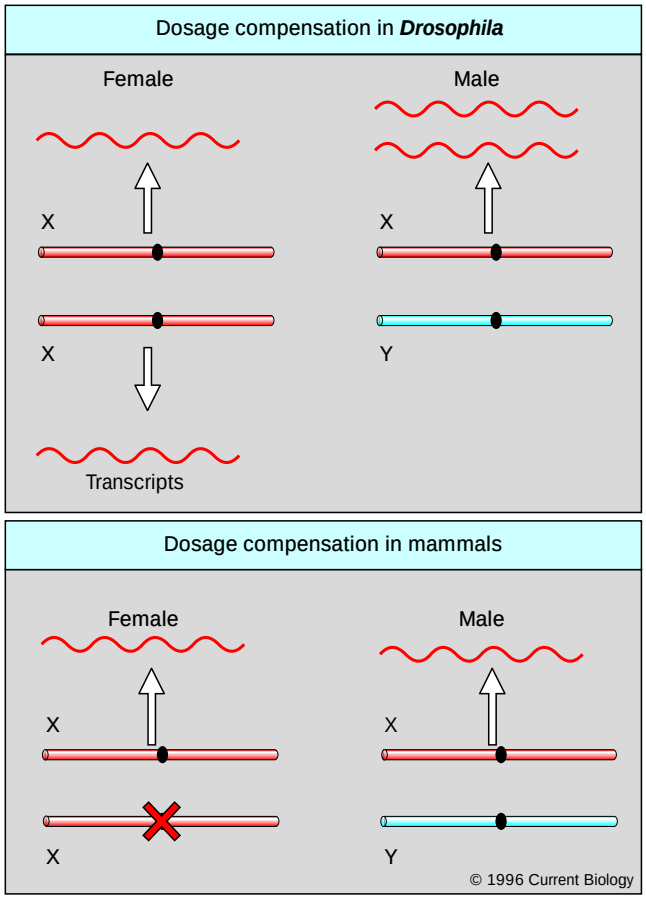
In some (but not all) groups with XX/XY sex determination, the possession of a genetically eroded Y chromosome is associated with dosage compensation, such that the activity of most X-linked genes is effectively the same in males and females [8–10] (Fig. 1). It has been suggested that the evolution of an eroded Y chromosome and of

Box 1

Features of genetic and chromosomal sex determination.

The genetic control of sexual phenotype is frequently associated with a locus, or set of closely-linked loci, that are homozyous in one sex – the homogametic sex – and heterozygous in the other – the heterogametic sex [1–3]. Male heterogamety has apparently evolved more often than female heterogamety [3]. Elaborations of this simple mode of sex determination have repeatedly evolved [3]. In the most primitive systems, there are no gross structural differences between the chromosomes carrying the sex-determining alleles. In more advanced systems, chromosomal rearrangements are present which prevent recombination around the sex-determining region in the heterogametic sex [1–3,5]. In the most advanced systems, the sex chromosomes are morphologically and genetically very different, and there is no recombinational exchange between them for all or part of their lengths. In such advanced systems, the chromosome that is restricted to the heterogametic sex (Y or W) lacks most of the genes carried on its partner (X or Z) [1–6]. In some cases, the heterogametic sex completely lacks a homologue of the X or Z chromosome (giving XX/XO sex determination) [2,3]. The Y or W chromosome may carry some genes which are absent from its partner, but which provide products that are useful for the heterogametic sex [6,24,96]. In several lineages with XX/XY or XX/X0 sex determination, there is a difference in gene activity between the X chromosomes in males and females, such that the overall level of X-linked gene products in males and females is approximately the same (dosage compensation; see Figure 1) [8–11].

Figure 1



Two different mechanisms of dosage compensation are shown. In *Drosophila* (top), there is a higher rate of transcription from the X chromosome in males than there is in females. This results in approximately equal amounts of gene products from most X-linked genes in males and females. In mammals (lower section), only one of the two X chromosomes is active in females. In marsupials, it is always the paternal X chromosome that is inactivated. In eutherians, either the paternal or the maternal X is inactivated in different cells of the same individual.

dosage compensation are both reflections of evolutionary forces which lead to selection for increased expression of genes on the X chromosome in the heterogametic sex, relative to their homologues on the Y, in response to mutation-driven decline in the genetic quality of Y-linked genes [4,11–14].

Here, I shall review possible mechanisms for the evolution of genetically eroded Y chromosomes and dosage compensation, in the light of recent advances in the theoretical understanding of relevant evolutionary forces and in our knowledge of the molecular and genetic basis of sex determination. I shall also consider the poorly understood evolution of X/autosomal balance sex determination (found in *Drosophila* [15,16], the flowering plant *Rumex acetosa* [17] and *Caenorhabditis elegans* [7,18]), and of XX/X0 sex determination. The well-known tendency for repetitive DNA sequences to accumulate on Y chromosomes will not be discussed, as this topic has recently been reviewed [19].

The evolution of incipient X and Y chromosomes

The evolutionary origin of primitive sex chromosomes is closely associated with the evolution of genetically determined separate sexes [1,3,6], and it is necessary to discuss this briefly before turning to the main topic of this review. The best understood path is when separation of the two sexual functions into different individuals (dioecy) evolves from an ancestral cosexual state, in which male or female functions are expressed in the same individual. Cosexuality is characteristic of most flowering plants, many invertebrate taxa and some fish species [3,6]. The simplest method of transition from cosexuality to a fully dioecious state is by mutations at two loci, one, *f*, controlling female function, and the other, *m*, controlling male function [1,20]. The selective forces causing an evolutionary transition between cosexuality and dioecy *via* an intermediate stage of gynodioecy (polymorphism for male-sterile and cosexual individuals) are outlined in Box 2, and have been discussed in more detail elsewhere [6,20].

Although this model is undoubtedly very oversimplified, its general outline is consistent with evidence from the genetics of sexuality in flowering plants, where transitions from cosexuality to dioecy are often of relatively recent evolutionary origin [1,6,20]. It also provides a simple explanation for the evolution of a male-determining proto-Y chromosome, and the predominance of male heterogamety [3,6,20]. A somewhat similar scenario can be envisaged for the evolution of genetic sex determination from environmental sex determination. Determination of sex by the temperature during early development occurs in many groups of lower vertebrates [3,21–23]. Alleles at two different loci that convert individuals of labile sex phenotype into males and females, respectively, can invade a population with environmental sex determination under suitable conditions on the relative fitnesses of males and females in different environments [3,23]. It is uncertain whether environmental sex determination in lower vertebrates is ancestral or derived [3,21].

After the establishment of the proto-X/proto-Y chromosome system, there is selection for closer linkage of the two sex-determining loci (Box 2). In addition, selection for alleles which are advantageous in males but disadvantageous in females will lead to further genetic differentiation between the two sex chromosomes, and to selection for suppression of recombinational exchange over most or all of their length [3,6,24]. This sets the scene for further evolution of the incipient Y chromosome and of dosage compensation, which are discussed in the next section.

Box 2

Evolution of separate sexes and primitive sex-determining mechanisms.

A male-sterility or female-sterility mutation which arises in an initially cosexual population may be favoured by selection for two reasons: first, avoidance of the production of unfit progeny by self-fertilization; and second, increased fertility through reallocation of resources to the sexual function that is retained by mutant individuals [6,20]. The first factor favours mutants that have nearly complete effects on fertility, as these must be fully outcrossing. In addition, male-sterility mutations are more likely to be favoured than female-sterility mutations, as reproduction through male gametes is of lower genetic value than reproduction through female gametes in a population which experiences some degree of self-fertilization [6,20]. Because loss-of-function mutations are usually recessive, the first step in the evolution of dioecy by this path is likely to be the establishment of a polymorphism for females and cosexuals (gynodioecy), with femaleness caused by a recessive mutation, m^s [6,20]. This model is supported by genetic and comparative evidence [1,6,20]. The presence of females means that reproduction through male gametes in a gynodioecious population is of greater value than in the original cosexual population. This enhances the prospects for selection of a female-sterility mutation, f^S, at another locus [6,20]. If linkage between the two loci is close, and f^S is completely dominant, most of the final population will consist of homogametic females ($f^f m^s / f^f m^s$) and heterogametic males ($f^S m^F / f^f m^s$). If f^S is incompletely dominant, full dioecy will not be established at once, although there will be selection for modification towards full dominance [20]. With the opposite dominance relations for m^s and f^S, female heterogamety will evolve. Clearly, if m^s and f^S are both recessive or both dominant, dioecy cannot evolve, and there is nothing to explain. If the two loci are loosely linked, recombination between them can generate genotypes, such as $f^S m^s / f^f m^s$, that are completely sterile. This means that close linkage between the *m* and *f* loci is required for the spread of the second mutation [6,20]. Genetic factors or chromosomal rearrangements that reduce recombination will be favoured, leading to the two loci being inherited as a unit [20]. Under this scenario, chromosomes carrying the combination $f^S m^F$ are proto-Y chromosomes, and chromosomes with $f^f m^s$ are proto-X chromosomes.

Erosion of the Y chromosome and the evolution of dosage compensation

There are two major models in the literature for the evolutionary erosion of an incipient Y chromosome. One invokes 'Muller's ratchet' [13], and the other the fixation of deleterious Y-linked mutations by 'hitchhiking' with selectively favourable mutations on the Y chromosome [14] (see below). Other models have been proposed [4,12,25], but they suffer from various difficulties which make them seem less plausible [13,26].

It has also been suggested that restricted recombination between the X and Y chromosomes may have led to a higher mutation rate for Y-linked alleles, and thereby to the mutational decay of the Y chromosome [5]. But it is unlikely that even a greatly elevated rate of mutation on an incipient Y chromosome would cause the fixation of deleterious alleles on the Y chromosome in opposition to selection in a large population [13,26], although it could accelerate the processes described below. Such an acceleration could also be caused by the higher rate of mutation in males than females in mammals [27,28]. But there is no evidence for a difference in mutation rate between males and females in *Drosophila* [29], despite the lack of crossing over in males, so that elevated mutation rates cannot contribute to Y chromosome degeneration in this genus.

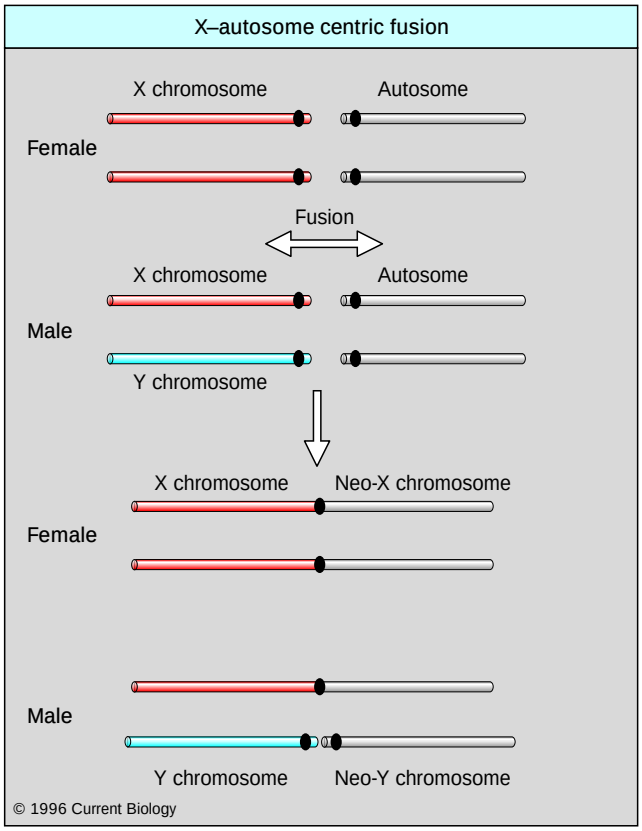
The same evolutionary principles apply to a proto-Y chromosome that has evolved restricted recombination, but still retains most of its genetic homology to the proto-X chromosome, and to a neo-Y chromosome that has evolved by fixation of a reciprocal translocation or centric fusion between a sex chromosome and an autosome (Fig. 2) [2]. All that is required is that the incipient Y chromosome be maintained permanently heterozygous over the X in the heterogametic sex, with no recombination over all or part of its length. Because of their relatively recent evolutionary origin, neo-X and neo-Y chromosomes provide very useful model systems for studying the degeneration of Y chromosomes and the evolution of dosage compensation [11].

I shall assess the plausibility of the two major models of Y chromosome degeneration in the light of empirical data on the mutation rates and fitness effects of deleterious alleles in *Drosophila*, and recent theoretical work on the evolutionary consequences of deleterious mutations. In addition, I shall discuss an alternative model, involving the 'background selection' effect of deleterious mutations on evolution at linked loci [30–36].

Deleterious alleles in *Drosophila*

Information on the per-genome rate of spontaneous mutation to deleterious alleles in *Drosophila* comes mainly from experiments by Mukai, Ohnishi and coworkers on egg-to-adult viability (reviewed in [37]). These involved measurements of the rate of decline of the mean viability, and the rate of increase in the between-line variance in viability, for *D. melanogaster* lines carrying second chromosomes that had accumulated mutations independently for many generations, in the effective absence of selection. A maximum likelihood reanalysis of these data indicates that the mean number of new mutations to non-lethal deleterious alleles (detrimentals) affecting viability is probably at least 0.2 per haploid second chromosome per generation [38]. Lethals contribute an additional component of the order of 0.005 events per generation [37]. Indirect estimates of the genomic mutation rate to deleterious alleles from the level of heterosis in highly selfing plant species are broadly consistent with this estimate of the mutation rate for detrimentals in *Drosophila* [39].

Figure 2



The formation of a neo-X/neo-Y sex chromosome system by a Robertsonian centric fusion between an autosome and the original X chromosome. The original state is shown in the upper section; the double-headed arrow indicates the chromosomes that undergo the centric fusion. Fixation of the fusion chromosome within the population results in males and females with the constitutions shown in the lower part of the figure. The neo-X chromosome is formed by the autosome that is fused to the X. The neo-Y chromosome is formed by its free homologue, which is present only in males.

Given that the X chromosome is approximately half the size of the second chromosome, these data suggest a mutation rate to deleterious alleles of ~0.1 per haploid X chromosome in *Drosophila*. But a large component of the homozygous genetic load in *Drosophila* is due to loci affecting male fertility [40], which may contribute little to the mutation rate estimated from experiments on viability. It is therefore probable that these experiments underestimate the total mutation rate for alleles affecting fitness [41]. The observed rate of decline in fitness of a non-recombining pair of chromosomes in a small laboratory population of *D. melanogaster* also indicates a high mutation rate to deleterious alleles [42]. Chromosome arms in the genus *Drosophila* are of similar size, and seem to be well conserved in evolution [43–45]. The mutation rate both for the primeval Y and for a neo-Y formed by an X–autosome or Y–autosome fusion (Fig. 2) should therefore be similar to that for the present-day X chromosome, assuming that the primeval sex chromosome system evolved in an ancestor with a genome structure similar to that of present-day members of the genus. An estimate of 0.1 for the deleterious mutation rate for a proto-X or neo-Y chromosome in *Drosophila*, with respect to loci affecting the fitness of males, is likely to be quite conservative.

In an infinitely large, randomly mating population, a balance between mutation and selection at many loci will be established, so that there is a stable distribution of the number of deleterious mutations per chromosome (Fig. 3), with a mean and variance which can calculated from the mode of selection, the mutation rate and the amount of recombination [46–48]. The equilibrium number of mutations per proto-Y or neo-Y chromosome in such an infinite population plays a major role in the processes discussed below.

Assume that there are m mutable sites on the proto-Y chromosome, the mutation rate at the ith site is u_i, and the corresponding selection coefficient against a heterozygous mutation at this site in males is t_i. (Data on *Drosophila* show that both lethal and detrimental mutations usually have significant deleterious effects on fitness when heterozygous [37,49], so that the assumption that selection takes place predominantly against heterozygous mutations in a randomly mating population is well supported.) The equilibrium mean number of deleterious mutations per incipient Y chromosome, $\bar{n}_Y$, is given by the following expression [46–48]:

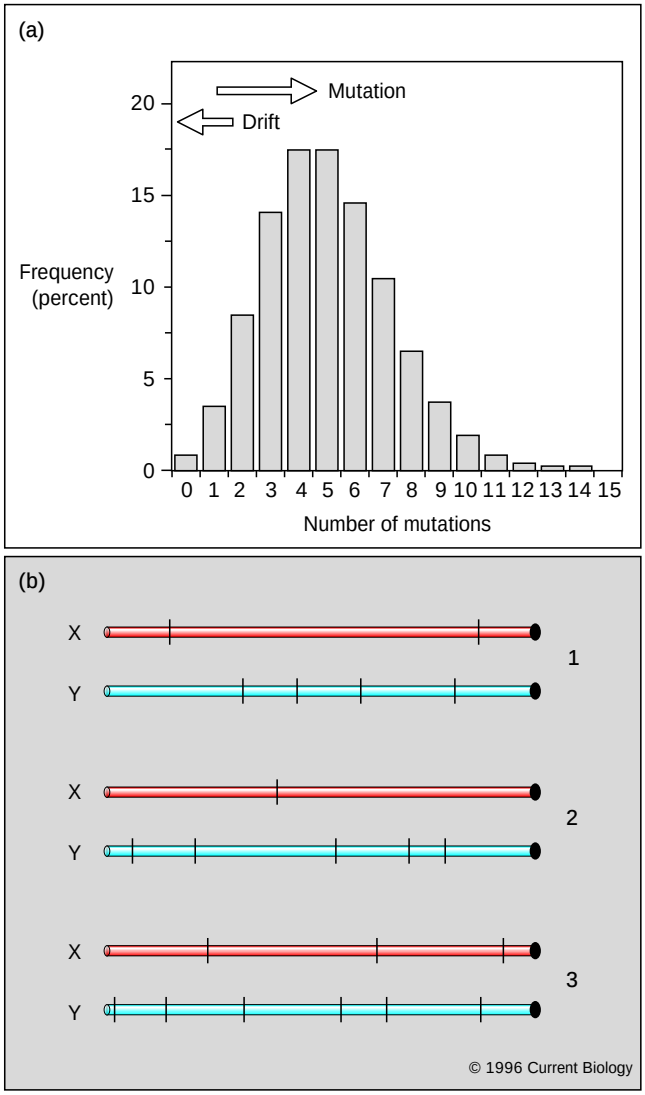
$$\bar{n}_Y = \sum_{i=1}^{m} \frac{u_i}{t_i} \qquad (1)$$

If detrimental mutations with small effects on fitness are more frequent than those with large effects, as seems likely [37,38], then $\bar{n}_Y > u_Y/t_H$, where u_Y is the mutation rate to deleterious alleles for the proto-Y chromosome, and t_H is the harmonic mean of the t_i. The value of u_Y/t_H thus provides an underestimate of the equilibrium mean number of mutations per proto-Y chromosome. An estimate of 0.02 for t_H has been obtained for the heterozygous effects of mutations on net fitness of *D. melanogaster* under natural conditions, from the ratio of the reduction in mean viability associated with homozygosity for non-lethal autosomes to the mutational decline in mean viability [37]. This is consistent with the apparently rather large mean homozygous effects of spontaneous deleterious mutations on net fitness compared with their effects on a single fitness component [41], and the partial recessivity of most detrimental mutations [37,39,49]. Given that u_Y is probably > 0.1, $\bar{n}_Y$ in *Drosophila* is therefore likely to exceed five.

Muller's ratchet

The term Muller's ratchet refers to the stochastic loss of the class of chromosomes carrying the smallest number of deleterious mutations in a population of finite size. In the

Figure 3



(a) The histogram shows the expected distribution of the number of deleterious mutations per proto-Y chromosome, in a population at equilibrium between mutation and selection with a mean number of mutations of 5 per chromosome. As indicated by the leftward arrow, genetic drift in a finite population causes the eventual loss of the chromosome class with the lowest number of mutations. Mutation pressure, indicated by the rightward arrow, causes a flow from lower to higher numbers of mutations. In the absence of recombination, these forces together result in the movement of the distribution to the right – Muller's ratchet. (b) The outcome of Muller's ratchet operating on an incipient Y chromosome. Incipient X and Y chromosome pairs in three independent individuals drawn from the population are shown. The vertical lines indicate the positions of deleterious mutations. The mean number of mutations on the proto-X chromosome, which is freely recombining in females, is given by the equilibrium between mutation and selection in a large population. Because of the operation of the ratchet on the non-recombining portion of the proto-Y, the mean number of deleterious mutations on the Y chromosome is greater than on the X. Note, however, that these mutations are still infrequent within the population at any given locus, so that they are present at different sites in different individuals.

absence of recombination and back mutation, this class of chromosome cannot be replaced after it has been lost. This results in a progressive increase in the mean number of deleterious alleles per individual [50,51] (Fig. 3). In large populations, this can occur in the absence of accelerated fixation of deleterious alleles at individual loci [51,52], an important difference from other mechanisms of Y chromosome degeneration (see below). There has recently been progress in the theoretical analysis of the progress of Muller's ratchet for strictly asexual populations, although a general analytic solution has not yet been obtained [53–55].

The behaviour of the non-recombining portion of an incipient Y chromosome is similar to that of the commonly analyzed case of a haploid asexual population. As only males bearing Y chromosomes are relevant, the number of breeding males, N_m, replaces the population size N used in the haploid models. The critical parameters that determine the rate of the ratchet are N_m and $\bar{n}_Y$. With multiplicative fitness interactions among loci, the equilibrium frequency of chromosomes free of deleterious mutations in an infinite population is $f_0 = \exp(-\bar{n}_Y)$ [46]. If the equilibrium number of individuals in the zero class, $f_0 N_m$, is of the order of 1 or less, the ratchet will advance rapidly, and the mean number of deleterious mutations per chromosome will increase by a large amount over a short period of evolutionary time [51–55]. If $f_0 N_m > 100$, the ratchet will proceed very slowly, with a rate of increase of $\bar{n}_Y$ of one every several thousand or millions of generations. The dependence of the speed of the ratchet on $f_0 N_m$ becomes exponential for such large values [53]. The corresponding rate of decline of the mean fitness of males is approximately equal to the rate of change of $\bar{n}_Y$ multiplied by the mean selection coefficient [52,54,55].

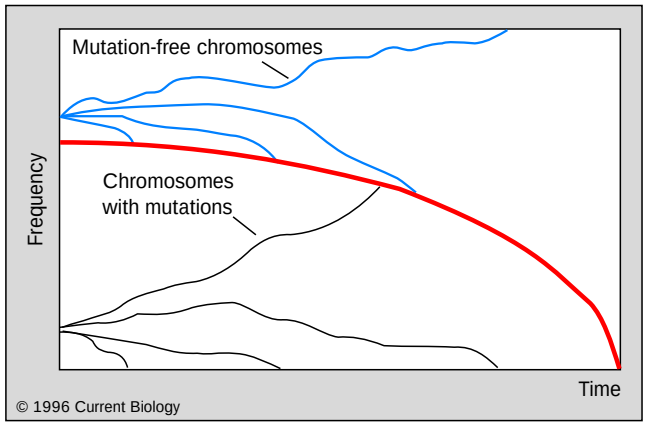
With the value of $\bar{n}_Y$ suggested by the *Drosophila* data, $f_0 = 0.0067$, so that $f_0 N_m = 670$ for a male population of one hundred thousand, and 6700 for a population of one million. Using the parameters assumed here, and equations (14) in [53], the average time between turns of the ratchet would be about 50 000 generations in the first case, and 3×10^{30} generations in the second. Data on DNA sequence variability at silent nucleotide sites suggest that total effective sizes of *Drosophila* species are in the millions [56,57]. Given that population subdivision is relatively weak in many species of *Drosophila* [58], the species population size, rather than local population size, is likely to be important in affecting the speed of the ratchet.

These considerations imply that the rate of advance of the ratchet for an incipient *Drosophila* Y chromosome is likely to be exceedingly slow, in contrast to what was believed when the ratchet was first proposed as an explanation of Y chromosome degeneration [13]. Synergistic fitness interactions — such that the selection coefficient against a new

mutation increases with the number of deleterious mutations already present in the genome — reduce the speed of the ratchet below this estimate [52,59,60]. The ratchet will move much faster if there is a large class of mutations with small effects on fitness, but the rate of decline in fitness will be greatly reduced if selection coefficients are much smaller than 0.02 [52,54,55,60].

The selective force that drives the evolution of inactivation of the Y chromosome and dosage compensation is related to the decline in mean fitness of carriers of the Y chromosome. The increased abundance of Y-linked deleterious mutations means that it is selectively advantageous to increase the activity of X-linked genes in males, even at the expense of the activity of genes on the Y chromosome (Fig. 3), provided that the total level of activity of genes on the two sex chromosomes combined is not greatly affected [13]. The end result of this process is a complete shutdown of expression of genes carried on the Y (except those which are required for function of the heterogametic sex and lack homologues on the X), and a doubling of the level of expression of genes on the X compared with the original state. But the above arguments imply that it is unlikely that this selective force will become very noticeable until many millions of years have elapsed after the formation of an incipient Y chromosome, unless the population size is much smaller than is indicated by the levels of DNA variation at silent nucleotide sites in *Drosophila* [56,57]. The ratchet is much more plausible for mammals, with their relatively small effective population sizes [61].

Figure 4



The fate of weakly selected variants when introduced into a large, non-recombining population in which strongly selected deleterious alleles are maintained at many loci by mutation pressure. In the initial generation, a new variant occurs either in a chromosome which is free of deleterious alleles (above the red line), or in a chromosome which contains one or more deleterious alleles (below the red line). As strongly selected deleterious alleles only persist in the population for a short time after they are created by mutation, the genetic make-up of future generations is increasingly dominated by the descendants of chromosomes that were mutation-free in the initial generation (blue lines). Hence, a new variant that arises in a chromosome which carries deleterious mutations will be ultimately lost from the population, even if it increases in frequency within the class of mutant chromosomes as a result of a selective advantage, unless its selective advantage is strong enough to outweigh the selective disadvantage of the associated deleterious mutations.

Hitchhiking by favourable mutations

An advantageous mutation that occurs on the non-recombining portion of an incipient Y chromosome will cause the fixation of all deleterious mutations present on the chromosome on which it occurs [14] — a process known as hitchhiking [62]. Successive 'selective sweeps' of this kind would cause the fixation of deleterious alleles at many Y-linked loci, leading to selection for increased activity of the non-mutant X-linked loci relative to the Y-linked loci that carry mutant alleles [14].

This model suffers, however, from the following difficulty. In a large, non-recombining population which is at equilibrium under mutation–selection balance at many loci, a new allele has a non-zero chance of survival only if it arises in a mutant-free chromosome, unless it is more strongly selected than most of the deleterious mutations [30–36]. This is because chromosomes carrying one or more deleterious mutations are rapidly eliminated from the population, carrying the new variant with them, unless the latter has a sufficiently large selective advantage to overcome the fitness disadvantage due to the deleterious alleles with which it is associated (a process known as 'background selection' [33]). Unless the population size is so small that Muller's ratchet operates very rapidly, or the selective advantage of the new variant is rather large compared with the mean selection coefficient for deleterious mutations, t_H, only chromosomes that are free of deleterious mutations contribute to the ancestry of future generations (Fig. 4).

In the absence of background selection, the probability of ultimate fixation of a favourable Y-linked new mutation with a small selective advantage, s, introduced into a population of N_m breeding males, is $\sim 2s$ if $sN_m \gg 1$ [63]. (For simplicity, it will be assumed here that the effective population number, whose reciprocal measures the effectiveness of genetic drift [63], is the same as the number of breeding individuals.) In the presence of background selection, and with no recombination, this probability is reduced by a factor of f_0 [31,34–36] if the favourable mutation can only be established on a background free of deleterious alleles. The probability of fixation of a very weakly selected Y-linked allele ($sN_m < 1$) approaches that for a neutral mutation [34]. In either case, the favourable variants that become established have arisen in mutant-free backgrounds, and hence do not cause hitchhiking of deleterious alleles. These results imply that only rather strongly selected favourable mutations (with s of the order of 0.02 or more), which can proceed to fixation even if they are associated with one or more deleterious alleles

[31,35,36], can contribute to the degeneration of the proto-Y chromosome. This process is, therefore, almost certainly less important than was originally envisaged [14].

Accelerated fixation of slightly deleterious mutations because of background selection

The reduction in effectiveness of selection on relatively weakly selected alleles because of background selection can also cause slightly deleterious mutations to experience an accelerated rate of substitution by drift [32,34]. With no recombination and with $f_0 N_m = 5000$, for example, mutations with a heterozygous selection coefficient of the order of 0.0001 will be effectively neutral, and the rate of substitution of new variants over evolutionary time will approach the rate at which they originate by mutation [63]. It is important to note that the selection coefficients which are compatible with the fixation of slightly deleterious alleles in large populations are at least two orders of magnitude smaller than the mean selection coefficients against detrimental mutations detected in the *Drosophila* experiments mentioned earlier. Any such alleles must therefore form a class which is quite distinct from the more drastic mutations responsible for background selection; in most cases of interest, these latter mutations can safely be assumed to be held close to their equilibrium frequencies. Recent studies of codon usage have provided evidence for very weak selection against mutations from preferred to less preferred codons [64]; the importance of the fixation of slightly deleterious alleles in protein sequence evolution is more controversial [65,66].

In *Drosophila*, a typical rate of substitution of silent nucleotide changes is of the order of 1 % per nucleotide site per million years [67]. As a result of background selection, weakly selected changes, which would otherwise be removed by selection with high probability, can become fixed at close to this rate on an incipient Y chromosome. This implies that the mean fitness of carriers of the Y chromosome will decline over evolutionary time [12], creating a selection pressure for increased activity of genes on the X relative to their homologues on the Y, as in the other two models discussed above [13,14].

An estimate of the size of this effect can be obtained as follows. Consider a Y-linked locus with a selection coefficient t against slightly deleterious sequence variants, in a population with N_m breeding males. Writing $\alpha = f_o N_m t$ and v for the rate of mutation to slightly deleterious variants per nucleotide site, equation (11) of [34] yields the following expression for the substitution rate of such variants per site per generation:

$$K = \frac{2\alpha v}{(e^{2\alpha} - 1)} \qquad (2)$$

If mutant effects at different loci interact multiplicatively, and the total mutation rate to slightly deleterious variants on an incipient Y chromosome is v_Y, this result implies that the rate of change of mean fitness, in time units of $f_o N_m v_Y^{-1}$ generations, is given by:

$$\frac{d\ln \bar{w}}{dt} = \frac{2\alpha^2}{(1 - e^{2\alpha})} \qquad (3)$$

A maximal value of 0.33 for the rate of decline of log mean fitness in these time units is attained when $\alpha = 0.8$. If the modest assumption is made that the ~22 megabase euchromatic part of the *Drosophila* X chromosome contains two hundred thousand sites subject to selection of this order of magnitude, each mutating at a rate of 10^{-2} per million years, and which interact multiplicatively to determine fitness, the mean log fitness of an incipient Y chromosome would decline at a maximal rate of $0.33 \times 10^3 / f_o N_m$ per million years. With $f_o N_m = 5000$, this is equal to 0.13 per million years — that is, the mean fitness of a Y chromosome would decline to 88 % of its initial value over a million years. Again, this creates a selection pressure for increasing the activity of X-linked loci at the expense of the homologous Y-linked loci, leading eventually to the evolution of inactive Y-linked loci [13,14].

This calculation assumes independence between substitutions at separate sites on the incipient Y chromosome. Mutual interference between the effects of selection on alleles at closely linked loci can accelerate the rate of fixation of deleterious alleles [53,68]. The rate of decline of mean Y chromosome fitness may therefore be substantially greater than is predicted by equation (3).

Evidence on the evolution of eroded Y chromosomes and dosage compensation

Direct experimental tests of these models are obviously hard to conduct, given the long time-scale over which they operate. But some useful, if only provisional, conclusions can be drawn using indirect inferences based on comparative data, and on the mechanisms of dosage compensation. Other tests, based on features of within-population variation and between-species divergence for loci on a neo-Y chromosome, are also feasible.

Comparative data from *Drosophila* provide evidence on the rate of degeneration of neo-Y chromosomes. The neo-Y of *D. americana* is the product of a centric fusion between chromosome arm B of the basic *Drosophila* karyotype [43] and the X chromosome, which are unfused in the closest relatives of *D. americana*, such as *D. texana* and *D. virilis* [69]. *D. americana* and *D. texana* are separated by a small genetic distance, as measured by divergence of electrophoretic alleles and DNA sequences [69–72], so that the neo-Y may only be a few hundred thousand years old. Hybridization with *D. virilis* strains homozygous for the recessive mutations *px* or *cd* located on arm B indicates that the neo-Y of *D. americana* carries active alleles at these loci [73]. Similar experiments with allozyme markers

indicate that the loci coding for enolase, phosphoglycerate kinase and alcohol dehydrogenase are located on arm B, and that the neo-Y chromosome carries active alleles at all of these loci (unpublished data).

D. miranda has a neo-Y chromosome, absent from its relative *D. pseudoobscura*, formed by the fusion of arm C with the Y chromosome [74]. Sequence data suggest a divergence of about 2 million years between these species, which provides an upper bound to the age of the neo-Y [67,75]. Genetic and molecular data indicate that a substantial fraction of the genes on the neo-Y of *D. miranda* have become non-functional or completely lost, and that their homologues on the X have become dosage compensated, but many loci still retain their function [74–78]. Finally, members of the *pseudoobscura* subgroup of the *obscura* species group have an ancient X–autosome fusion, generating a neo-Y homologous to arm D. This has completely lost genetic activity, and genes on the neo-X chromosome appear to be fully dosage compensated [10,11,79]. This fusion is absent from the *obscura* sister clade, which is separated from the other clade by ~13 million years [67].

These facts are compatible with the theoretical models described above, which require a long period of time for Y chromosome degeneration when the effective population size for the whole species is in the millions or hundreds of thousands. They shed no light, however, on which process has been primarily responsible for the degeneration of the neo-Y chromosome. One possible approach for investigating this question would be through DNA sequence comparisons of homologous neo-Y and neo-X linked loci, and their autosomal homologues, in a related species that lacks the fusion. If the background selection or selective sweep models apply, one would expect to see an accelerated rate of amino-acid replacement substitutions, and of silent-site substitutions that change preferred to non-preferred codons [34,64,80]. This is not a requirement of the Muller's ratchet model, so that detection of an accelerated rate of substitution would support the other mechanisms. Of course, these mechanisms are not mutually exclusive. For example, if the ratchet is operating slowly in a moderately large population (which requires small f_0), background selection will also take place, as the times between turns of the ratchet are so long that the future population is mainly descended from the currently least-loaded class [31]. The question is then whether background selection is solely responsible for the degeneration of the Y, or whether the ratchet also contributes. This may be hard to determine.

The role of selective sweeps relative to the other two mechanisms could be tested by examining the frequency distributions within populations of silent-site variants on the neo-Y chromosome. Recent theoretical studies have shown that hitchhiking in chromosomal regions with little or no crossing-over can produce a large excess of rare variants at neutral sites over classical expectation [81,82], whereas little departure from expectation would be likely to result from selection against deleterious mutations [83,84]. But a neo-Y chromosome that has recently evolved by selection for a fusion between an autosome and the primary Y chromosome will necessarily have experienced a selective sweep, so that it is useless for this purpose. A neo-Y chromosome of intermediate age, as in *D. miranda*, would be likely to have recovered from its initial selective sweep, and would thus provide suitable material for this test. This problem does not arise in the case of a neo-Y formed by an X–autosome fusion, as in *D. americana*, as here the neo-Y chromosomes in the population are derived from a large number of males.

It is perhaps worth noting that there is good evidence that selective sweeps and/or background selection are operating in *Drosophila*, so that these mechanisms have empirical support in addition to the facts presented above. Several regions of the genome of *D. melanogaster* exhibit greatly reduced rates of meiotic crossing over. These include the telomeric region of the X chromosome, the pericentric regions of the major chromosomes, and chromosome four (which lacks meiotic exchange under normal conditions) [85]. DNA variability in natural populations of *D. melanogaster* is lower in such regions, compared with regions where crossing over occurs at normal frequencies [57,86,87]. In addition, genes in regions of reduced recombination seem to have a lower codon bias [80], suggesting that natural selection is less effective in such regions. Both selective sweeps and background selection are expected to produce these patterns, and the extent of their relative contributions is a subject of current research [33,81–84,86–89].

The mechanism of dosage compensation may also shed light on the processes involved in the degeneration of the Y chromosome. The selective sweep model can in principle lead to the fixation of deleterious mutations that have sizeable effects at individual loci on an evolving Y chromosome, creating a selection pressure for enhancing the activity of the X-linked alleles at all loci that have experienced such sweeps [14]. As sweeps must occur sporadically, and will be randomly distributed over the chromosome, dosage compensation is most likely to evolve on a locus-by-locus basis if the selective sweep model applies [14].

In contrast, Muller's ratchet in a large population involves an increase in mean number of mutations per chromosome, rather than the fixation of deleterious variants at each locus (Fig. 3). So if Muller's ratchet applies, dosage compensation should evolve by the modification of gene expression over sufficiently large blocks of the chromosome for there to be a sizeable selective advantage to enhancing X-activity at the expense of Y-activity [13]. The fact that compensation for experimentally produced changes of gene dosage at autosomal loci is often observed

in *Drosophila* and maize [90] suggests that it would in principle not be difficult for a reduction in transcriptional activity from a block of Y-linked loci to result in a corresponding increase in activity from their homologues on the X chromosome. This could lead to the evolution of a primitive system of dosage compensation.

The enhanced rate of substitution of deleterious variants as a result of background selection is intermediate between the other two models in this respect. While this model involves the fixation of deleterious variants, these will be most likely to have minor fitness effects — of the order of 10^{-4} or less — so that it would take the accumulation of several such variants at a locus before there would be much selection for dosage compensation at the level of an individual locus. It would therefore seem that the initial mode of evolution of dosage compensation by this mechanism would be at the level of blocks of loci, although gene-by-gene evolution could occur if enough time had elapsed for multiple substitutions at individual loci to have occurred.

In *Drosophila*, there is solid evidence that dosage compensation is regulated by *cis*-acting sequences, which may be so close to the locus itself that cloned X-linked genes remain at least partially dosage compensated when inserted into autosomes [79,91]. These sequences appear to respond to proteins produced by the autosomal *msl* and *mle* loci, which bind specifically to the X chromosome in males [91]. Dosage compensation of X-chromosome genes that have been transferred to autosomes is often incomplete or may even fail completely [91]. Cloned autosomal genes often become dosage compensated when inserted into the X chromosome [91]. These observations suggest that dosage compensation in *Drosophila* is controlled by relatively remote *cis*-acting regulatory sequences, as well by sequences close to, or even within [92], the genes themselves.

While not conclusive, this evidence suggests that *Drosophila* dosage compensation may have evolved as a dual process of both gene-by-gene enhancement of X-linked gene expression in males, and regulation of X-linked gene activity over larger chromosomal domains. It remains obscure how the male X-chromosome specific proteins and the X-chromosome specific regulatory sequences could evolve by any of the mechanisms discussed here. But the mechanistic difficulties of the selective sweep hypothesis on the one hand, and the fact that Muller's ratchet fails to predict gene-by-gene dosage compensation on the other (and is also likely to be very slow in species with large effective population sizes), suggest that these processes may not have caused the evolution of dosage compensation in *Drosophila*. The background selection model seems to be the most plausible explanation of this system.

Dosage compensation in *Caenorhabditis* differs from that in *Drosophila* and Sciarid flies [93], in that X chromosomal gene activity seems to be actively turned down in XX individuals [18]. Similarly, in mammals, one X chromosome is almost completely inactivated in females (Fig. 1) [9,94,95]. At first sight, a reduction in X chromosome activity in XX individuals seems paradoxical, if dosage compensation has indeed evolved concomitantly with reduced Y-chromosomal gene activity in males. But this situation can be explained in the following way [13]. If increased X-chromosomal gene activity were initially not confined to males, but affected females as well, there would be selection to reduce gene activity in females, in order to restore the balance between X-chromosomal and autosomal gene products. This would eventually lead to a halving of gene activity for the X in females, either by shutting down one X chromosome, as in mammals, or by reducing transcription from both X chromosomes, as in *C. elegans*.

The small effective population sizes of mammals, and the fact that mammalian dosage compensation operates at a high level of chromosomal organization, mean that the ratchet is still a viable hypothesis for mammalian Y chromosome evolution. The only serious difficulty is the fact that not all mammalian X-linked loci that lack partners on the Y are dosage compensated, and some that do have partners are dosage compensated [95,96]. There is no difficulty in understanding the lack of dosage compensation for loci in the pseudo-autosomal region, which cross-over freely with the Y chromosome and hence are not exposed to the forces discussed above. It is also not surprising that some loci on the X chromosome that lack partners on the Y are not dosage compensated; this may simply represent the incomplete evolution of dosage compensation, for example because of peculiarities of the chromatin configuration in the region where they are located. It is harder to understand cases of dosage compensation of loci that have partners on the Y chromosome, such as *Zfx* in the mouse [95,96]. (This difficulty applies, of course, to all explanations of dosage compensation based on an evolutionary response to the loss of Y-linked genes that originally had homology to X-linked counterparts.) Such cases could perhaps be explained in terms of divergence of function between X-linked and Y-linked gene copies, leading to selection for enhanced activity of the X-linked copy in males, and subsequently to down-regulation in females if the enhanced activity were not male-limited. Alternatively, the dosage compensation of these loci could simply be a by-product of their inclusion in a chromosomal region where other loci are dosage compensated.

The *XIST* locus controls X inactivation in eutherian mammals [95]. It is transcribed from the inactive but not the active X after inactivation, but is expressed early in development from the paternal X before being expressed from both X chromosomes prior to X inactivation. *XIST* is expressed in male meiosis before inactivation of the X in spermatocytes [95]. These facts suggest that the original

role of *XIST* may have been the control of X inactivation in spermatocytes [95], and that X inactivation in females may have evolved from imprinting of the *XIST* locus. Carry-over by imprinting of X inactivation in the male germ cells may have led to the marsupial system of paternal X inactivation [13,97].

It appears likely that the eutherian mammalian system of random maternal X inactivation evolved from the marsupial system [13,97,98]. There is a clear selective advantage to such a change, as inactivation of the paternal X chromosome means that every cell of an individual is hemizygous for all deleterious mutations carried on the maternal X chromosome. With random X inactivation, half the cells will be hemizgygous for such mutations, and half will be hemizygous for mutations carried on the paternal X. There should be a fitness advantage in covering up the expression of hemizygous recessive or partially recessive deleterious mutations [13,97], at least for the many loci whose expression is not cell-autonomous [94]. This would be similar to the evolutionary advantage of diploidy over haploidy [99,100].

Finally, it is worth noting that the models discussed here are compatible with the cases where the Y chromosome is genetically inert, but dosage compensation has not evolved, as in Lepidoptera [101] and birds [102]. If the evolution of increased activity of X chromosomal loci is not limited to the male sex, there may not be a large selective premium to reducing their activity in the homogametic sex in order to restore balance with the products of autosomal loci, particularly if the X chromosome forms only a small portion of the genome. Hence, it is possible for the Y chromosome to become inactive without the concomitant evolution of dosage compensation. It is even possible that parts of the X may evolve dosage compensation while others fail to do, as seems to be the case for the part of the X chromosome in eutherian mammals which is autosomal in marsupials, and hence is of a more recent evolutionary origin than the part that is X chromosomal in both types [95].

Evolution of X/A sex determination and X0 sex chromosome systems

Sex determination by a Y chromosome is common but not universal [3]. An alternative mechanism involves the specification of sex by the balance between the number of X chromosomes and the number of autosomes, as first discovered by Bridges in *D. melanogaster* [15]. Individuals with two X chromosomes and two sets of autosomes develop as females, and individuals with one X chromosome and two sets of autosomes develop as males. Individuals with an intermediate X/autosome (X/A) ratio are intersexes. Recent genetic and molecular research has shown that the control of sex differentiation by this mechanism in *Drosophila* and *C. elegans* is achieved by regulation of the activity of a primary sex determining 'counter' locus by the products of X chromosome 'numerator' and autosomal 'denominator' loci [16,18,103,104]. Clearly, the X/A balance mechanism must operate in the numerous groups where one sex is XX and the other sex is X0 [2,3]. A full understanding of the evolution of sex chromosome systems must include X/A and XX/X0 sex determination, but this is a problem that has hardly been tackled either theoretically or empirically. Some tentative ideas are outlined below.

X/A balance

Sex determination systems based on the X/A balance are known to exist in *Drosophila*, *C. elegans* and *Rumex* [15–18], and possibly also in birds [105]. There is an indication of a role for X chromosome dosage in some aspects of sexual differentiation in marsupials [95], but the detailed mechanisms involved are obscure. It seems likely, as suggested by Westergaard [1], that X/A balance systems have evolved secondarily from male-determining Y chromosome systems, as it is difficult to imagine steps by which they could evolve from cosexual ancestors or from systems of environmental sex determination. A scenario for such an evolutionary transition is sketched here, based on the known mechanisms of X/A balance sex determination in *Drosophila* and *C. elegans*. I do not mean to imply that the proposed scenario actually represents the course of evolution of sex chromosome systems in *Drosophila* or other groups. Comparative data on the evolutionary relationships between sex chromosome systems and modes of sex determination in different groups are currently inadequate to permit a reconstruction of the history of the evolution of these systems.

One assumption is crucial for this scenario to work: the m^F allele postulated above (Box 2) must not be required for the development of phenotypically male cells, although it is required for male fertility. It is thus analogous to the Y-chromosomal loci of *Drosophila* needed for male fertility, rather than the *SRY* gene of mammals, which acts as a primary determinant of maleness [95,96]. Species where a gene such as m^F is required for male development are clearly constrained to retain a male-determining Y mode of sex determination, unless this can be overriden by other genetic mechanisms [106]. Differences between major taxa in mode of sex determination are thus likely to be a product of historical accidents, reflecting the types of mutations that arose in the early stages of the evolution of these mechanisms in different groups, rather than the outcome of different selective pressures.

It is also assumed that expression of f^f is required for female development, and that lack of f^f product — for example, because of the presence of a dominant female-sterility allele f^S — leads to partial or complete development as male. There is an obvious analogy between the postulated function of f^f and the properties of *Sxl* in *Drosophila*. Expression of *Sxl* gene product is required for development as a female, at least in the soma [16,107]; its

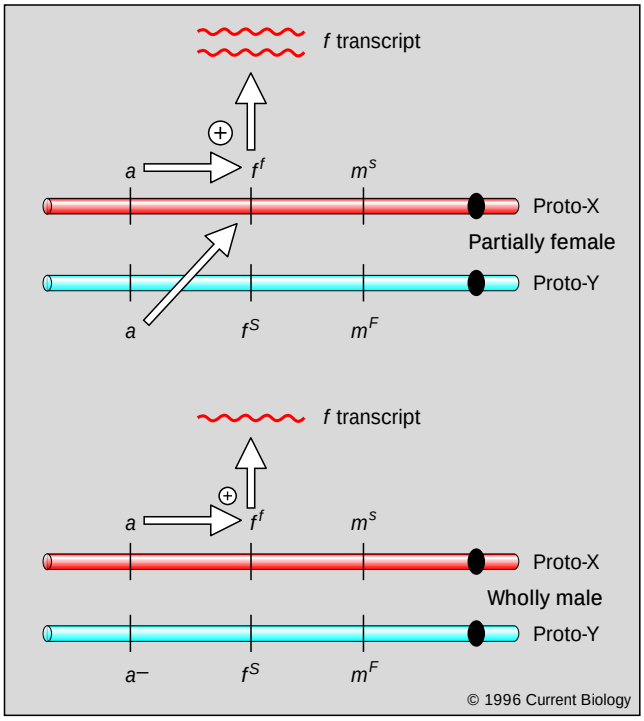
role in the germ line is less clear [108]. Loss-of-function mutations in *Sxl*, which lead to failure of development of female carriers, can be wholly or partially dominant, depending on genetic background [107], which is consistent with the postulated behaviour of f^S.

It is useful to note here that partial development as a male — that is, development as a cosexual biased towards maleness — is compatible with an extension to the scenario discussed earlier for the evolution of proto-X and proto-Y chromosomes. In this extension, the second stage of evolution to dioecy involves partial rather than complete maleness [20]. If this is the case, further genetic factors suppressing femaleness in the presence of f^S are needed to complete the evolution of dioecy. This could be achieved either by epistatically-interacting alleles that specifically suppress female function in cosexuals carrying f^S, or by close linkage to f^S of non-specific female-suppressor alleles. This more gradual evolution to dioecy is plausible from the point of view of the population genetic mechanisms involved, and is consistent with data on the evolution of plant sexual phenotypes [20].

By this route, the formation of a proto-Y chromosome carrying f^S and m^F creates individuals who are partially male in phenotype. In order for X/A sex determination to evolve from this state, the further step is postulated of the evolution of a Y-chromosomal loss-of-function allele that reduces expression of the single copy of f^f in males, leading to a higher probability of maleness (Fig. 5). Assume that the functional allele at the locus in question (carried initially on both the proto-X and proto-Y) is *a*, and that the loss-of-function allele is a^-. Assume also that $a/a^- f^S/f^f$ individuals have a lower probability of expressing f^f compared with $a/a\ f^S/f^f$ individuals, and so have a higher probability of developing a fully male phenotype. The replacement of *a* by a^- on the proto-Y chromosome, but its retention on the proto-X, produces a situation in which the sex of a developing individual is likely to be female if it has two doses of the proto-X chromosome (carrying two doses of *a*), but to be male if it has only one dose of the proto-X (as f^S is a loss-of-function allele, its presence on the proto-Y is irrelevant if f^f expression is repressed). This argument can easily be extended to multiple genes with a mode of action similar to that of *a*. There is an obvious analogy between the *Drosophila* X/A ratio signalling genes *sis-A*, *sis-B*, *sis-C* and *runt*, which help to maintain the activity of *Sxl* when present in double dose [16], and these hypothetical regulators of f^f expression.

There are two possible reasons why a^- alleles might spread through a population of proto-Y chromosomes. The first is that the greater degree of maleness of $a/a^- f^S/f$ compared with $a/a\ f^S/f^f$ individuals, on an m^F/m^s background, is selectively advantageous under conditions when the evolution of dioecy is favoured. Conversely, any

Figure 5



A path for the evolution of X/A sex determination. It is assumed that development as a female is promoted by the product of the *f* locus. If this is above a certain threshold, individuals can develop fully or partially as females. The product of the *a* locus promotes activity of the *f* locus. f^f/f^S individuals still produce enough *f* product to be partially female when sufficient *a* product is present (as in *a/a* homozygotes), but not when the amount of *a* product is insufficient (as in a/a^- heterozygotes).

impairment of the expression of f^f on an m^s/m^s background will be disadvantageous. There is therefore sexual antagonism between the fitness effects on males and females of *a* and a^-, favouring Y-linkage of a^- [3,24]. The second possibility is that a^- is neutral in combination with f^S and m^F, although deleterious in combination with f^f and m^s, and so simply drifts to fixation within the population of proto-Y chromosomes. This seems less plausible. By analogy with the functions of the *Drosophila* X/A ratio signalling genes in developmental processes unconnected with sex determination [16], the *a* gene product might well serve functions other than sex determination, so that a complete loss-of-function allele could incur a fitness cost that would prevent its spread through the population of proto-Y chromosomes in the absence of a countervailing advantage.

A slightly different scenario must be postulated to explain the X/A balance mechanism in *C. elegans*, where XX individuals develop as hermaphrodites and X0 individuals as males [18]. This system is probably a result of secondary evolution to hermaphroditism from the XX (female), X0 (male) system found in most other species of *Caenorhabditis* [7], so that XX individuals will be described here as female.

In this case, the locus *xol* plays a role similar to that of *Sxl* in *Drosophila*, except that *xol* activity represses female development [18]. The proto-Y chromosome allele f^S in an ancestor of *Caenorhabditis* with X/Y sex determination might therefore correspond to a constitutive allele of *xol*. In this case, the *a* gene product must turn off activity of the *f* locus, if present in sufficient dose. An a^- mutation on the proto-Y chromosome that enhances *f* expression might be favoured because it allows fuller expression of the male pathway. There is evidence for several X-linked loci that control the expression of *xol*, consistent with this scenario [104]. The accumulation of mutations of this kind at several loci like *a*, coupled with the evolution of a general enhancement of gene activity on the X chromosome at the expense of the Y, might lead to a situation in which the activity of *f* on the Y chromosome is no longer required, as sufficient *f* gene product is produced on the X. This would permit the total loss of gene activity on the Y chromosome, and pave the way to the final loss of the Y chromosome (see below).

The further evolution of the system under both the *Drosophila* and *Caenorhabditis* scenarios follows a similar path. Once an a^- mutation has been fixed on the proto-Y chromosome, there is no selective pressure to maintain the integrity of the locus on the proto-Y, and it would probably eventually be lost through further mutation and genetic drift, or by the evolution of reduced Y-chromosome activity and dosage compensation. This would lead to a situation in which only the X chromosome carries alleles that differentially regulate the activity of *f* in males and females, as is now the case in *Drosophila* and *Caenorhabditis*. Once this has happened, there would no longer be a requirement for the presence of the *f* locus on the Y chromosome, and null mutations could spread by drift, or even be favoured by selection because they lower the probability of *f* gene expression in males. Eventually, all trace of the *f* locus (or loci) would disappear from the Y chromosome.

X0 sex determination

The evolution of XX/X0 sex determination systems is also a puzzle that requires some exploration [109]. It is difficult to see how this mechanism could be anything other than the end product of the erosion of Y-chromosomal gene activity that is observable in so many systems. However, for the complete elimination of functional genes from the Y chromosome to occur, X/A sex determination must have evolved, and any genes required for male function must first be removed to another chromosome or be replaced by other loci that fulfill their functions.

It is easy to devise a scenario in which a three-break translocation between the Y chromosome and an autosome could result in the transposition of male-essential Y-linked genes to an autosome. The fixation of such a translocation by drift, or by selection for a pleiotropic position effect, would eliminate the need to maintain the Y chromosome [109]. Alternatively, the fact that the path to male development is shut down in XX individuals means that mutations from m^s to m^F on the X chromosome are neutral, once an X/A balance system has been established, or possibly even advantageous if the product of the *m* locus has functions other than male fertility. The restoration of *m* activity on the X would permit the erosion of activity of the Y-linked allele; once this has gone to completion, the Y chromosome can be dispensed with.

Evidence on the evolution of X/A sex determination and X0 sex chromosomes

It is hard to see how critical evidence could be obtained on the evolution of the X/A balance mechanism from an X/Y sex-determination system, other than by characterizing the genes concerned in a group such as *Rumex*, where both mechanisms can be found in species of the same genus [17]. This would need to be coupled with a phylogenetic analysis of this group, in order to establish whether or not the X/Y system is indeed primitive, as is widely assumed. Circumstantial evidence supporting this assumption is provided by comparative evidence showing that X/Y sex determination is taxonomically far more widely distributed than X/A, especially in groups, such as fishes and plants, with relatively poorly developed sex chromosome systems [3].

Conclusions

If the mechanisms of sex chromosome evolution discussed here have been important, then a widespread feature of genomic organization in higher organisms has evolved in response to the steady input of deleterious alleles into the population by recurrent mutation. At first sight, this seems contrary to the orthodox Fisherian conclusion that mutation pressure plays a minor role in controlling the direction of evolutionary change [30]. But this conclusion is based on the demonstrable effectiveness of selection against the very low rate of mutation to deleterious alleles at individual loci, in large sexually reproducing populations [30,110]. When the relevant conditions are not fulfilled, it should occasion no surprise that the conclusion breaks down. If the selection pressure is reduced to the same order of magnitude as the mutation rate or the reciprocal of the effective population size, mutations that were formerly held at low frequencies can accumulate, either deterministically under the pressure of recurrent mutation, or stochastically by genetic drift [111]. The accumulation of loss-of-function mutations in pseudogenes [63], and the loss of many components of the chloroplast genome in parasitic species of flowering plants [112], illustrate this principle.

The mechanisms for degeneration of the Y chromosome discussed here rely on the reduction in the ability of selection either to remove deleterious mutations from the population or to fix beneficial mutations when genetic recombination is absent from a sizeable portion of the

genome, provided that the mutation rate to deleterious alleles for this region as a whole is sufficiently large. This is a phenomenon which was first pointed out by Fisher himself [30]. In the light of subsequent theoretical work, and increasingly solid evidence that there is a high mutation rate per genome to deleterious alleles in higher eukaryotes [37–39], it seems virtually certain that one or more of the processes that involve this general principle have played a major role in the evolutionary erosion of Y chromosomes. It also seems likely that, as suggested by its discoverer [113], the phenomenon of dosage compensation is an adaptive response to the loss of gene activity on the Y chromosome. While a good deal of further research is needed to check the validity of the assumptions of the various theories discussed here, and to discriminate among them by testing their predictions, there is reason to be optimistic that we now have a useful general framework for thinking about the evolution of sex chromosomes.

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