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Genetic linkage between a sexually selected trait and X chromosome meiotic drive

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Previous studies on the stalk-eyed fly, *Cyrtodiopsis dalmanni*, have shown that males with long eye-stalks win contests and are preferred by females, and artificial selection on male relative eye span alters brood sex-ratios. Subsequent theory proposes that X-linked meiotic drive can catalyse the evolution of mate preferences when drive is linked to ornament genes. Here we test this prediction by mapping meiotic drive and quantitative trait loci (QTL) for eye span. To map QTL we genotyped 24 microsatellite loci using 1228 F2 flies from two crosses between lines selected for long or short eye span. The crosses differed by presence or absence of a drive X chromosome, X^D, in the parental male. Linkage analysis reveals that X^D dramatically reduces recombination between X and X^D chromosomes. In the X^D cross, half of the F2 males carried the drive haplotype, produced partially elongated spermatids and female-biased broods, and had shorter eye span. The largest QTL mapped 1.3 cM from drive on the X chromosome and explained 36% of the variation in male eye span while another QTL mapped to an autosomal region that suppresses drive. These results indicate that selfish genetic elements that distort the sex-ratio can influence the evolution of exaggerated traits.

Keywords: *Cyrtodiopsis dalmanni*; sex-ratio; mate choice; meiotic drive

1. INTRODUCTION

In many species females prefer mating with males that display extraordinary ornaments (Andersson 1994). Some hypotheses for the evolution of female mate choice assume that the expression of a male ornament indicates genetic benefits that offspring can inherit (Pomiankowski 1987; Grafen 1990; Iwasa *et al.* 1991). Females could choose mates that are in better condition (Andersson 1986), perhaps because they carry fewer deleterious mutations (Pomiankowski 1988) or are more resistant to pathogens (Hamilton & Zuk 1982). Alternatively, females might choose males with a trait that, due to pleiotropy or linkage, directly indicates a gene or gene region that influences offspring fitness. While evidence for good genes effects is accumulating (Møller & Alatalo 1999), the nature of the genetic benefits indicated by male ornaments remains obscure in most cases.

Flies in the family Diopsidae have eyes at the ends of stalks that in some males can greatly exceed their body length. Sexual dimorphism in eye-stalks varies across species (Burkhardt & de la Motte 1985; Wilkinson & Dodson 1997) and is probably due to sexual selection: males use eye span to assess rivals in combat (Panhuis & Wilkinson 1999), and females prefer to mate with males that have long eye span relative to body length (Burkhardt & de la Motte 1988; Wilkinson *et al.* 1998*a*; Hingle *et al.* 2001). The ratio of eye span to body size (relative eye span) exhibits condition dependence (David *et al.* 2000). The allometric relationship between male eye span and body length has increased in highly dimorphic diopsid species (Wilkinson & Taper 1999; Baker & Wilkinson 2001), which allows eye span to indicate body size better than body size itself, assuming both are measured with comparable accuracy. In sexually dimorphic species of the genus *Cyrtodiopsis* some males produce predominantly daughters as a consequence of X chromosome meiotic drive (Presgraves *et al.* 1997; Wilkinson *et al.* 2003).This phenomenon occurs when males that possess a driving X chromosome (X^D) pass X^D-bearing sperm but few or no Y-bearing sperm to females (Cazemajor *et al.* 2000) and results in the production of highly female-biased broods. With a two fold transmission advantage an X^D chromosome should spread rapidly and lead to female-biased populations that, in the absence of opposing selection or suppression, go extinct (Hamilton 1967). In many taxa where X drive exists, the sex-ratio (SR) phenotype is associated with one or more inversions on the driving X chromosome (Jaenike 2001; Jutier *et al.* 2004). These inversions are thought to restrict recombination and maintain linkage disequilibrium among drive and responder loci, such that X^D gene products do not destroy X^D-bearing sperm (Haig & Grafen 1991). In some species with X drive, loci on the Y chromosome or autosomes completely or partially suppress the effects of the driving X chromosome on brood sex-ratio (Stalker 1961; Atlan *et al.* 1997; de Carvalho *et al.* 1997; Jaenike 1999; Atlan *et al.* 2003).

The presence of sex chromosome meiotic drive may provide a reason for females to mate non-randomly. Females that avoid mating with SR males should produce more sons who will have enhanced reproductive success in a female-biased population (Lande & Wilkinson 1999). In *Cyrtodiopsis dalmanni*, both male relative eye span and female preference respond to artificial selection (Wilkinson 1993; Wilkinson & Reillo 1994) and lines selected for short eye span show higher frequencies of

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males with female-biased broods than lines selected for long eye span (Wilkinson *et al.* 1998*b*). This association between eye span and brood sex ratio suggests that females might use eye span as an indicator of X drive or drive suppression. Simulations reveal that selection for an equal sex-ratio allows autosomal genes for female preference to spread if the genes for the male trait and SR are tightly, but not perfectly, linked on an X chromosome (Lande & Wilkinson 1999). If the loci affecting eye span and brood sex ratio never recombine, then strong female preference should eventually eliminate drive. Linked suppressor and ornament genes on a Y chromosome should not evolve, in contrast to earlier suggestions (Wilkinson *et al.* 1998*b*), because such a polymorphism is unstable (Lande & Wilkinson 1999; Reinhold *et al.* 1999).

Here we test whether the conditions for X drive to catalyse sexual selection exist in *C. dalmanni*. We do this by mapping the location of SR in males relative to quantitative trait loci (QTL) for eye span using a cross between flies that were selected for long and short relative eye span. The presence of a major QTL for eye span tightly linked to SR would support the hypothesis that sex chromosome meiotic drive facilitated the evolution of female preferences for extreme eye-stalks. However, linkage disequilibrium between a QTL for eye span and SR could be a consequence of artificial selection rather than physical linkage. To examine this possibility we also map X-linked eye span QTL in a cross between selected line flies lacking SR.

2. MATERIAL AND METHODS

(a) *Experimental crosses*

To map SR and QTL for eye span we conducted two F2 intercross experiments between different pairs of replicate lines of *C. dalmanni* selected for either long (high line) or short (low line) male relative eye span. Below we use 'experimental cross' to indicate that X^D was present in the parental male and 'control cross' to indicate that X^D was absent in the parental male.

After 32 generations of selection, we initiated the control cross by mating a high line standard (ST, i.e. no biased sex-ratio) male with a low line female. We then mated F1 sons each to a sister in 2.5 l plastic jars, allowed females to oviposit in cups of pureed corn collected twice each week, and subsequently used the F1 family that produced the largest number of F2 offspring. In the QTL analysis we used 490 F2 flies from this family: 259 males and 231 females, a sex-ratio that did not differ from unity ($X^2=1.6$, $p=0.21$). Unbiased F1 family sex ratios, as well as the X-linked haplotypes of F2 males (see below), indicated that the X^D chromosome was not present in either the male or female parent. Thus F1 females had two standard X chromosomes, one each from selected high and low lines, X_L/X_H, and F1 males had the standard low line X chromosome, X_L.

We began the experimental cross after 45 generations of selection by mating a low line SR male with a high line female. We intended this cross to produce F1 females with a single low line X^D chromosome and a standard high line X chromosome, $X^D{}_L/X_H$, and F1 males with a high line X chromosome, X_H. SR and ST males were present in both replicate low lines after 22 generations of selection (Wilkinson *et al.* 1998*b*). We let males from the low line replicate with the shortest relative eye span mate with three virgin females and we tallied the sex of all progeny produced during two weeks of oviposition. Four of 18 males produced less than 10% male offspring, suggesting that X^D frequency was roughly 22% at generation 45. We mated the male with the most extreme sex-ratio to a virgin high line female to start the cross. They produced six sons and more than 300 daughters. We then mated each of the six F1 males to a sister, allowed females to oviposit until death, froze female progeny after eclosion, and kept 310 male progeny from three families alive for at least a month before scoring spermatocysts for evidence of meiotic drive (see below). We used 738 F2 flies from two F1 families of the experimental cross for QTL mapping. Three families were frozen immediately after eclosion and had equal sex ratios: 124 males, 102 females ($X^2=2.1$, $p=0.14$), 70 males, 63 females ($X^2=0.4$, $p=0.54$), and 34 males, 49 females ($X^2=2.7$, $p=0.10$). The two families used in the QTL analysis contained 194 males, 271 females ($X^2=12.8$, $p=0.0004$) and 76 males, 197 females ($X^2=53.6$, $p<0.0001$). The female-biased sex-ratio in these families is due, at least in part, to male mortality after eclosion but prior to dissections of testes.

In the lines that produced the flies for these crosses, selection on the ratio of eye span to body length produced change in eye span largely independent of body size (Wilkinson 1993; Wolfenbarger & Wilkinson 2001). Although the two crosses used parental flies from different generations and lines, the difference in mean relative eye span between the two pairs of lines was similar because the response to selection reached a plateau after 30 generations (Wilkinson *et al.* 2005). The differences between the lines for males and females, respectively, expressed as standard deviations of relative eye span in the base population, were 6.7 and 4.2 s.d. at generation 32, and 7.2 and 4.9 s.d. at generation 45. The high line we used at generation 32 had a mean ($\pm$s.e.) male eye span of 10.2 ± 0.05 mm and body length of 7.3 ± 0.03 mm while the low line had a mean male eye span of 8.8 ± 0.05 mm and body length of 7.5 ± 0.03 mm. At generation 45 mean male eye span and body length measured 10.1 ± 0.10 and 7.4 ± 0.04 mm for the high line and 7.7 ± 0.05 and 7.0 ± 0.03 mm for the low line.

(b) *Phenotype measurement*

The eye span, body length, and thorax width of all F2 males and females were measured from 11$\times$ dissecting scope images at a resolution of 50 pixels per mm using NIH Image, version 1.59[1]. In the experimental intercross we scored spermatocyst morphology from 310 F2 males for evidence of drive by staining testes with 10^{-7} M Hoechst 33258 (Sakaluk & O'Day 1984; Wilkinson & Sanchez 2001), and examined at least 10 disassociating cysts from each at 200$\times$ magnification with UV fluorescence as described elsewhere (Wilkinson & Sanchez 2001). To determine the degree to which cyst morphology predicts brood sex-ratio, we mated a subset of 59 F2 males each to three virgin outbred females and collected offspring for three weeks before examining cysts. Screened males produced an average of 164 ± 9 offspring. We tested brood sex ratios for deviation from 1 : 1 using a chi-squared test corrected for continuity. Males that produced highly female-biased sex ratios (less than 20% male and $p<0.0001$) were designated SR males and assumed to carry an X^D chromosome while all others were scored as ST males. Males producing significant, but weakly biased sex ratios (32–43% male, $0.001<p<0.01$) were labelled partial SR males.

(c) *Genotype assignment*

We extracted genomic DNA from flies in the control cross using phenol–chloroform and ethanol precipitation (Sambrook & Russell 2001) and in the experimental cross with Qiagen DNeasy kits (Qiagen, Valencia, CA). We screened 52 microsatellite markers described elsewhere (Wright *et al.* 2004) to identify 24 informative markers. We used fluorescently labelled primers in 5.5 μL PCR reactions (Wright *et al.* 2004) to amplify four X-linked loci for all 1228 F2 flies from both crosses and 20 autosomal loci for 738 flies from the experimental cross. We separated amplification products by capillary electrophoresis using an ABI 3100 (Applied Biosystems, Foster City, CA) and used GeneScan 3.1.2 (Applied Biosystems) and Genotyper 2.5 (Applied Biosystems) to assign genotypes to individuals.

(d) *Linkage and QTL analysis*

We excluded five autosomal loci from the experimental cross-dataset because the genotypes at those loci failed to conform to expected segregation patterns. Then for each family we used Joinmap v. 3.0 (Van Ooijen & Voorrips 2001) to assign the remaining 19 marker loci and SR phenotype to linkage groups using Kosambi map distances. We incorporated sex-specific segregation expectations of X-linked loci in Joinmap by assigning backcross segregation expectations to females and doubled haploid segregation expectations to males. Because some microsatellite loci were informative in only one family, we used Joinmap to produce a joint linkage map for males and females from both families. We then used this linkage map to constrain marker order for each sex before estimating QTL separately for males and females.

To locate QTL for eye span we first corrected for any differences in body length and, in the experimental cross, between families, by calculating least squares (LS) estimates for the eye span of each male and female fly. Then, using the linkage map obtained for each sex we estimated QTL location and effect size by composite interval mapping (Zeng 1994) using MapManager QTXb20 (Manly et al. 2001). Each sex was analysed separately. In analysing each chromosome in the experimental cross, we controlled for a single background locus near the largest significant QTL on each of the other two chromosomes. We performed 1000 genome-wide permutation tests to determine likelihood ratio (LR) critical values above which QTL were significant at $p<0.001$ (Doerge & Churchill 1996). We confirmed QTL location by recording the maximum LR obtained in each of 1000 bootstrap samples for each chromosome (Walling *et al.* 1998). If more than one QTL was indicated by the bootstrap analysis, we included a background locus from that chromosome in the analysis.

We used JMP v 5.0.1.2 (SAS Institute, Cary, NC) to perform all other statistical analyses.

3. RESULTS

(a) *Meiotic drive and sperm morphology*

F2 males from the experimental cross produced two distinct types of spermatocysts. Twenty-seven (46%) of the males screened for brood sex-ratio contained cysts in which about half of the 128 spermatid heads were not fully elongated and resembled singed-hairs (figure 1*a*) while the other 32 males had normal, uniformly elongated spermatids (figure 1*b*). The 59 F2 males also produced a bimodal distribution of brood sex ratios (figure 1*c*), where males

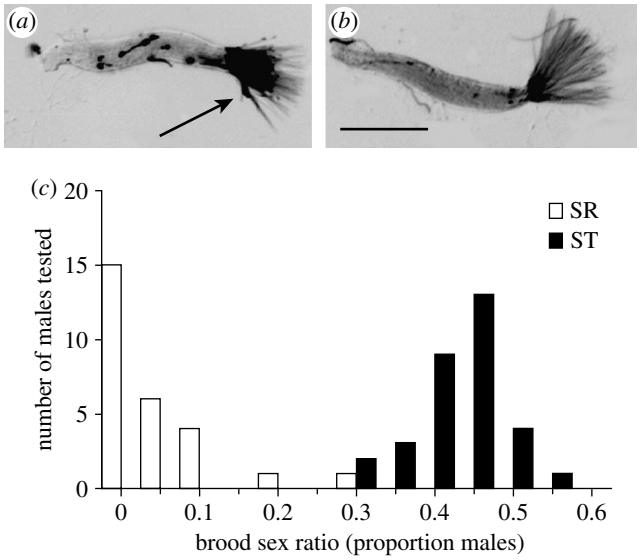

Figure 1. Inverted images of (*a*) SR and (*b*) non-SR male spermatocyst bundles in the process of differentiating into 128 spermatids. DNA stained with Hoechst appears dark. Arrow points to non-elongated spermatid heads found in the majority of cysts from SR males. Bar indicates 50 μm. (*c*) Distribution of brood sex ratios for 59 F2 males. Open bars indicate males with SR cysts and filled bars indicate males with normal cysts.

containing non-elongated spermatids sired broods with a lower percentage of sons ($6.5\pm1.5\%$) than males with normal cysts ($44.9\pm1.1\%$; $t=21.6$, $p<0.0001$). Spermatid elongation predicted brood sex-ratio correctly for all males with either highly female-biased ($n=26$) or equal ($n=26$) sex ratios. Only one of the seven males with partial SR brood sex ratios had cysts like an SR male. In total, 159 males contained incompletely elongated spermatids and were scored as SR and 151 were scored as ST, consistent with a 1 : 1 ratio ($X^2=0.2$, $p>0.5$).

(b) *Linkage maps*

Linkage analysis of the control cross revealed that the four X-linked microsatellite markers produced a single linkage group that spans 32.7 centimorgans (cM) in males and 29.7 cM in females (figure 2). Support for this marker order was high with logarithm of odds (LOD) scores between markers ranging from 16.4 to 47.7. Linkage analysis of the experimental cross, in which X^D was segregating, yielded three linkage groups, consistent with the chromosome complement reported for *C. dalmanni* (Wolfenbarger & Wilkinson 2001). LOD scores between markers ranged from 9.2 to 101.0. In males, eight markers spanned 42.5 cM on the first linkage group and seven markers spanned 35.6 cM on the second linkage group (figure 2). In females, the same markers spanned 56.4 and 27.5 cM, respectively. However, in contrast to the control cross we found little evidence of recombination between X and X^D chromosomes. In the experimental cross, X-linked loci spanned only 5.2 cM in females, while in males all four X-linked loci segregated together 5.4 cM from the SR phenotype, as scored by cyst morphology (figure 2). We found only three X haplotypes at the four microsatellite loci among males from the experimental cross (table 1). This observation is consistent with three different X haplotypes in the two parental flies of the experimental cross. In contrast, among the F2 males of the

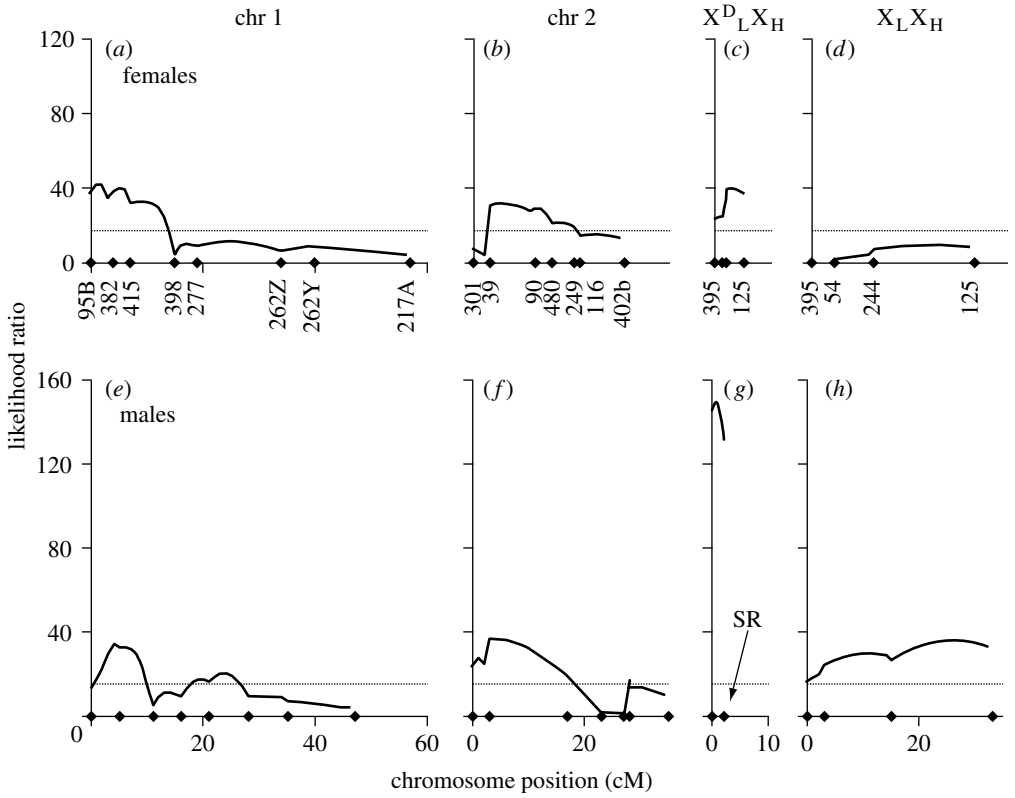

Figure 2. Quantitative trait loci (QTL) estimated by composite interval mapping for eye span, adjusted for body length by least squares, in (*a–d*) female and (*e–h*) male stalk-eyed flies. Chromosome source is listed above each panel. (*d*,*h*) X-linked QTL, estimated from an intercross without an SR parental male. (*a–c*,*e–g*) QTL estimated from F2 flies segregating for X^D. Closed diamonds and labels indicate the position and identity (Wright *et al.* 2004) of microsatellite markers. Note that in (*g*) (X^D male QTL), all four X-linked markers cosegregate 2.1 cM from SR. Dotted lines indicate likelihood ratio (LR) significance thresholds for $p<0.001$.

Table 1. Number of F2 males with sex-ratio (SR) or standard (ST) sperm phenotypes by X haplotype.

haplotype[a]	SR	ST
BABC	138	8
ABAB	1	41
BBAA	4	110

[a] Letters refer to alleles at four X-linked microsatellite loci (Wright *et al.* 2004): ms-54 ($A=160$ bp, $B=162$ bp), ms-125 ($A=148$ bp, $B=153$ bp), ms-244 ($A=226$ bp, $B=236$ bp), and ms-395 ($A=202$ bp, $B=216$ bp, $C=238$ bp), respectively.

control cross we found eight X haplotypes, which included the two unique parental and all six single-recombinant haplotypes.

X haplotype predicted spermatocyst morphology in 297 of the F2 males in the experimental cross (table 1). Ninety-seven percent of the males scored as SR had the same X haplotype (henceforth referred to as the X^D haplotype) while 95% of the ST males had one of two other X haplotypes ($X^2=248.6$, $p<0.0001$). Three X-linked loci had unique alleles present only in SR males and not in any of the seven partial SR males in the experimental cross or in any of the F2 flies from the control cross.

The 13 mismatches between haplotype and cyst morphology could be due to recombination between the X-linked marker loci and SR or to autosomal factors that suppress drive. In eight of the 13 mismatches, normal cysts were found with the X^D haplotype, consistent with drive suppression. Three adjacent loci on the first linkage group exhibited highly significant associations (MS392: $X^2=20.5$, $p<0.0004$; MS415: $X^2=25.7$, $p<0.0001$; MS398: $X^2=12.4$, $p<0.0020$) between genotype and these eight mismatches, strongly implicating chromosome 1 as the location of a suppressing factor. After Bonferroni correction ($p<0.0025$) no other marker exhibited a significant association between mismatch and genotype. If the remaining five exceptions (males with non-drive haplotypes and SR cysts) are due to recombination, then the map distance between the four X-linked markers and the SR element is 2.1 cM.

(c) *Quantitative trait loci*

In the experimental cross, we found five significant QTL with bootstrap support for LS eye span in males and females (figure 2). On the first chromosome we found three QTL in males, explaining 7, 3, and 7% of the variation in eye span, and two QTL in females, explaining 9 and 7% of the variation in eye span. The three adjacent marker loci implicated in suppressing SR occur in the same region as these QTL. On the second chromosome we found a single QTL in males that explained 7% and two in females that explained 7 and 6% of the variation in eye span. The QTL with greatest additive effect, accounting for 36% of the variation in male eye span (figure 3), was on the X chromosome, 1.3 cM from SR (figure 2*g*). The corresponding X-linked QTL in females explained 9% of eye span variation. Data from the control cross revealed two X-linked QTL in males, which

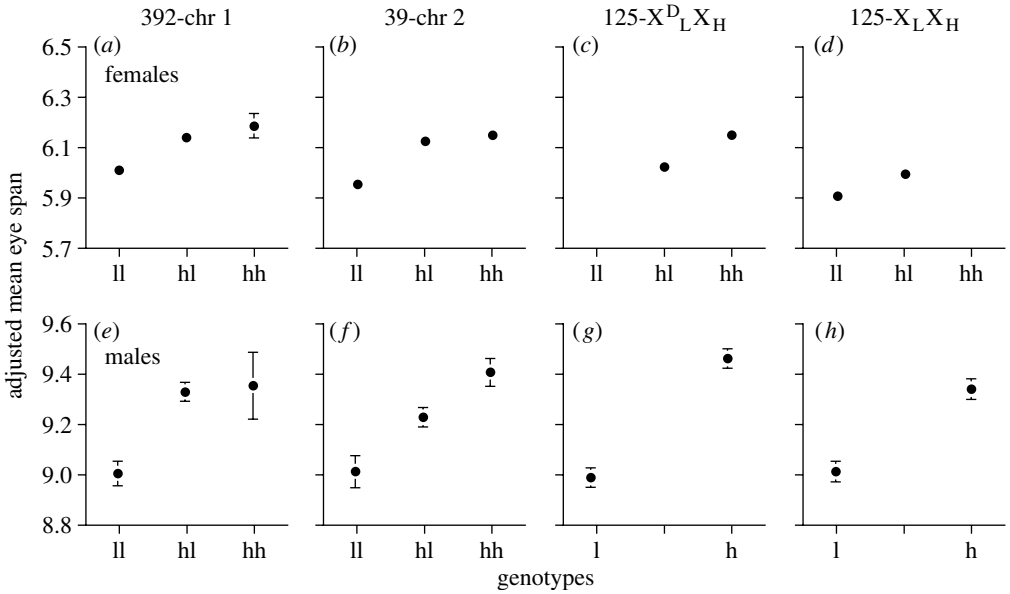

Figure 3. Mean (±s.e.) eye span, adjusted for body length by least squares, for (*a–d*) female and (*e–h*) male F2 flies that were homozygous or heterozygous at microsatellite loci (Wright *et al.* 2004) closest to the QTL of largest effect on each chromosome. Source for alleles is given below each panel, where l and h refer to selected low and high lines, respectively. (*c*,*g*) The X-linked low line allele is inherited from an SR parental male. (*d*,*h*) Means for F2 flies from the control intercross with a standard parental male (see text).

explained 8 and 13% of eye span variation, and no significant QTL for females (figure 2).

The X-linked QTL estimated for male eye span in the experimental cross was negatively associated with SR: LS eye span of SR males (8.9±0.03 mm) was shorter than that of ST males (9.4±0.03 mm; $F_{1,262}=85.1$, $p<0.0001$). Furthermore, when we estimated the strength of the QTL by regressing LS male eye span on the genotype of the marker nearest the largest QTL on each chromosome, we found that the effect of the X-linked QTL from the experimental cross was greater than the X-linked effect from the control cross (table 2, figure 3).

In contrast to the results for LS eye span, in the experimental cross we found no significant QTL for male body length and only a single significant QTL for female body length (LR=35.2, $p<0.001$) on the second linkage group.

Table 2. Variance[a] (%) in LS eye span explained by regression on marker genotype.

chromosome	marker[b]	males	females
1	ms-392	19	
	ms-95		14
2	ms-39	8	6
X^D	ms-125	34	6
X	ms-125	14	4

[a] All regressions are significant with $p<0.0001$.
[b] Microsatellite marker (Wright *et al.* 2004) nearest the QTL of largest effect on each chromosome.

4. DISCUSSION

The evidence presented here indicates that SR males in our experimental cross possess an X chromosome, X^D, that is unambiguously associated with a single X haplotype at four microsatellite loci. This X chromosome influences spermatocyst development by preventing full elongation of presumably Y-bearing spermatids. Males that produce weakly biased female sex ratios do not carry this X^D chromosome. While the SR phenotype is also present in a related species, *C. whitei* (Presgraves *et al.* 1997), how drive alters spermatogenesis appears to differ. In *C. whitei* drive is predicted better by the presence of Hoechst-staining material in the tail of a sperm bundle (Wilkinson & Sanchez 2001) than by incomplete elongation of spermatid heads. These differences may represent two points on a temporal continuum of spermatid elongation.

Linkage analysis revealed that the presence of the X^D chromosome dramatically influences recombination. In the cross lacking the X^D chromosome, the four microsatellite markers spanned 33 cM in males, whereas they were 0 cM apart in the cross where the parental male carried the X^D chromosome. This difference may be due to an X-linked paracentric inversion that is large enough to contain all four microsatellites and renders recombinant haplotypes inviable in male eggs. Chromosomal inversions are associated with meiotic drive in several other cases of SR in Diptera (Jaenike 2001). Comparison of two-locus autosomal haplotypes in F2 flies indicates that recombination does not occur in males (G. S. Wilkinson, unpublished data), as is true for other Diptera (Lenormand 2003).

The QTL analysis indicates that male ornament genes may be sex-linked and have sex-biased effects. Depending on the cross, we found either five or six QTL for eye span in males and five or four QTL for females, which is similar to previous estimates for the number of segregating factors that influence eye span (Wolfenbarger & Wilkinson 2001). It should be noted, though, that each cross generated only two of the three possible X-linked female genotypes. Assuming additive gene action, any X-linked effect in females should, therefore, be doubled to compare to males. Even after doubling, X-linked QTL explain 12% of the variance in experimental cross females, which is three fold less than the 36% explained in males. These results are consistent with low, but significant, genetic

correlations between male and female eye span indicated by correlated responses to selection (Wilkinson 1993) and by phylogenetic correlations (Baker & Wilkinson 2001; Baker & Wilkinson 2003). The absence of body length QTL in males confirm that the effects we measured are not due to incidental selection on body size or condition in the selected lines.

One consequence of a paracentric inversion is that inverted chromosomal regions can evolve independently of non-inverted regions. For example, the SR X chromosome of *Drosophila pseudoobscura* shows greater sequence similarity to the SR X chromosome of *D. persimilis* than to the standard X chromosome of *D. pseudoobscura* (Babcock & Anderson 1996). Consequently, inversions may contain unique alleles with different phenotypic effects. A paracentric inversion on the X^D chromosome could include a unique allele for eye span, which would explain why the X-linked eye span QTL estimated from the experimental cross had a greater effect than the X-linked QTL estimated from the control cross.

Another consequence of a paracentric inversion is that genes in the inverted region will be inherited together as a single Mendelian factor. Thus, our QTL analysis revealed two QTL for male eye span on the X in the control cross, but only a single QTL of large effect in the experimental cross. Furthermore, the X^D haplotype and SR phenotype were almost always inherited together. Most of the mismatches between X haplotype and SR phenotype were associated with an 11 cM region on the first chromosome, a region that also carries QTL for both male and female eye span. These results suggest that male eye span is influenced both by the type of X chromosome a male carries and by the presence or absence of at least one autosomal factor that can modify the expression of drive.

Although most good genes models of sexual selection assume that ornament expression indicates male genetic quality at many loci (Pomiankowski 1988), we find that the largest QTL for a male ornament are associated directly or indirectly with X chromosome meiotic drive. Thus, male eye span in *C. dalmanni* advertises the presence or absence of a selfish genetic element that can distort brood sex ratios or a factor that suppresses this effect. Diopsid stalk-eyed flies are not the only organisms in which meiotic drive or other selfish genetic elements may influence mate preference (Lenington *et al.* 1994; Randerson *et al.* 2000; Moreau *et al.* 2001). However, in these other systems, biased sex ratios are not known to be associated with ornaments. Elaborate head ornaments have evolved several times among flies (Wilkinson & Dodson 1997) and sex chromosome meiotic drive is common in this order (Jaenike 2001), which suggests that the general phenomenon we describe should be investigated in other taxa.

Hypotheses for the evolution of mate choice have historically had difficulty explaining how genetic variation for a male trait and genetic quality are maintained, given strong female preference (Charlesworth 1987). We presume that variation in genetic quality is maintained, in part, by balancing selection on the X^D chromosome and continuous evolution of new drive and suppressor elements (Jaenike 2001; Jutier *et al.* 2004). SR polymorphisms are present in multiple populations of *C. dalmanni* and *C. whitei* (Wilkinson *et al.* 2003) and more than one X^D chromosome has been detected in a single population of *C. dalmanni* (P. M. Johns & G. S. Wilkinson, unpublished data), indicating that the drive system is both dynamic and persistent in these flies. Association mapping studies could determine if all SR elements indicate eye span or if some are no longer informative as a consequence of past recombination, as predicted (Lande & Wilkinson 1999).

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ENDNOTE

[1]http://rsb.info.nih.gov/nih-image/index.html.

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As this paper exceeds the maximum length normally permitted, the authors have agreed to contribute to production costs.