Genet. Res., Camb. (2003), **81**, *pp.* 213–220. © 2003 Cambridge University Press
DOI: 10.1017/S0016672303006190 Printed in the United Kingdom

Quantitative genetic analysis of among-population variation in sperm and female sperm-storage organ length in *Drosophila mojavensis*

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(*Received 27 August 2002 and in revised form 30 December 2002*)

Summary

In *Drosophila*, sperm length and the length of the females' primary sperm-storage organ have rapidly coevolved through post-copulatory sexual selection. This pattern is evident even among geographic populations of *Drosophila mojavensis*. To understand better these traits of potential importance for speciation, we performed quantitative genetic analysis of both seminal receptacle length and sperm length in two divergent populations. Parental strains, F_1, F_1 reciprocal (F_{1r}), F_2, F_{2r}, backcross and backcross reciprocal generations were used in a line-cross (generation means) analysis. Seminal receptacle length is largely an autosomal additive trait, whereas additivity, dominance and epistasis all contributed to the means of sperm length. Either an X-chromosome or a Y-chromosome effect was necessary for models of sperm length to be significant. However, the overall contributions from the X and Y chromosomes to sperm length was small.

1. Introduction

Sperm morphology exhibits dramatic evolutionary divergence (e.g. Jamieson, 1987, 1991; Briskie & Montgomerie 1992; Gage, 1994, 1998; Stockley *et al*., 1996). Rapid sperm evolution is well illustrated by the genus *Drosophila*, in which sperm length varies from 0·23 mm in *Drosophila subobscura* (Snook, 1997) to over 58 mm in *Drosophila bifurca* (Pitnick *et al*., 1995*b*), with giant sperm having evolved many times (Pitnick *et al*., 1995*a*). The selective advantage of longer sperm is especially intriguing given their large energetic and developmental costs (Pitnick, 1993, 1996; Pitnick *et al*., 1995*a*).

Comparative studies of a diverse array of taxa have found a positive relationship between sperm length and the risk of sperm competition (Gomendio & Roldan, 1991; Briskie & Montogomerie, 1992; Gage, 1994; Briskie *et al*., 1997; LaMunyon & Ward, 1998; Balshine *et al*., 2001; but see Hosken 1997; Stockley *et al*., 1997), thus implicating post-copulatory sexual selection as the causal agent driving sperm length evolution. Although these correlational studies cannot establish causation, a recent study of the nematode *Caenorhabditis elegans* demonstrated that the volume of their amoeboid sperm increases when the risk of encountering sperm competition is experimentally increased (Lamunyon & Ward, 2002). This effect was attributable exclusively to male–male sperm competition.

In species with more traditional flagellated sperm, a female role in postcopulatory selection has been suggested (Keller & Reeve, 1995; Eberhard, 1996; Birkhead, 1998; Parker & Partridge, 1998). For example, various dimensions of the female reproductive tract have been found to correlate positively with sperm length in birds (Briskie & Montgomerie, 1993), beetles (Dybas & Dybas, 1981), stalk-eyed flies (Presgraves *et al*., 1999), butterflies (Gage, 1994), moths (Morrow & Gage, 2000) and *Drosophila* (Pitnick *et al*., 1999). Among *Drosophila* species, the females' primary sperm-storage organ, the seminal receptacle (SR), ranges from 0·41 mm to 81·67 mm long and is highly correlated with sperm length ($r^2 = 0{\cdot}900$, $P < 0{\cdot}001$; Pitnick *et al*., 1999). This correlated evolution of sperm and female reproductive tract morphology might be the result of coevolution driven by postcopulatory sexual selection, with female tract morphology serving as the proximate mechanism underlying female sperm choice (Keller & Reeve, 1995; Eberhard, 1996; Birkhead, 1998, 2000; Pitnick *et al*., 1999; Pitnick & Brown, 2000).

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This hypothesis was supported by a recent investigation of the interaction between sperm and SR length using artificially selected lines of *Drosophila melanogaster* (Miller *et al*., 2001; Miller & Pitnick, 2002, 2003). Sperm length evolution was demonstrated to occur as a correlated response to selection on female SR length. Moreover, results of experiments in which males with sperm of different lengths were competed within females with different SR lengths confirms that male fertilization success is largely determined by an interaction between sperm and SR length (Miller & Pitnick, 2002).

As sperm and female reproductive tract morphology are central to successful reproduction, their divergence might contribute to reproductive isolation between populations (Parker & Partridge, 1998; Howard, 1999; Pitnick *et al*., 1999; Eady, 2001). *Drosophila mojavensis* has geographically isolated populations that are considered to be in the early stages of speciation (Markow & Hocutt, 1998). Examination of geographical variation in sperm and SR length throughout the range of *D. mojavensis* has revealed significant among-population differences in both traits in addition to a pattern of strong correlated evolution between them (Pitnick *et al*., 2003).

Few studies have examined the genetics of sperm or female sperm storage organ morphology. Miller *et al*. (2001) found that SR length was heritable and could be fully explained by a completely additive model in *D. melanogaster*. Because of the experimental design, no evaluation of sex-linked genetic effects could be made. Several studies have suggested a predominant role for the X or Y chromosomes in determining sperm length. If true, this would have implications for sexual selection theory because most quantitative genetic models assume autosomal inheritance of characters under strong sexual selection (Andersson, 1994).

Two studies using regression analysis suggest a substantial role for the X chromosome in sperm length. In the dungfly *Scathophaga stercoraria*, Ward (2000) found that a maternal grandfather–grandson regression was greater than a paternal grandfather–grandson regression, implying an X-chromosome contribution to sperm length. Furthermore, the maternal grandfather–grandson regression was nonlinear suggesting that epistatic interactions between the X chromosome and the autosomes contributed to sperm length. In the cricket *Gryllus bimaculatus*, Morrow & Gage (2001) also suggested an X-chromosome contribution to sperm length, noting that the realized heritability after four generations of selection was greater than the heritability measure by father–son regression.

The only genetic analyses of *Drosophila* sperm length have used hybrid crosses between *Drosophila simulans* and *Drosophila sechellia* (sperm lengths of 1·2 mm and 1·6 mm, respectively; Joly *et al*., 1997; Macdonald & Goldstein, 1999). These closely related species can be successfully crossed in one direction only (*D. simulans* females by *D. sechellia* males) to produce fertile hybrid females and sterile hybrid males. Using several recessive markers in a *D. simulans* strain in a backcrossing design, Joly *et al*. (1997) demonstrated that *D. simulans* cyst length (mature cysts were measured as an estimate of sperm length) was autosomally dominant over *D. sechellia*, and that there were strong epistatic interactions between interspecific chromosomes. Although the X-chromosome had no major effect on sperm length, they did find that the Y chromosome of *D. sechellia* in a *D. simulans* background reduced sperm length. By contrast, MacDonald & Goldstein (1999) performed a quantitative trait locus analysis using a similar backcrossing scheme and found that cyst length quantitative trait loci were few and limited to the X chromosome.

The rapid divergence of sperm and SR lengths among populations of *D. mojavensis* (Pitnick *et al*., 2003) allows for detailed intraspecific genetic analysis. Here, we report a quantitative genetic analysis of both an organ of female sperm choice (SR length) and the interacting male character undergoing sexual selection (sperm length). Means analysis was performed on populations collected from Organ Pipe National Monument (AZ, USA) (sperm length $\pm$ SE $= 1{\cdot}847 \pm 0{\cdot}007$; SR length $\pm$ SE $= 4{\cdot}753 \pm 0{\cdot}076$) and from Whitmore Canyon of the Grand Canyon (AZ, USA) (sperm length $\pm$ SE $= 1{\cdot}766 \pm 0{\cdot}013$; SR length $\pm$ SE $= 4{\cdot}099 \pm 0{\cdot}071$), hereafter referred to as the high (H) and low (L) lines, respectively. These two populations were chosen because of their relatively large differences in sperm and SR length despite their relative geographical proximity. Line crosses between the populations were used to model the additive, dominance, epistatic, maternal, cytoplasmic and X- and Y-chromosome genetic contributions. It should be realized that the LP inversion in the high line (Ruiz *et al*., 1990; Etges *et al*., 1999) will limit some recombination on the second chromosome.

2. Materials and methods

(i) *Culturing and crosses*

Laboratory cultures of the two populations were established from multi-female collections made in April of 1998 for the H line and in November of 1996 for the L line (generously provided by T. A. Markow). We maintained the lines in our laboratory for one year prior to analysis. All crosses were performed with flies reared at low density on standard banana medium in 200 ml bottles. The H and L lines were used to generate ten lines, four of which were non-segregating lines (parental H and L, and F_1 and F_{1r}) and six

https://doi.org/10.1017/S0016672303006190 Published online by Cambridge University Press

Table 1. *Source of genetic lines used in the generation means analysis*

Line	Designation	Source	
		Female	Male
High	H	H	H
Backcross high	BH	H	F_{1p}[a]
Backcross high reciprocal	BH_r	F_{1p}	H
First filial generation	F_1	H	L
First filial generation reciprocal	F_{1r}	L	H
Second filial generation	F_{2a}	F_1	F_1
Second filial generation reciprocal	F_{2b}	F_{1r}	F_{1r}
Backcross low reciprocal	BL_r	F_{1p}	L
Backcross low	BL	L	F_{1p}
Low	L	L	L

[a] F_{1p} is pool $F_1 + F_{1r}$.

of which were segregating lines (F_{2a}, F_{2b} and four backcrosses). Line designations are listed in Table 1. Breeding was scheduled so that all crosses were reared and examined contemporaneously, thus negating any temporal environmental influence. The means and variances of SR length and sperm length were used to analyse the quantitative genetics by generation means or line-cross analysis (Mather & Jinks, 1982; Lynch & Walsh, 1998). The methods for the means analysis are outlined below.

(ii) *Measurement of sperm and SR lengths*

The sperm length of each anaesthetized male was measured by dissecting the seminal vesicles into phosphate-buffered saline (PBS) on a subbed (gelatine coated) slide. After releasing a few hundred sperm into the saline, preparations were dried in a 60 °C oven, fixed in methanol : acetic acid (3 : 1) and then mounted with glycerol : PBS (9 : 1) under a glass coverslip. Digital images of sperm using dark-field optics at a magnification of 200 × were obtained using a Dage CCD72 camera mounted on an Olympus BX60 microscope. Sperm were measured to the nearest 10 μm using NIH Image public-domain software (http://rsb.info.nih.gov/nih-image).

For each female, following anaesthetization with ether, the reproductive tract was dissected into PBS on a microscope slide. A glass coverslip was placed on top with clay at the corners that allowed flattening of the SR to two dimensions without stretching the organ. The preparation was then viewed at 200 × using differential interference contrast microscopy. A digitized image of the SR was obtained and organ length determined by tracing its lumen using NIH Image.

(iii) *Means analysis*

Alternative genetic models for both sperm length and SR length were evaluated based on the ten line crosses. The initial models assessed discrete autosomal, maternal, cytological and X and Y genetic effects. The best-fitting models were used as the basis upon which systematically to add other genetic effects, which were then retained in future models only if they improved the overall fit of the model. In these crosses, a maternal cytoplasmic effect is not distinguishable from a maternal nuclear composite additive effect.

For all models, weighted least-squares procedures were used to estimate the parameters contained in vector **y** and their variances from the diagonal of their variance covariance matrix **S** (Mather & Jinks, 1982; Lynch & Walsh, 1998). The estimates of **y** and **S** are obtained as

$$\hat{\mathbf{y}} = (\mathbf{C}^{\mathrm{T}}\mathbf{V}^{-1}\mathbf{C})^{-1}\mathbf{C}^{\mathrm{T}}\mathbf{V}^{-1}\mathbf{x}$$

and

$$\hat{\mathbf{S}} = (\mathbf{C}^{\mathrm{T}}\mathbf{V}^{-1}\mathbf{C})^{-1},$$

where **C** is the coefficient matrix for the contribution of effects to each line mean, **V** is the diagonal matrix of the error variances of each line mean and **x** is the vector of observed line means. Goodness of fit of each model was tested using a χ^2, where $\chi^2 = \mathbf{x}^{\mathrm{T}}\mathbf{V}^{-1}\mathbf{x} - \mathbf{x}^{\mathrm{T}}\mathbf{V}^{-1}\mathbf{C}\hat{\mathbf{y}}$ (Hayman, 1958).

The degrees of freedom for this χ^2 is the number of line means minus the number of parameters in the model. The significance of model parameters was evaluated using *F* statistics by comparing improvement of the goodness of fit after modifying model parameters. Notice that *P* values increase as the fit of the model improves; models are considered significant at $P \geqslant 0{\cdot}05$.

3. Results

The means, variances and sample sizes for sperm length and SR length are listed for all line-crosses in Table 2. Because SR length was positively correlated with thorax length, analysis of covariance (ANCOVA) was used to statistically control for body size effects. There was no relationship between body size and sperm length.

(i) *Results of line cross for sperm length*

Table 3 shows the basic sperm length models (1–9) and additional models (10–20) involving both autosomal and sex-chromosome effects. When analysing all effects separately, the best-fitting model was an autosomal model including additive [d], dominance [h],

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Table 2. *Means, standard errors and sample sizes* (n) *for both sperm and SR length for all line crosses*

	Sperm			SR		
Line	Mean	SE	*n*	Mean	SE	*n*
H	1·847	0·0068	15	4·768	0·1011	15
BH	1·868	0·0082	30	4·591	0·0715	30
BH_r	1·875	0·0071	30	4·723	0·0714	30
F_1	1·822	0·0084	20	4·618	0·0879	20
F_{1r}	1·844	0·0059	20	4·572	0·0879	20
F_{2a}	1·833	0·0057	50	4·417	0·0553	50
F_{2b}	1·832	0·0086	49	4·508	0·0565	50
BL_r	1·812	0·0083	29	4·495	0·0722	30
BL	1·853	0·0089	30	4·250	0·0717	30
L	1·766	0·0126	15	4·101	0·1009	15

additive × additive [i] and dominance × dominance [l] epistasis. However, this model was not adequate ($\chi^2 = 16{\cdot}9$, $P < 0{\cdot}05$) to describe the means. The inclusion of additive × dominance [j] epistasis did not improve the model and was removed from further analysis.

Additional models for sperm length capturing X- and Y-chromosomal effects, mixed non-epistatic autosomal genetic activity in combination with X, Y or both X and Y chromosomal effects were tested (10–15) but none was adequate (Table 3). Models that included epistatic autosomal effects with sex chromosomes acting independently or epistatically with additive autosomal effects (Table 3, 16–20) showed that the best autosomal model (including d, h, i and l) was significantly improved by adding X-chromosomal effects [d_x] (model 16, $\chi^2 = 5{\cdot}8$, $P = 0{\cdot}21$) and by including X-additive × autosomal-additive interactions [i_{ax}] (model 17, $\chi^2 = 1{\cdot}9$, $P = 0{\cdot}59$).

Even though the autosomal–X model was adequate, we were interested in the possibility that the Y chromosome could be involved. Table 3 shows that models involving Y and autosomal–Y interactions (18, 19) are also adequate ($\chi^2 = 10{\cdot}3$, $P = 0{\cdot}04$ and $\chi^2 = 6{\cdot}7$, $P = 0{\cdot}08$) for describing the line means but not as good as the autosomal–X model. Model 20, which included both X and Y effects, did not show an improvement over the model showing only X effects (16).

Table 4 shows the relative contributions to the means of each effect for models 16–19. For each model, dominance [h] and dominance by dominance [l] epistasis were the largest contributors. Although additive and sex-chromosome effects were necessary for models to be acceptable, their relative contributions to sperm length were small.

(ii) *Results of line cross for SR length*

Table 5 shows the most relevant models tested for SR length. Of the basic effects tested separately, only the autosomal effects were adequate. The best fit was a simple model with additive [d] and dominance [h] effects (model 2, $\chi^2 = 10{\cdot}3$, $P = 0{\cdot}17$). Epistatic interactions did not improve the model. Maternal (4–6), cytoplasmic (7, 8), and X-chromosome effects (9–11) also did not significantly improve the overall model. The relative contribution of additive genetic variance to the mean of SR length was 0·31.

4. Discussion

Sperm length differences between two divergent populations of *D. mojavensis* were mainly the result of dominance and epistatic genetic interactions, with smaller contributions from additive genetic elements. Although they were mainly autosomal, either an X-chromosome or a Y-chromosome effect must be included in the analysis in order for models to be statistically adequate. These sex-linkage effects nonetheless offer a relatively minor contribution to the overall model (Table 4) and compose substantially less than might expected because the X-chromosome constitutes a large portion of the genome (20–40 %) in *Drosophila* (Turelli & Begun, 1997). As reported for *D. melanogaster* (Miller *et al*., 2001), SR length in *D. mojavensis* is mainly determined by large additive genetic effects, possibly with an autosomal dominance contribution and no apparent sex linkage. Although similar results for SR length were found for *D. melanogaster* and *D. mojavensis*, the current study is based on crosses between only two populations and thus the generality of our results should be considered with caution.

Previous examinations of sperm length have found extremely high heritabilities in the mouse *Mus musculus* ($0{\cdot}76 \pm 0{\cdot}02$; Woolley, 1971), the yellow dungfly *S. stercoraria* (0·67; Ward, 2000), the cricket *G. bimaculatus* ($1{\cdot}04 \pm 0{\cdot}06$; Morrow & Gage, 2001) and the dung beetle *Onthogophagus taurus* (between $0{\cdot}57 \pm 0{\cdot}31$ and $1{\cdot}14 \pm 0{\cdot}61$; Simmons & Kotiaho, 2002). All of these estimates are much greater than average estimates of heritability found for fitness-related traits ($0{\cdot}26 \pm 0{\cdot}02$; Mousseau & Roff, 1987), suggesting that sperm length is not subject to strong selection in these species. Simmons & Kotiano (2002) argue that sperm competition will have little impact on sperm length, and Morrow & Gage (2001) found that sperm length in *G. bimaculatus* had no impact on male fertilization success. As modelled by Parker (1998), sperm length is generally subject to stabilizing selection, whereas sperm numbers are the focus of directional sexual selection.

By contrast, the phylogenetic distribution of sperm lengths in *Drosophila* suggests rapid directional selection of this trait throughout the genus (Joly *et al*., 1991; Pitnick *et al*., 1995*a*). This conclusion is further supported by the observation of significant differences

https://doi.org/10.1017/S0016672303006190 Published online by Cambridge University Press

Table 3. *Sperm length models. It is not possible to include [h] and [i_{xy}] in the same model because they effectively estimate the same quantity for the given set of crosses*

	Autosomal						Maternal					Interactions					
Model	m	[d]	[h]	[i]	[j]	[l]	[d_m]	[h_m]	Cyto c	X [d_x]	Y [d_y]	[i_{xy}]	[i_{ax}]	[i_{ay}]	χ^2	d.f.	*P*
1	+	+	+												45·3	7	<0·01
2	+	+	+	+	+	+									16·9	4	<0·01
3	+	+	+	+		+									16·9	5	<0·01
4	+						+	+							90·3	7	<0·01
5	+								+						91·6	8	<0·01
6	+									+					90·4	8	<0·01
7	+										+				58·8	8	<0·01
8	+									+	+				53·2	7	<0·01
9	+									+	+	+			50·7	6	<0·01
10	+	+	+							+					37·0	6	<0·01
11	+	+	+							+			+		35·6	5	<0·01
12	+	+	+								+				40·0	6	<0·01
13	+	+	+								+			+	29·2	5	<0·01
14	+	+	+							+	+		+	+	26·8	3	<0·01
15	+	+								+	+	+	+	+	26·8	3	<0·01
16	+	+	+	+		+				+					5·8	4	0·21
17	+	+	+	+		+				+			+		1·9	3	0·59
18	+	+	+	+		+					+				10·3	4	0·04
19	+	+	+	+		+					+			+	6·7	3	0·08
20	+	+	+	+		+				+	+				5·8	3	0·12

Labels: m, means; [d], additive; [h], dominance; [i], additive × additive epistasis; [j], additive × dominance epistasis; [l], dominance × dominance epistasis; [d_m], maternal additive; [h_m], maternal dominance; c, cytological; [d_x], additive on X chromosome; [d_y], additive on Y chromosome; [i_{xy}], X-chromosome × Y-chromosome epistasis; [i_{ax}], autosome × X-chromosome epistasis; [i_{ay}], autosome × Y-chromosome epistasis.

Table 4. *Estimates for models 16–19 showing the relative contributions of the various factors and the standard errors for the contribution*

	16	17	18	19
m	1·730 ± 0·026*	1·730 ± 0·026*	1·746 ± 0·027*	1·743 ± 0·027*
[d]	0·054 ± 0·007*	0·055 ± 0·007*	0·032 ± 0·007*	0·032 ± 0·007*
[h]	0·298 ± 0·065*	0·310 ± 0·065*	0·271 ± 0·066*	0·282 ± 0·066*
[i]	0·073 ± 0·026*	0·045 ± 0·029	0·060 ± 0·026*	0·094 ± 0·032*
[l]	−0·200 ± 0·041*	−0·208 ± 0·041*	−0·184 ± 0·041*	−0·191 ± 0·041*
[d_x]	−0·014 ± 0·004*	−0·015 ± 0·004*		
[d_y]			0·009 ± 0·003*	0·009 ± 0·003*
[i_{ax}]		0·032 ± 0·016		
[i_{ay}]				−0·030 ± 0·016
χ^2	5·83	1·90	10·32	6·73
d.f.	4	3	4	3
P	0·21	0·59	0·04	0·08

* Significant contribution ($\alpha = 0·05$).

among geographic populations of *D. mojavensis* (Pitnick *et al.*, 2003). The relatively small additive contribution to sperm length reported here is consistent with a history of strong postcopulatory selection on this trait. In *Drosophila*, variation in sperm length probably contributes more to differential male fertilization success than does sperm numbers (Miller & Pitnick, 2002*b*).

The degree to which traits are sex linked might also be a function of the mode of selection. In a recent survey examining reciprocal crosses, Reinhold (1998) found that X-chromosome genes significantly influenced sexually selected traits, whereas non-sexually selected traits showed little or no X linkage. Reinhold (1999) further demonstrated that, if sex-limited traits are undergoing fluctuating selection, the heterogametic sex should possess traits that are predominantly influenced by X-chromosome genes. Fluctuating selection on sex-limited traits is considered to be widespread (Hamilton & Zuk, 1982;

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Table 5. *Models tested for SR length*

	Autosomal						Maternal		Cyto	X				
Model	m	[d]	[h]	[i]	[j]	[l]	[d_m]	[h_m]	c	[d_x]	[i_{ax}]	χ^2	d.f.	*P*
1	+	+										13·5	8	0·10
2	+	+	+									10·3	7	0·17
3	+	+	+	+	+	+						8·9	4	0·06
4	+	+					+					12·9	7	0·08
5	+	+					+	+				12·1	6	0·06
6	+	+	+				+	+				8·8	5	0·12
7	+	+							+			13·3	7	0·07
8	+	+	+						+			10·2	6	0·12
9	+	+								+		12·1	7	0·10
10	+	+	+							+		9·0	6	0·18
11	+	+								+	+	11·5	6	0·07

Labels: m, means; [d], additive; [h], dominance; [i], additive × additive epistasis; [j], additive × dominace epistasis; [l], dominance × dominance epistasis; [d_m], maternal additive; [h_m], maternal dominance; c, cytological; [d_x], additive on X chromosome; [d_y], additive on Y chromosome.

Reinhold, 2000) and might partly explain the large genetic variance found in many traits undergoing sexual selection (Pomiankowski & Møller, 1995). This process might also explain the X linkage, high heritabilities and presumably high additive genetic variance that has been found for sperm length in mice (Woolley, 1971; Wang *et al*., 2001), dungflies (Ward, 2000) and crickets (Morrow & Gage, 2001).

The low additive genetic variability and limited X-chromosome effect on sperm length observed here might be due to a lack of fluctuating selection on sperm length in *Drosophila*. In addition to rapid directional selection, the mechanics of sperm selection within the SR might also limit reverse selection for sperm length. In *D. melanogaster*, lines selected for increased SR length resulted in postcopulatory sexual selection for increased sperm length (Miller & Pitnick, 2002). By contrast, lines selected for reduced SR length resulted in no change of sperm length. Thus, in *Drosophila*, selection on sperm length by female 'sperm choice' (Birkhead, 1998, 2000; Pitnick & Brown, 2000) might only be capable of increasing sperm length.

In *Drosophila*, genes that cause hybrid sterility in the heterogametic sex map predominantly to the X chromosome (Coyne, 1992). For example, MacDonald & Goldstein (1999) noted that, in the backcrossed flies from a cross between *D. simulans* and *D. sechellia*, 29% or 59% (depending on the direction of the backcross) of flies used for measurement of sperm cyst length did not produce viable spermatozoa. Interspecific epistasis between autosomes and sex chromosomes can cause increased reproductive abnormalities (Orr, 1995) and substantially increase the likelihood of mapping cyst length to the X chromosome (Macdonald & Goldstein, 1999) or indicate an effect of the Y chromosome (Joly *et al*., 1997) on cyst length. Studies such as these provide valuable insights into the genetics of reproductive isolation (Wu *et al*., 1996) but might not reliably identify genes responsible for intra- or interspecific variation in characters. Genetic analysis of divergent populations within a species greatly reduces the likelihood of mapping traits to sex chromosomes as a result of aberrations arising from chromosomal incompatibility.

Several studies have indicated that genes found on the Y chromosome are linked to spermatogenesis (Roldan & Gomendio, 1999). In *Drosophila*, the Y chromosome has a substantial effect on male fertility (Charlesworth, 2001). Mutant genes that are advantageous to the heterogametic sex but disadvantageous to the homogametic sex are much more likely to spread in a population if they are located on the Y chromosome (Fisher, 1958; Rice, 1984). However, loss of function and eventual degeneration of genes on the Y chromosome is expected (Rice, 1994; Charlesworth, 1998). Although our results show the possibility of only minor Y-chromosome contributions to sperm length in *D. mojavensis*, relatively few genetic elements on the Y chromosome might nevertheless be instrumental to male fitness (Chippindale & Rice, 2001).

The current study serves as a preliminary report of the genetics of a model system for examining postcopulatory sexual selection and female choice. The results obtained here represent the first step in identifying the basic genetic properties of male and female traits exhibiting a pattern of coevolution throughout the genus *Drosophila*.

We thank J. Reagan for superb technical assistance and T. A. Markow for providing fly strains. Financial support for this work was provided by The National Science Foundation (grants DEB-9806649 and DEB-0075307 to S.P.).

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