Electronic Supplementary Material

METHODS

Isofemale lines and populations

Each isoline was founded by a single gravid female and maintained for at least 35 generations before experiments began.

Populations were maintained in 30 x 30 x 30 cm cages (Bioquip, Knutsford, UK) at densities of approximately 1000 individuals with overlapping generations. All flies were maintained on a cornmeal based medium (standard Jazz mix, Applied Scientific) and housed in incubators at 25°C on a 12:12h light:dark cycle unless otherwise specified.

Experimental animals

Experimental animals were collected as first instar larvae from Petri dishes containing 2% agar in equal parts apple juice and water placed overnight inside population cages. Larvae were collected at a density of 100 larvae per vial to limit larval competition and standardise rearing conditions. Virgin adults were separated by sex under light CO_2 anaesthesia at a density of 10 individuals per vial in order to standardise social experience and access to nutrition.

Flies were not exposed to CO_2 within 24 hours of any behavioural assays as recent exposure to CO_2 is known to alter mating behaviour [1]. Wing size was measured as a proxy for body size using ImageJ [2]. In all assays females were housed in experimental vials for 24 hours before experimentation to allow them to habituate.

DDT resistance assay

Assay vials also contained 4g of ‘Drosophila quick mix medium’ sprinkled with live baker’s yeast (Blades Biological, Edenbridge, UK) as this medium can be made in glass vials at room temperature.

qRT-PCR

Relative *Cyp6g1* expression was performed using the primers in ESM Table 1, and the thermal profile in ESM Table 2.

ESM Table 1. qRT-PCR primer sequences

Transcript	Direction	Sequence
Cyp6g1	F	ACCCTTATGCAGGAGATTG
Cyp6g1	R	TAGGCTGTTAGCACGAATG
Rpl32	F	TAAGCTGTCGCACAAATGG
Rpl32	R	GGGCATCAGATACTGTCCC

ESM Table 2. qRT-PCR thermal profile

Duration	Temperature (°C)	Cycle number
10:00	50	1
03:00	95	
00:20	95	40
00:20	60	
01:00	95	1
00:30	60	
00:30	95	

A mathematical model of intralocus sexual conflict

The model of Rostant *et al.* [3] is a single-locus, two allele, nonlinear recursive model that calculates allele frequencies in each subsequent generation by taking the proportions of males and females of each genotype in the current generation, and then calculating the frequencies of each mating combination (RR x RR, RR x RS/SR, RS/SR x SS, SS x SS, where R is the resistant allele, and S is the susceptible allele). Here follows a brief summary of the model, but full details are available in Rostant *et al.* [3].

In each generation the proportion of sires of each genotype, *y*, is given by equations 1-3, where *x* is the frequency of the genotype, and *m* is the relative mating success of resistant males.

$$y_{RR} = \frac{mx_{RR}}{m\,(x_{RR} + x_{RS}) + x_{ss}} \tag{1}$$

$$y_{RS} = \frac{mx_{RS}}{m\,(x_{RR} + x_{RS}) + x_{ss}} \tag{2}$$

$$y_{SS} = 1 - y_{RR} + y_{RS} \tag{3}$$

The relative mating frequencies of each genotype combination, *λ*, can then be calculated from the genotype frequencies, *x*, and the proportion of sires of each genotype, *y*, where *i* and *j* represent the male and female genotypes (equation 4).

$$\lambda_{ij} = x_i y_j \tag{4}$$

The relative numbers of each genotype in the next generation, *n*, can then be calculated using equations 5-7, where *F* is the relative fecundity of resistant females including offspring viability effects, and *P* is the relative pupal viability of resistant offspring.

$$n_{RR} = FP\lambda_{RRRR} + {}^1\!/_2\,\lambda_{RRRS} + {}^1\!/_2\,\lambda_{RSRR} + {}^1\!/_4\,\lambda_{RSRS} \tag{5}$$

$$n_{RS} = FP\lambda_{RRSS} + {}^1\!/_2\,\lambda_{RRRS} + {}^1\!/_2\,\lambda_{RSRR} + {}^1\!/_2\,\lambda_{RSRS} + {}^1\!/_2\,\lambda_{RSSS} + P\lambda_{SSRR} + {}^1\!/_2\,\lambda_{SSRS} \tag{6}$$

$$n_{SS} = F\,{}^1\!/_4\,\lambda_{RSRS} + {}^1\!/_2\,\lambda_{RSSS} + {}^1\!/_2\,\lambda_{SSRS} + \lambda_{SSSS} \tag{7}$$

Genotype frequencies for the next generation can then be calculated using equation 8.

$$x'_k = \frac{n_k}{\Sigma n} \tag{8}$$

RESULTS

ESM Table 3. GLMM summary table for female offspring production.

Effect	χ^2	**df**	***p***
Fixed			
Male Genotype	0.8829	6	0.3474
Female Size	7.9679	5	**0.004761**
Female Genotype	6.5046	5	**0.01076**
Random	**Variance**	**St Dev**	
Block	0.0276	0.1661	
Individual	0.5579	0.7469	

ESM Table 4. GLM summary table for female egg production.

Effect	***F***	**df1**	**df2**	***p***
Female Size	0.2245	1	87	0.6368
Male Genotype	0.5124	1	88	0.476
Female Genotype	10.882	1	89	**0.001397**
Male Genotype x Female Size	0.8489	1	83	0.4316
Female Genotype x Male Genotype	1.176	1	85	0.2812
Female Genotype x Female Size	1.3936	1	86	0.2411
Female Genotype x Male Genotype * Female Size	1.0566	1	83	0.3833

ESM Table 5. GLM summary table for egg-larvae viability.

Effect	***F***	**df1**	**df2**	***p***
Female Size	0.0189	1	86	0.8909
Male Genotype	2.8933	1	87	0.09252
Female Genotype	0.1626	1	88	0.6878
Male Genotype x Female Size	3.8595	1	85	0.05273
Female Genotype x Male Genotype	0.0397	1	84	0.8426
Female Genotype x Female Size	0.0007	1	83	0.9791
Female Genotype x Male Genotype x Female Size	1.0173	1	82	0.3161

ESM Table 6. GLM summary table for larvae-adult viability.

Effect	***F***	**df1**	**df2**	***p***
Female Size	0.21	1	85	0.6479
Male Genotype	0.0342	1	84	0.8537
Female Genotype	0.9052	1	86	0.3441
Male Genotype x Female Size	0.5821	1	82	0.4477
Female Genotype x Male Genotype	2.5587	1	83	0.1135
Female Genotype x Female Size	0.0101	1	81	0.9201
Female Genotype x Male Genotype x Female Size	0.9503	1	80	0.3326

ESM Table 7. GLM summary table for male sperm defence (P1).

Effect	*F*	df1	df2	*p*
Relative Copulation Duration	3.5418	1	66	0.06425
Offspring Before Remating	4.8984	1	68	**0.03024**
Genotype	0.0845	1	67	0.7722
Genotype x Offspring Before Remating	0.1663	1	63	0.6848
Genotype x Relative Copulation Duration	0.3594	1	64	0.5509
Offspring Before Remating x Relative Copulation Duration	0.408	1	65	0.5252
Genotype x Offspring Before Remating x Relative Copulation Duration	0.1512	1	62	0.6987

ESM Table 8. GLM summary table for male sperm offence (P2).

Effect	*F*	df1	df2	*p*
Relative Copulation Duration	0.7491	1	64	0.39
Offspring Before Remating	0.2158	1	62	0.6439
Genotype	0.1496	1	63	0.7002
Genotype x Offspring Before Remating	2.5281	1	61	0.117
Genotype x Relative Copulation Duration	0.7317	1	59	0.3958
Offspring Before Remating x Relative Copulation Duration	1.3318	1	59	0.2718
Genotype x Offspring Before Remating x Relative Copulation Duration	0.9852	1	58	0.325

ESM Table 9. GLM summary table for *Cyp6g1* expression.

Effect	*F*	df1	df2	*p*
Sex	229.48	1	9	**1.035E-07**
Genotype	186.99	1	9	**2.513E-07**
Sex x Genotype	1.4818	1	8	0.2582

References

1. Barron, A. B. 2000 Anaesthetising *Drosophila* for behavioural studies. *J. Insect Physiol.* **46**, 439–442. (doi:10.1016/S0022-1910(99)00129-8)
2. Schneider, C. a, Rasband, W. S. & Eliceiri, K. W. 2012 NIH Image to ImageJ: 25 years of image analysis. *Nat. Methods* **9**, 671–675. (doi:10.1038/nmeth.2089)
3. Rostant, W. G., Kay, C., Wedell, N. & Hosken, D. J. 2015 Sexual conflict maintains variation at an insecticide resistance locus. *BMC Biol.* **13**. (doi:10.1186/s12915-015-0143-3)