Sex-linked inheritance of endosulphan resistance in *Helicoverpa armigera*

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Endosulphan resistance is common in Australian populations of *Helicoverpa armigera* (Hübner), but resistance frequencies have not increased above 10–40% of the population, despite widespread use of this insecticide. We examined the inheritance of endosulphan resistance as part of a study of its evolution. Two field-resistant strains were selected in the laboratory. Results from a series of F₁s and backcrosses in both larvae and adults indicated that resistance was sex-linked, with almost complete dosage compensation in the hemizygous females. In males, resistance was partially dominant in fourth instar larvae but semi-recessive in adults. Estimates of resistance factors depended on the life stage and instar tested and on the bioassay method used. In larvae, resistance factors were highest in neonates (14-fold) and then declined to twofold in fifth instar larvae. Our results indicate that the genetic basis of resistance should enhance the rate of evolution in field populations compared with an autosomal locus.

Keywords: endosulphan resistance, *Helicoverpa armigera*, sex-linked resistance.

Introduction

Insecticide resistance provides a good model for the study of evolution in natural populations (McKenzie, 1996). This is because phenotypic differences are usually caused by major genes, and changes in allele frequencies are measurable over periods of 3–5 years. These changes arise from an interaction of the biological characteristics of the insect — those that determine effective population size and rates of gene flow — with characteristics of the resistance alleles, such as relative dominance, relative fitness or linkage relationships.

In insects, resistance alleles are rarely sex-linked (for examples, see Heather, 1986; McDonald & Schmidt, 1987; Argentine *et al.*, 1989; Brun *et al.*, 1995), so the consequences of this mode of inheritance on the evolution of resistance have not been well documented. However, from theoretical studies, Avery (1984) showed that directional selection for a sex-linked trait could lead to more rapid increases in allele frequency than if the allele was autosomal. This hypothesis has recently been supported by Follet *et al.* (1995), who modelled rates of adaptation to permethrin in field populations of Colorado potato beetle *Leptinotarsa decemlineata* (Say) for a

sex-linked trait. In contrast, McDonald & Schmidt (1987, 1990) proposed that sex-linked resistance in the horn fly *Haematobia irritans* (L.) could retard the rate of evolution compared with autosomally inherited traits.

Daly (1992) provided preliminary evidence that resistance to the cyclodiene insecticide, endosulphan, in *Helicoverpa armigera* (Hübner) is sex-linked. Detailed studies in this species, on both its ecology (Fitt, 1989) and the evolution of pyrethroid resistance (Daly, 1993; Forrester *et al.*, 1993), provide an opportunity to explore hypotheses about the importance of genetic factors on the evolution of endosulphan resistance in natural populations, because resistance to the two insecticides evolved concurrently during the 1980s in field populations of *H. armigera* in eastern Australia.

In response to the emergence of pyrethroid resistance throughout eastern Australia in 1983, the Australian Resistance Management Strategy for *Helicoverpa* control was introduced to delay the further development of resistance to both pyrethroids and endosulphan (Forrester *et al.*, 1993). The impact of the strategy on the subsequent evolution of pyrethroid resistance has been well described (Daly, 1993; Forrester *et al.*, 1993) but, to date, there is limited evidence of its impact on endosulphan resistance.

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Resistance to cyclodiene insecticides (endrin, endosulphan and toxaphene) in *H. armigera* was common and widespread in the early 1970s (Goodyer *et al.*, 1975). Resistance frequencies declined to almost undetectable levels in field populations during 1979–83 when endosulphan was replaced by pyrethroids, and other cyclodienes were deregistered (Forrester *et al.*, 1993; Gunning & Easton, 1994). Endosulphan use increased after 1984 in response to widespread pyrethroid resistance.

Despite the general association between the use of cyclodienes and the presence of resistance, the frequency of endosulphan resistance has not increased to high levels, in contrast to pyrethroid resistance (Daly, 1993; Forrester *et al.*, 1993). Until 1991, the frequency of resistant individuals fluctuated from 10% to 30% in populations sampled in the Namoi/Gwydir area of New South Wales, even though over 50% of pesticide applications for *Helicoverpa* control in cotton crops were with endosulphan. Frequencies have increased more recently, so that, by 1995, they reached 40% by late summer (Forrester & Bird, 1996). In contrast, pyrethroid resistance rose from maximum frequencies of around 15% in 1983 to 80–90% by 1995, even though pyrethroid applications account for less than 20% of insecticide applications in cotton crops (Forrester *et al.*, 1993; Gunning & Easton, 1994).

In addition to this year-to-year variation, frequencies of both endosulphan and pyrethroid resistance increase from spring to autumn, in association with the use of insecticides (Forrester & Bird, 1996), but they usually decline in the first spring generation after winter diapause, probably because of dilution from an influx of susceptible individuals into the cropping regions (Daly, 1993; Forrester *et al.*, 1993).

Forrester *et al.* (1993) proposed a number of hypotheses to account for the difference in evolutionary response between pyrethroid and endosulphan resistance. First, selection pressure could be lower for endosulphan resistance, because it is used predominantly in springtime when a sibling species, *H. punctigera* (Wallengren), is the dominant pest species, compared with the pyrethroids, which are used in mid-summer when *H. armigera* becomes more common. Secondly, endosulphan resistance may have reduced fitness in the absence of insecticide. Thirdly, the mode of inheritance and relative dominance of resistance alleles could differ between endosulphan and pyrethroid resistance. Another hypothesis could be that the discrimination in the field between genotypes may be less with endosulphan resistance than it is with pyrethroid resistance.

In this paper, we examine the third of these hypotheses — that the slower loss of susceptibility to endosulphan is a result of the mode of inheritance and expression of resistance. Daly (1992) reported preliminary evidence for the fourth hypothesis — that discrimination between genotypes under field selection is poor in larvae.

Materials and methods

Strains

Rearing techniques for *H. armigera* strains in the laboratory are given in Daly & Fisk (1992). Two endosulphan-resistant strains were derived from individuals collected by N. W. Forrester (New South Wales Department of Agriculture, Narrabri) as part of his resistance monitoring programme (Forrester *et al.*, 1993). Resist-1 was collected in midsummer and resist-2 was collected in the spring of the 1989/90 cotton season. The level of resistance in these strains was increased after a series of selections at 2, 20 and 40 µg per larva of endosulphan. The endosulphan-susceptible strain, suscept-1, was also susceptible to pyrethroids, whereas a second strain, suscept-2, was homozygous for metabolic resistance to pyrethroids (called susceptible and field, respectively, in Daly & Fisk, 1992).

F₁s and backcrosses were carried out in larvae using resist-1 and suscept-1 strains. All other experiments, including bioassays in adults and the Potter tower experiments, used the resist-2 and suscept-2 strains, because they were healthier.

The parent resist-2 line was selected for five generations at the LD₇₅, 80 µg per larva of endosulphan, using topical application on fourth instars. Survivors of the selecting dose were reared and mass-mated. An unselected line was kept as a control. Larvae from the selected strain were bioassayed each generation. Adults were bioassayed in the first and fifth generations.

Bioassays

We used three techniques to assay resistance in *H. armigera*.

1 Fourth instar larvae 28–40 mg in body mass were tested using a topical application of 1 µL of a solution of endosulphan (technical grade; Hoechst Australia Ltd, Sydney, Australia) dissolved in acetone (Daly & Fisk, 1992). Survivors were classified as resistant.

2 Larvae from hatchlings to 10-day-olds were exposed to cotton leaves, *Gossypium hirsutum*, culti-

var Deltapine 90, treated with a 0.5 mL solution of endosulphan applied using a Potter tower (Burkard Manufacturing, UK). All dilutions were a mixture of acetone, ethanol, white oil, 0.05% Triton X in water, in the ratio 1:1:2:6. The white oil and Triton X were needed to increase the solubility of endosulphan at high concentrations. Cotton leaves were sampled from the third to the fifth nodes. Treated leaf discs were allowed to dry, between one and five larvae were put onto the treated surface, and then the disc was placed over the top of a 30 mL plastic container, treated surface facing inwards, and the container was sealed with a snap lid. Mortality was recorded after 48 h and moribund larvae were counted as dead.

3 Adults were tested using the adult vial test, described in detail in Daly & Fisk (1993). A 0.5 mL solution of acetone with dissolved endosulphan was added to each vial. The concentration of endosulphan per vial was between 0.3 and 100 µg. Prepared vials were stored at 4°C for up to 14 days until required. Adults were scored after 2 days; individuals that were dead, moribund or showed uncoordinated movements were all counted as dead.

Analysis

All statistical analyses were performed using the statistical program GLIM 3.77 (Payne, 1985). A logit transformation was used for the analysis of all dose-response bioassay data.

Sex-linked resistance in *H. armigera*

In *H. armigera*, females are XY and males XX, as is the case in most Lepidoptera (White, 1973; Fisk, 1992). If a resistance locus is on the X chromosome,

the dose-response of the F₁ generation will differ between the reciprocal crosses, and male and female progeny will have different genotypes and phenotypes, as given in Table 1. In both crosses, sons would be heterozygous (X^RX^r), but daughters will be hemizygous resistant (X^RY) or susceptible (X^rY), depending on the father's genotype. However, as larvae are difficult to sex, it is not possible to determine directly the phenotype of heterozygous males in this life stage.

Backcrosses also yield different results depending on the direction of cross. The types of backcrosses that were informative and their predicted outcomes are given in Table 1. Data were not available for backcross D in the larval stage. Backcross lines were compared with the appropriate susceptible or F₁ line using GLIM log-linear regression.

Results

Fourth instar larvae with topical dose test

Bioassay results for fourth instar larvae are given in Table 2. Overall, resistance factors in the topical dose test in fourth instar larvae were calculated to be 42-fold and 62-fold in the two lines. There was no significant difference between the two susceptible lines ($F_{1,33} = 0.2$, $P > 0.05$), but significant differences were observed between the two resistant strains ($F_{1,26} = 6.3$, $P < 0.05$). The resist-2 strain, which had been subject to stronger selection in the laboratory, had a higher LD₅₀ and a steeper slope. However, there was no change in dose-response after five generations of selection: generation 1 (LD₅₀ = 45 µg per larva, slope 2.2) compared with generation 5 (LD₅₀ = 45 µg per larva, slope 2.0; $\chi^2_1 = 0.0$, $P > 0.05$).

The two reciprocal F₁ crosses were significantly different from each other ($F_{1,27} = 118$, $P < 0.001$).

Table 1 Predicted progeny in F₁ crosses and backcrosses for sex-linked endosulphan resistance in *Helicoverpa armigera* [females are the heterogametic sex (XY) and males are homogametic (XX)]

Cross	Genotype of progeny	Predicted phenotype	
		Larvae	Adults
F ₁			
A. (X ^r X ^r × X ^R Y)	X ^R X ^r , X ^r Y	Low F ₁ line	Heterozygous ♂, susceptible ♀
B. (X ^R X ^R × X ^r Y)	X ^R X ^r , X ^R Y	High F ₁ line	Heterozygous ♂, resistant ♀
Backcrosses			
A. X ^r X ^r × (X ^r X ^r × X ^R Y)	X ^r X ^r , X ^r Y	All susceptible	All susceptible
B. X ^r X ^r × (X ^R X ^R × X ^r Y)	X ^R X ^r , X ^r Y	Low F ₁ line	Heterozygous ♂, susceptible ♀
C. X ^R X ^R × (X ^r X ^r × X ^R Y)	X ^R X ^r , X ^R Y	High F ₁ line	Heterozygous ♂, resistant ♀
D. X ^R X ^R × (X ^R X ^R × X ^r Y)	X ^R X ^R , X ^R Y	All resistant	All resistant

Table 2 Dose-response of fourth instar larvae of *Helicoverpa armigera* to topical application of endosulphan (crosses are given in Table 1)

Strain	LD ₅₀ (μ g per larva)	95% CI	Slope	RF
Suscept-1	1.0	0.9, 1.1	2.3	
Suscept-2	0.9	0.8, 1.0	2.8	
Backcross A susceptibles	0.9	0.8, 1.0	2.5	
Resist-1	42	36, 48	1.5	× 42
Resist-2	54	49, 60	2.5	× 62
F ₁ A — low F ₁	3.9	3.1, 4.9	1.0	
Backcross B — low F ₁	1.8	1.5, 2.2	1.0	
F ₁ B — high F ₁	37	30, 46	1.0	
Backcross C — high F ₁	46	34, 62	1.3	

The low slope and the difference in LD₅₀ of the two crosses, 10-fold in this case, is consistent with sex-linked inheritance of resistance. The LD₅₀ from the F₁ B cross, 37 μ g per larvae, is similar to that in the parental resistance lines, 42–54 μ g per larvae, which is consistent with partial dominance of a sex-linked trait.

The backcross data also support the sex-linked inheritance of resistance in larvae (Table 2). Backcrosses A and C produced progeny with the predicted dose-response curve for sex-linked inheritance (Table 2) ($F_{1,29} = 0.5$, $P > 0.05$, susceptible parents cf. backcross A; $F_{1,17} = 2.4$, $P > 0.05$, high F₁ cf. backcross C). There were significant differences

between the 'low F₁' produced by the backcross B and the original low F₁ line ($F_{1,36} = 18$, $P < 0.01$), but the differences in LD₅₀ were small.

Dose-response in adults

Bioassay data for adults are given in Table 3. These data provide further evidence that the resistance is sex-linked. However, there appears to be extra variation from the susceptible strains that contributes to the expression of resistance in hybrid crosses.

Resistance factors in fully resistant adults were 22-fold in females and 30-fold in males. The differences between males and females were small and,

Table 3 Dose-response of adult *Helicoverpa armigera* (the crosses are as given in Table 1)

Strain	Sex	LD ₅₀ (p.p.m.)	95% CI	Slope	RF	
Susceptible	Male	8.8	8.1, 9.6	3.7		
	Female	9.2	8.5, 9.9	3.7		
	Combined	8.9	8.5, 9.5	3.7		
Resistant	Male	266	244, 289	1.8	30	
	Female	205	186, 224	1.8	22	
	Combined	250	226, 276	2.0	28	
F ₁	A	Susceptible daughters	13.8	13.1, 14.4	3.9	
		Heterozygous sons	66	61, 71	2.3	7.5
	B	Resistant daughters	310	270, 357	2.6	23
		Heterozygous sons	62	58, 68	2.3	7.0
Backcrosses						
A and B	Susceptible offspring	12.4	11.8, 13.0	4.1		
B and C	Heterozygous males	51	47, 55	4.6		
C and D	Resistant daughters	370	330, 415	3.5		
D	Resistant sons	497	410, 602	2.8		

although these differences were not statistically significant in either the susceptible ($F_{1,30} = 0.2$, $P > 0.05$) or the resistant strains ($F_{1,106} = 2.4$, $P > 0.05$) analysed, resistant males were consistently more tolerant of endosulphan than were females (see Table 3).

Data from single-pair crosses provide evidence for extra genetic variation that contributes to the resistant phenotype. Five single-pair crosses of type ($X^R X^R \times X^R Y$) yielded estimates of LD_{50} from 119 to 267 p.p.m. in females and from 185 to 444 p.p.m. in males. Three of these crosses were reared as isofemale lines over three generations. They maintained their relative differences during subsequent generations ($F_{5,34} = 3.6$, $P < 0.05$; for differences between lines over generations). For example, the LD_{50} s for isofemale line 1 were 156, 139 and 91 p.p.m. compared with 331, 406 and 425 p.p.m. for isofemale line 2. Nevertheless, after five generations of selection in the parental resist-2 strain, there was no increase in resistance levels. Indeed, there was a small but significant drop ($F_{1,15} = 17$, $P < 0.01$) in LD_{50} between generation 1 (399 p.p.m.) and generation 5 (280 p.p.m.). The unselected control strain had an LD_{50} after five generations of 183 p.p.m.

Results in the F_1 crosses indicate the importance of genetic background on phenotype. F_1 crosses yielded heterozygous sons (sevenfold) and either resistant (23-fold) or susceptible daughters. Although there was no significant difference in the LD_{50} between F_1 males from either direction of cross

(Table 3; $F_{1,50} = 0.1$, $P > 0.05$), the LD_{50} s of the susceptible and resistant F_1 females were about 1.5-fold greater than those observed in the parental strains for both susceptible ($F_{1,27} = 11$, $P < 0.01$, for differences in LD_{50}) and resistant strains ($F_{1,35} = 2.5$, $P > 0.05$). Thus, hybrid females were more tolerant of endosulphan compared with females in the original parent strains, although the difference was only significant for susceptible genotypes (see above). These differences were evident in the four estimates of LD_{50} s for single-pair crosses of F_1 resistant daughters ($X^R X^R \times X^r Y$) — they ranged from 512 to 817 p.p.m., all of which were greater than those obtained in mass matings above.

In the backcross generation, the progeny of each cross exhibited the predicted phenotype for sex-linked inheritance, given in Table 2, except that again the LD_{50} values of the backcross homozygous males and hemizygous females were 1.5- to twofold higher relative to the original parental values (Table 3).

Dose-response of first to fourth instars using Potter tower

The dose-responses of larvae exposed to treated leaves are given in Table 4. An age range of 0 to 10 days old corresponds to first to early fifth instar; and 7-day-old larvae are fourth instar, the same age as larvae treated with the topical dose test. The LC_{50} s increased within each strain as larvae

Table 4 Comparison of dose-response of susceptible and resistant *Helicoverpa armigera* larvae exposed to cotton leaves treated with endosulphan

Age of larvae (days)*	Strain	LC_{50} (p.p.m.)	95% CI	Slope	RF
0	Susceptible	39	32, 47	1.9	14
	Resistant	547	451, 664	1.9	
2	Susceptible	249	195, 1280	1.6	6.5
	Resistant	1620	1280, 2050	1.6	
3	Susceptible	499	425, 587	2.8	6.5
	Resistant	3220	2780, 3730	2.8	
5	Susceptible	941	771, 1150	2.3	5.2
	Resistant	4870	4020, 5900	2.3	
7	Susceptible	2340	2040, 2681	4.9	4.4
	Resistant	10 310	8990, 11 825	4.9	
10	Susceptible	5656	4663, 6862	2.9	2.1
	Resistant	11 871	10 013, 14074	2.9	

*Ages 0, 2, 3, 5, 7 and 10 days old correspond to first, early second, late second, third, fourth and fifth instars, respectively.

increased age: 146-fold within the susceptible strain, but only 22-fold within the resistant strain. Thus, the differences between resistant and susceptible insects at a given age were greatest in neonates (14-fold) and decreased to 2.1-fold by early fifth instar, 10-day-old larvae. Overall, resistance factors were lower than those observed with the topical dose test for fourth instar larvae (62-fold) compared with those observed with larvae on treated leaves (4.4-fold).

Neonate larvae died when exposed to endosulphan with only minimal or no signs of feeding. However, in larger larvae, particularly at 10 days old, significant amounts of feeding were observed even at concentrations of endosulphan that led to 95% mortality.

Sex-linkage and frequency of resistance

The frequencies of genotypes were calculated for a hypothetical resistance gene *R*: for either a sex-linked or an autosomal locus. It was assumed that (i) all genotypes were in Hardy-Weinberg equilibrium; (ii) the heterozygotes had intermediate expression; and (iii) full dosage compensation occurred at the sex-linked locus. The relative abundance of fully resistant individuals given the sex-linked locus ($X^R Y$ plus $X^R X^R$ individuals) was compared with the autosomal locus (RR individuals) at a range of allele frequencies from 10^{-5} to 10^2 . At 10^{-5} , there were 10 000 times more fully resistant individuals in the population if the allele was sex-linked rather than autosomal. The difference between sex-linked and autosomal loci disappeared rapidly as the allele became common. Note, however, that if all resistant genotypes ($X^R X^R$, $X^R Y$ and $X^R X^r$) are taken into account, then the relative abundance of resistant individuals is similar for both sex-linked and autosomal loci.

Discussion

Our results confirm that the major genetic factor for endosulphan resistance is sex-linked. The simplest hypothesis is that resistance is conferred by a single, major gene. At this stage, however, it is not possible to exclude a more complex mode of inheritance, namely that resistance is determined by a group of closely linked genes on the X chromosome. Furthermore, in the F_1 s and backcrosses, evidence of further genetic variation that contributes to the resistance phenotype was found. The nature of this variation is not defined, nor subject to further study at this time. Chromosomal mapping (see below) may help to

resolve the relative contribution of other genes to the expression of resistance.

Estimates of resistance factors conferred by the major gene varied depending on the life stage tested and on the technique used. Differences between susceptible and resistant insects were smaller when larvae were exposed to treated leaves compared with the topical dose test. The former bioassay technique resembles more closely exposure of insects under field conditions in which larvae move across and eat treated plants. Thus, our results in the laboratory indicate that discrimination between resistant and susceptible larvae under field conditions may be poor, except in neonate larvae.

Forrester *et al.* (1993) summarized earlier reports on endosulphan resistance in *H. armigera*. Resistance factors for the period 1971–81 varied from twofold to 62-fold. These estimates were obtained from topical bioassays using fourth instar larvae collected from the field as part of resistance monitoring programmes, so a major source of variation in their estimates would be the genetic heterogeneity within each sample. These earlier studies did not identify the genetic basis of resistance, so it is not clear if the same loci have been involved in endosulphan resistance since the early 1970s. However, the magnitude of resistance factors in earlier studies is similar to that reported here.

The mechanism of endosulphan resistance in *H. armigera* is unknown, but Forrester *et al.* (1993) observed high levels of cross-resistance to other cyclodienes, dieldrin and endrin. ffrench-Constant *et al.* (1993) reported that cyclodiene resistance in a variety of *Drosophila* species and in other insects (Thompson *et al.*, 1993) is associated with the *Rdl* gene that codes for the GABA_A receptor. They observed that resistant and susceptible individuals in these species all appeared to differ by only a single basepair at this locus.

In contrast, Trowell *et al.* (1994) and Daly & Trowell (1996) could find no difference in the *Rdl* gene in either susceptible or resistant *H. armigera* in the region described by ffrench-Constant *et al.* (1993). Furthermore, D. G. Heckel and L. J. Gahan (pers. comm.) have constructed linkage maps for two closely related heliothine species, *H. zea* and *Heliothis virescens*, and the *Rdl* gene is not sex-linked in these two species. This evidence taken together would suggest that the *Rdl* locus is not involved in sex-linked cyclodiene resistance in *H. armigera*.

Forrester *et al.* (1993) hypothesized that the mode of expression of endosulphan resistance might be one possible explanation for the gradual loss of susceptibility, compared with the situation with

pyrethroid resistance. This hypothesis has been proposed for the evolution of sex-linked permethrin resistance in the horn fly *Haematobia irritans* (L.). McDonald & Schmidt (1987, 1990) suggested that a combination of an almost completely recessive resistance locus linked to a sex-determining locus would delay the establishment of resistance in the horn fly because, initially, males could not be homozygous for resistance, and females would all be susceptible. Their conclusions, however, are dependent on the sex-determining system of some muscoid flies, such as the horn fly, in which sex is determined by a single locus rather than by the XX/XY or XX/XO chromosome complement that is more typical of insects.

Our results suggest that, in *H. armigera*, the sex-linked mode of endosulphan resistance should enhance, rather than retard, the rate of evolution for two reasons, as was proposed by Avery (1984). First, dosage compensation occurs in females, so that even at low frequencies of the X^R allele, fully resistant females ($X^R Y$) would be present, a situation not normally found with an autosomal locus until resistance reaches appreciable frequencies. Secondly, resistance is partially dominant in larvae, so that $X^R X^r$ males have a resistant phenotype.

Yet the field data indicate that the average frequency of resistance year by year has been slow to increase (Forrester & Bird, 1996), although there is some indirect evidence that selection is operating in field populations. Phenotype frequencies do change within a season in association with the use of endosulphan in spring and summer (Forrester & Bird, 1996). So, why has intraseasonal variation in endosulphan not led to a more rapid increase in resistance frequencies, as has been observed with pyrethroid resistance?

We hypothesize that other factors are also important. Daly (1992) reported that, in the field, poor discrimination between phenotypes occurred in larvae. Mortality of all phenotypes was high when very young larvae were exposed to cotton plants freshly exposed to commercial applications of endosulphan. Two days later, however, all phenotypes had high survival when exposed to the same plants. Thus, the window for selection appears to be small in field populations, smaller than that observed for pyrethroid resistance (Daly, 1993).

Monitoring data for resistance may also be giving a biased view of intraseasonal changes. Sampling is generally restricted to populations found in cotton fields where endosulphan is used (Forrester *et al.*, 1993). But *H. armigera* is polyphagous, and many alternative hosts, which are not sprayed with insecti-

cide, exist in spring and early summer (Fitt, 1989) when endosulphan usage is high. In midsummer, few alternative hosts exist, yet this coincides with the period of maximum pyrethroid usage. Thus, in these terms, although endosulphan resistance frequencies increase in response to selection in cotton fields, less of the total population in a region would be under selection compared with the situation with pyrethroid resistance. Thus, the observed increases in endosulphan resistance within a season may be overestimated relative to those observed for pyrethroid resistance.

In conclusion, although the genetic basis of resistance can influence the rate of evolution, the actual consequences of sex-linkage will depend on how sex is determined and on the relative dominance of the heterozygous and hemizygous individuals. Dominance relationships are not necessarily predictive of fitnesses of genotypes under selection in field populations (Daly, 1993; McKenzie, 1996).

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