

**BIOCHEMICAL GENETICS OF RESISTANCE AND
CROSS RESISTANCE OF *Spodoptera littoralis*
(Boisd.) PROGNEY TO CHLORPYRIFOS USING
DEVELOPED REARING TECHNIQUE**



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ABSTRACT

In laboratory tests, a unique technique was carried out to distinguish and obtain developed strains originated from separately reared *Spodoptera littoralis* (Boisd.) egg-masses (field and laboratory strains); traditional susceptible strain (Tss) from laboratory strain, and traditional resistant strain (Trs) from field. Two other strains were obtained in laboratory; the first developed traditional susceptible strain (dTss) was originated from the high susceptible larvae of Tss, and was subjected to relaxation for four successive generations. while the second, developed traditional resistant strain (dTrs) was resulted from moderately to high level larvae of Trs, and was subjected to selection pressure with chlorpyrifos as a selected agent for four successive generations, at the LC₅₀ level. The obtained results revealed that LC₅₀ value of Tss and dTss were 0.905 and 0.607, respectively. Also dTrs strain was exhibited 14.47 fold of resistance as compared to 8.45 fold for Trs strain.

Cross resistance spectrum was tested on dTss, Tss, uTss parent, Trs, and dTrs. Using 3 OP's compounds, one carbamate, 2 synthetic pyrethroids, and one demeloid.

All strains tested proved that increase in resistance level was accompanied by steady increase in cross resistance of all tested compounds. All strains tested were in between vigor tolerance to true resistance (positive cross resistance). Analysis of data showed that developed resistance strain exhibited a moderate level of resistance to the selected agent (chlorpyrifos).

Degree of dominance of resistance in F_{1a} (dTss males × dTrs females) and F_{2b} (dTrs males × dTss females) exhibited a moderate level of recessive. Reciprocal crosses in F₁ progeny indicated that increase in LC₅₀ and slope in F_{2b} (heterozygous) was done.

Enzyme activity of acid phosphatase (Acph) and alkaline phosphatase (Alkph) and transaminases enzyme were established for all strains. Generally, it was obvious that chlorpyrifos resistance in *S.littoralis* expressed as a high activity of enzyme was more exhibited in the developed and hybrid strains, while traditional strains recorded a fluctuated slightly levels.

Keywords: *Spodoptera littoralis*, Resistance, Susceptible, Enzyme

INTRODUCTION

Resistance is a simple mechanisms that can be attributed to a singular mode of action of a pesticide, which disrupts only one genetically controlled process in the populations appear suddenly, either by selection of resistant individuals in populations or by a mutation, which appear to be the less common of the two means. Furthermore, populations of several insect pests

now have such a high proportion of individuals resistant to all known insecticides, (Ware, 1982). Resistance is a genetic change in response to selection by toxicants that may impair control in the field (Sparks *et al.* 1989). As already emphasized, exaggerating the differences between resistant and susceptible individuals improves the resolution between genotypes, (Devonshire *et al.* 1986).

The frequencies of resistance alleles to each insecticide must be low. These individuals surviving on insecticide are likely to be killed by the other. This assumes that resistant genotype have statistically lower fitness than the susceptible one. Therefore, their frequency declines during those generations between application of the compound. (Georghiou *et al.* 1980). Selection for resistance can occur if a small population able to survive treatment with insecticides can reproduce and pass on their resistance to offspring (FAO, 2012) and Xin- Ju and Hui-Min 2012). There is a relationship between the increase of insecticide resistance and the activity of detoxification enzymes. (Rupa *et al.* 2010). The analysis of reciprocal genetic crosses between resistant *Helicoverpa armigera* (Hübner) and the susceptible strain showed a semi dominance resistant (Huiru Yan *et al.* 2012), Glutathion S transferases (GSTs) play an important role in protecting organisms against the toxicity of reactive oxygen species (ROS). Glutathion S transferase enzymes activity was increased in treated larvae, this enzyme plays a role in detoxification mechanism. Therefore this study aimed to investigate the biochemical genetics of resistance and cross resistance of *Spodoptera littoralis* (Boisd.) progeny to chlorpyrifos using developed rearing technique.

MATERIALS AND METHODS

Two strains were used in this study, parent (field) strain which was collected from Kaliobeya, Egypt, during two seasons of 2013, 2014 of cotton growing and laboratory strain, supplied by Central Laboratory of Pesticides (C.L.P.), Research Center of Agricultural, Giza, Egypt. Developed technique, based on some egg masses in a completely fitness in each shape and size were chosen by eye fitted from the production of each of two parent strains. Each egg mass was reared alone in a separate container (jar) labeled with a mean number. A portion of 50 larvae of 4th instar larvae from each separate colony were treated by LC₅₀ concentration with the selected agent (chlorpyrifos) at level of 50% mortality obtained from parent strain Ldp- line. All egg-masses which exhibited low mortality (20%) were collected together to become a base line of resistant strain which generated from field strain. On the contrary, for laboratory strain (80% death). The technique based on Mohanna (1998 a & b), that used to conduct a sufficient number of pairs (♂ and ♀) of fit moths from first generation after the first technique was carried out. Each pair (♂ and ♀) in a separate lantern glass was left to copulate, each of them were labeled for stress. The same technique was also carried out on a fit egg mass of strain which exhibited a moderate to high

level of susceptibility were taken to accomplish to the target of this investigation.

Strains:

- 1-Laboratory strain, traditional susceptible strain (Tss.) from Central Laboratory of Pesticides.
- 2- Untreated traditional susceptible strain (uTss.), of number 1 in relaxation for 4 generations.
- 3- Developed traditional susceptible strain (dTss.), resulted from laboratory strain after developing technique for four generations.
- 4- Parent strain (original), Kaliobeya field strain.
- 5- Traditional resistant strain (Trs.), parent after selection for 4 successive generations.
- 6- Developed traditional resistant strain dTrs, originated from parent strain after concentrating the resistant individuals and subjected to selection pressure for 4 successive generations.

Rearing Program:

Rearing program used to those mentioned by El- Defrawi *et al.* (1964). All strains were reared in a room conditions (25C° ± 5% and 65± 5% RH) with continuous cleanup in all steps.

Insecticides:

Three organophosphate compounds [profenfos, chlorpyrifos,(selected agent) and cyanofos], two synthetic pyrethroids namely, fenvalerate, and decamethrin, and a carbamate methomyl, besides to the demeloid diflubenzuron, were used in this study.

Toxicological STUDIES:

Treated strains of *Spodoptera littoralis* (Boid.), were subjected to chemical selections with chlorpyrifos for four successive generations. Selection was carried out on 4th instar larvae at a level of LC₅₀.

Cross RESISTANCE:

Cross resistance spectrum to different insecticides was determined at the end of selection in G4 selected generation, on all strains. Mortality data were corrected according to the formula of Abbot (1925). The LC₅₀ and slope values were estimated by method of Busvine (1957). The rate of development of resistance to chlorpyrifos together with cross resistance ratio were assessed and based on LC₅₀ of selected strain compared with their corresponding values of the susceptible strain, dTss.

Genetic ANALYSIS:

Larvae of dTss and dTrs strains were permitted to pupate, Pupae were differentiated, males and females were kept separately petri dishes placed in cages to prevent uncontrolled mating after emergence. Equal numbers of moths from dTss and dTrs (60 males and 60 females), were prepared. Reciprocal crosses were between [25 male from dTss and 25 female dTrs (F₁a)], besides to [25 males dTrs and 25 female of dTss. (F₁ b)]. A portion of each of two reciprocal F₁ hybrid was tested to determine LC₅₀ values. F₁ hybrid was pooled together and left to give F₂ progeny. LC₅₀ of F₂ was established . The degree of dominance (D) of resistance in F₁ offspring was calculated by Falconer formula (1964) according to Stone (1968).

$$D = \frac{2 \times LC_{50}(RS) - LC_{50}(RR) - LC_{50}(SS)}{LC_{50}(RR) - LC_{50}(SS)}$$

Where:

RR, RS, SS, represent resistant, heterozygote, (F_1), and susceptible population, respectively. The LC_{50} are expressed in terms of their logarithm. The values thus derived indicates whether resistance is fully recessive ($D = -1$), intermediate ($D = 0$) and fully dominant ($D = 1$). Other results in between the two extremes indicate partial or incomplete inheritance of character.

Enzyme Assays:

Insensitive enzyme activities were measured for each of six strains. Larvae of 4th instar (average weight 37- 40 mg.) were collected for each strain at G4. Larvae of all samples were starved for about 4 hrs. before being homogenized in distilled water (5 larvae /ml.). The homogenates were centrifuged for 15 min. at 10,000 r.p.m. at 2°C and supernatant fraction was used for enzyme assay. The method described by Pawel and Smith (1954), was used for determination of acid phosphatase, and alkaline phosphatase activity. The procedure was based on the hydrolysis of disodium phenyl phosphate (substrate) in acid media (pH 4.5) for Acp., or (pH 10.5) for alkaline media, to yield phenol. Hydrolyzed phenol after reacting with 4 amino antipyrine yield red color, which read spectrophotometrically.

Transaminases enzyme activities glutamic oxaloacetic transferase (GOT), and glutamic pyruvic transferase (GPT), were determined according to the method described by Reitman and Frankel (1957), using alpha - keto glutarate / dL- aspartate (2mM. + 200mM.) and alpha ketoglutarate / dL. Alanine (2 Mm. + 200 M.) as substrates for GOT and GPT, respectively. Enzyme activity was expressed as μ M pyruvate / min. / larvae. All colorimetric determinations were based on a minimum of four replications for each sample, and all homogenates were incubated with substrates at 37°C for half an hour.

Enzyme assays in hybrid strains, was done. In these experiments, 4th instar larvae average weight, (40-45 mg.), were collected from each of strains in last generation besides to the samples of F_{1a} , F_{1b} , F_{2a} , and F_{2b} of genetic analysis.

RESULTS AND DISCUSSION

The present work represents a trial to accelerate the susceptibility of any strain either low or high, depending on developed technique of Mohanna (1998 a & b). Susceptibility of 4th instar larvae of Tss. and dTss. Throughout four successive generations without any exposure to insecticide treatments are shown in Table (1). The base line strain Tss, LC_{50} was 0.905, while the susceptibility increased during four generations of relaxation to 0.607 in dTss. with 1.57 fold of susceptibility as compared to dTss. On the other hand, the rate of development of resistance to chlorpyrifos on parent (field), LC_{50} was 1.99, with 3.28 fold of susceptibility as compared to dTss. With continuous selection to field or parent, resistant ratios in moderate to tolerate led to 8.45

fold, LC₅₀ 5.13 at the end of selection. The strain reached to 14.47 fold, (LC₅₀, 8.79) when developed technique was carried out in it, after 4 successive generations with selected agent .Mohanna and El Sharkawy, (2015)

Table(1):Comparative studies of the development of resistance to chlorpyrifos in 4th instar larvae of *S. littoralis* in a traditional and developed technique forms.

Strain	Slope	LC ₅₀	R R
	5% fiducial limits	5% fiducial limits	
dTSS	2.209	0.607 (0.480- 0.771)	-----
uTss	2.157	0.856 (0.682-1.140)	1.41
Tss	2.446	0.905(0.718- 1.114)	1.57
parent	2.635	1.991(1.572-2.483)	3.28
Trs	2.301	5.128(3.710-6.520)	8.45
dTrs	2.257	8.786(5.868- 11.255)	14.47

Cross resistance pattern to several insecticides tested on dTss, uTss, Tss, parent, Trs, and dTrs, strains is presented in Table (2), The standard (developed) susceptible strain, dTss, was remarkably highly susceptible to all tested insecticides. Untreated traditional susceptible strain uTss, was exhibited slightly effect on *S. littoralis*, 4th instar larvae against all tested compounds. Poorly effects are also observed on traditional susceptible strain, when tested with the same compounds. Regarding to parent strain which exhibited a low level of tolerance to cyanofos, (RR= 2.49 fold), and chlorpyrifos (3.28 fold) followed by diflubenzuron with 4.17 fold. The carbamate methomyl and diflubenzuron recorded 6.28 and 4.17 fold of tolerance as compared to dTss. The profenfos came the next with 7.35 fold of vigor tolerance.

On the other hand, the pyrethroid fenvalerate was more resistant than the selective agent with 25.57 fold of resistance.

Traditional resistant strain Trs which was selected with chlorpyrifos for four successive generations and experimented to cross resistance agents were exhibited a slight level of tolerance to the cyanofos with 5.34 fold, followed by diflubenzuron, 7.69 fold, while methomyl recorded 9.34 fold of resistance at the end of selection in G4. Truly cross resistance was clear when used synthetic pyrethroids decamethrin which recorded 12.84 fold, while the poorly fenvalerate exhibited clear resistant ratio 71.43 fold as compared with dTss. The developed resistant strain dTrs for chlorpyrifos manifested a high level of resistance to each of fenvalerate 284.74 fold and diflubenzuron 28.19 fold followed by profenfos 27.21 fold, but decamethrin came the next with 26.05 fold of resistance. Positive resistance was recorded by cyanofos, 19.83 fold, followed by methomyl, which recorded 19.19 fold of resistance on chlorpyrifos selected strain (14.47 fold) at the end of selection in G4 as compared to dTss.

T2

Those findings agreed either totally or partially with results of Georghiou and Calmen (1969), and Cert & Georghiou(1974), indicating that resistance to organophosphate compounds and organochlorines, also possess high level of cross resistance to insect growth regulator. Also agree with findings of El-Sayed *et al.* (1982- 1983), indicating certain level of resistance established before the selected strain is ready to confer cross resistance to other insecticides and that the level of cross resistance increased with progressive selection. Mohanna and Hossain (1999)and Moshtaq *et al.* (2007), Generally level of resistance were very low to endosulfon, chlorpyrifos, phoxim, quinalphos, profenfos, bifenthrin, and thiocarb, and a moderate to high level to methomyl and cyfluthrin. Correlation analysis indicated that insecticides belonging to the same class such as in *S. littoralis*. Positive correlation was also found between endosulfon and carbamate, Adel and Freed (2014); and Ghasem *et al.*(2015).

Data of the degree of dominance for resistance are shown in Table(3). It is cleared that F_{1a} (dTss males \times dTrs females) and F_{1b} (dTrs males \times dTss females), exhibited a high recessive degree ($D = -0.79$) and ($D = -0.84$), respectively. Resistant ratio of F_{1a} and F_{1b} were 6.35 and 6.02 folds. Slightly decreases in slope values from 2.373 to 2.247 accompanied with decreasing LC_{50} value from 3.857 to 3.653, means that the strain being slightly heterozygous. On the other hand, F_{2b} was exhibited an increased in the slope of Ldp- line from 2.226 to 3.446 and decreased in LC_{50} from 5.4507 to 4.9407, that refers to more heterozygosity in the individuals of this generation. These data agree with Hoskins and Gordon (1956)and Rossi & Prescutti (1996),they mentioned that crosses between unselected and selected strains of *Piophilila cosei* (L.), were done. Results of survival analysis in F_1 hybrids were analogous for males and females and were similar in both reciprocal crosses. LC_{50} 's for deltamethrin were intermediate between those of the parental strains. Similarly, several authors arrived almost to the same findings; Head *et al.*(1995), Gunning *et al.* (1995); Argentine *et al.* (1995);Thomas and Boethal (1995); Rupa *et al.*(2010), the analysis of reciprocal genetic crosses between resistant *Helicoverpa armigera* strain (227.9 fold), and susceptible strain showed dominance ($D = 0.299 - 0.782$), suggesting CryI Ac resistance as a semi – dominance trait. Also agree with Muhammed *et al.* (2015).

Either acid phosphatase (Acp) or alkaline phosphatase (Alkp) activities were fluctuated between strains summarized in Table (4);, Regarding to Acp which inhibited by Tss and uTss, recording -30.12 and - 28.29 % activity as compared to dTss. Parent strain exhibited high level of activity (25.30 % , followed by Trs which was exhibited a moderate level of (79.51 %) activity. On the other hand, developed traditional resistant strain was the top of activities with 306.02 %, and F_{2a} hybrid strain came the next with 249.4 % activity as compared to the standard strain (dTss.).Strains which generated from hybrids recorded a moderately level of 13.73, and 116.87 % activity for dTss and dTrs respectively. Similarly results were obtained from Alkp., when experimented on dTrs, recorded the highest level of activity,(196.83 %), accompanied by F_{2a} hybrid strain with (188.47 %), and F_{2b} recorded

(184.44 %), but F₁b come in the latent effect of this group with (165.42 %). The second category of activity was F₁a,Trs, and parent strains; 150.72, 134 and 97.69 % activities. Low level of activity was recorded by uTss. 12.97 %. The monostatic which exhibited a moderate level of inhibition was Tss recorded -38.90 % activity for alkaline phosphate, as compared to dTss.. The present results are in agreement with those of, Van Asperen (1960) Abdallah *et al.* (1973),; Hassan (1979);Abd Elsamie *et al.* (1979));Gomaa *et al.* (1982),Abdel Hafez *et al.* (1993) Mohanna&Hossain(1999) and Muhammed *et al.* (2015).

Table (3): F₁ generation generated from reciprocal crosses between dTss and dTrs, strains, and F₂ derives from pooled F₁ hybrids using chlorpyrifos in 4th instar larvae of *S. littoralis*.

Hybrid strains	Slope	LC ₅₀ (5% fiducial limits)	Degree of dominance	RR
F ₁ a(dTss male × dTrs females)	2.373	3.8570 (3.0786- 4.8469)	0.79-	6.35
F ₁ b (dTrs male× dTss female)	2.247	3.6529 (2.4830- 4.5637)	0.84-	6.02
F ₂ a,(reciprocal crosses)	2.266	5.4507 (4.3675- 6.9666)	---	8.98
F ₂ b,(reciprocal crosses)	3.446	4.9407 (4.1327- 5.9163)	---	8.15

Table (4): Activity of Acp., Alkp., and (GOT&GPT) transaminases enzyme in 4th instar larvae of *S.littoralis* for different strains developed by certain technique , and having different level of susceptibility to chlorpyrifos , besides to strains resulting from hybrids.

Strain	Slope	LC ₅₀	Acid phosphatase		Alkalinephosphatase		GOT		GPT	
			Acp	%	alkp	%	GOT	%	GPT	%
dTss	2.22	0.607	1.66	-----	3.47	-----	10.73	-----	0.19	-----
uTss	231	0.056	1.18	28.29-	3.92	12.97	9.88	- 7.92	0.16	15.75-
tss	2.45	0.905	1.16	- 30.12	2.12	-38.90	10.97	2.24	0.13	-31.58
Parent	2.64	1.991	2.08	25.30	6.68	97.69	10.44	- 2.27	0.28	47.37
Trs	2.30	5.128	2.98	79.51	8.12	134	13.07	21.81	0.31	63.16
dTrs	2.26	8.786	6.74	306.02	10.30	196.83	13.91	29.64	0.36	89.47
F ₁ a	2.32	3.857	3.60	116.87	8.70	150.72	12.11	12.86	0.33	73.68
F ₁ b	2.25	3.653	3.88	133.73	9.21	165.42	13.06	21.71	0.44	131.58
F ₂ a	2.27	5.451	5.80	249.40	10.01	188.47	12.97	20.88	0.31	63.15
F ₂ b	3.45	4.978	6.12	268.67	9.87	184.44	14.36	33.83	0.37	94.74

Regarding, transaminases enzyme, data in Table (4) also proved that uTss, and parent strain inhibited GOT with – 7.92 and -2.27 % ,respectively. Whereas, uTss, and Tss strains were in the same behavior GPT with -15.75 and31.58 %. High low to low activity to GOT were recorded in Tss, Trs, F₁a and F₂a, ranged between 2.24 to 21.81 % activity .Moderate level of activity was recorded by F₂b with 33.83 % as compared to dTss. Glutamic pyruvic transferase inhibition was clear in uTss, and Tss, recording – 15.75 and -

31.58 % inhibition respectively. The hybrid F₁b was the highest activity , recorded 131.58 % activity, followed by F₂b with 94.74%, while dTrs came the next with 89.47 % activity. The third category which included F₁a, Trs, and F₂b recorded 73.68, 63.16, and 94.74 % activity. Parent strain was recorded 47.37 % activity as compared to dTss. In other word, increases of enzyme activities were clearly observed in strains developed by modified techniques and hybrids than strains developed by traditional once. Similar results with modified techniques are also recorded by Shaaban *et al.*(1978-1979)and Mohanna (1998 a&b), where they mentioned that Ch-E and acid phosphatase in *S. littoralis* seem to play a considerable role in resistance to OP's insecticides, Shoji and Hisaak (2005), glutathione s transferase , play an important role in protecting organisms against the toxicity of reactive oxygen species (ROS).Glutathion s transferase enzyme activity was increased in treated larvae, this enzyme play a role in detoxification mechanism in insects, therefore may be an over production of this enzymes occurred as a result of treated .Sarita *et al.* (2010), Wang *et al.* (2010) and Ghasm *et al.*(2015)who a reported that glutathione s transferase was a higher in treated larvae than in untreated one.

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وراثة بيو كيميائية حيوية لمقاومة نسل دودة ورق القطن لمبيد الكلوربيريفوس وعبور المقاومة باستخدام طرق تربية مطورة عبد الحميد مهنا وحزمه محمد الشرقاوي قسم الانتاج النباتي – كلية التكنولوجيا والتنمية – جامعة الزقازيق – مصر

تم تطوير المقاومة لسلالة حقلية وكذلك زيادة الحساسية لسلالة معملية بطريقة مطورة وذلك للتمييز والحصول على سلالات حديثة من اصل دودة ورق القطن والتي تم تربيتها منفردة من لطم من آباء للسلائتين الحقلية التقليدية (Trs) والمعملية الحساسة (Tss) عن طريق اختبارها بالجرعة المميثة لنصف عدد الأفراد المعاملة، والعكس صحيحا بالنسبة للسلالة المعملية للحصول على أفراد أكثر حساسية لمركب الانتخاب (الكلوربيريفوس) – جمعت الأفراد المقاومة وذات التحمل معا لتكون نواة للسلالة المقاومة (dTrs) ، والتي يتم تطويرها بالانتخاب بمركب الانتخاب- أما الأفراد الحساسة والقريبة منها من السلالة المعملية (dTss) جمعت مع بعضها أيضا ولكن دون تعرضها لأي مبيدات لمدة أربعة أجيال متعاقبة- وصل معدل المقاومة للسلالة المطورة إلى 14,47 ضعف مقارنة بالسلالة الحساسة المطورة.

شملت الدراسة أيضا ظاهرة عبور المقاومة باستخدام البروفينفوس والسيانوفوس (فوسفورية عضوية)، والفينفاليريت ، الديكاميثرين من مجموعة البيريثرويد المصنعة بالإضافة إلى الديميلويد دايفلوزينبيرون ، وأيضا الكربامات كانت ممثلة بالميثوميل- وقد أظهرت النتائج أنه كلما زادت درجة المقاومة تبعها زيادة في إيجابية عبور المقاومة لجميع المبيدات المستخدمة عن السلالة المقاومة والمطورة للكلوربيريفوس.

تضمنت الدراسة أيضا تقدير درجة السيادة والتحي لمبيد الانتخاب (الكلوربيريفوس) وتم عمل هجن بين ذكور حساسة وإناث مقاومة بين السلالة المقاومة المطورة ونظيرتها الحساسة (1) ، والعكس (2)- لإنتاج هجن الجيل الأول ، ثم ترك كلا منهما لإنتاج الجيل الثاني خلطيا للشق (1) وكذلك للشق (2) ، وتحليل النتائج المتحصل عليها كان واضحا أن للكلوربيريفوس درجة تنحي معقولة في كلا الحالتين. وقد تم إنهاء هذا العمل بدراسة النشاط الإنزيمي للسلائت المختبرة والمهجنة لإنزيمات الأسيد فوسفاتيز والألكالين فوسفاتيز ، كذلك الترانسامينيز (جلوتاميك أوكسالواسيتك ترانسفيريز والجلوتاميك بيروفيك ترانسفيريز) حيث أظهرت النتائج أن السلالات المقاومة كان لها نشاطا إنزيميا ، وبصفة عامة يفوق نشاط السلالات ذات التحمل الفائق والتحمل وكذلك الحساسة .

Table (2): Cross resistance studies on 4th instar larvae of *S.littoralis* for dTss, Tss, uTss, parent, Trs, and dTrs strains to chlorpyrifos with different insecticides.

Strain	dTss		Tss			uTss			Parent		Trs			dTrs			
Insecticide	Slope	LC ₅₀ (5% fiducial limits)	slope	LC ₅₀ (5% fiducial limits)	RR	slope	LC ₅₀ (5% fiducial limits)	RR	slope	LC ₅₀ (5% fiducial limits)	RR	Slope	LC ₅₀ (5% fiducial limits)	RR	Slope	LC ₅₀ (5% fiducial limits)	RR
Chlorpyrifos	2.24	0.6072 (0.480-0.771)	2.45	0.9054 (0.718-1.114)	1.49	2.16	0.8563 (0.684-1.141)	1.41	2.63	1.99110 (1.372-2.493)	3.28	2.30	5.2216 (3.710-6.519)	8.43	2.26	8.7858 (5.868-11.265)	14.47
Profenfos	2.29	0.7781 (0.614-0.970)	2.24	1.3113 (1.1008-1.737)	1.69	3.17	2.2034 (1.973-2.652)	2.83	2.58	5.7206 (4.567-7.445)	7.35	2.54	11.4412 (9.135-14.888)	14.20	2.95	21.1692 (17.313-24.255)	27.21
Cyanofos	1.97	3.0608 (2.0608-4.020)	2.22	5.0000 (3.812-6.551)	1.63	2.20	2.4601 (2.703-4.364)	1.13	2.98	7.6079 (5.194-9.233)	2.49	3.08	16.3476 (13.105-18.628)	5.34	3.48	60.6908 (50.986-70.001)	19.83
Methomyl	2.26	0.3964 (0.312-0.496)	2.99	0.5613 (0.452-0.683)	1.42	4.02	0.8612 (0.701-1.000)	2.17	3.08	2.4907 (2.034-3.016)	6.28	1.80	3.7030 (2.797-4.954)	9.34	2.98	7.6079 (5.940-9.233)	19.19
Diflubenzuron	2.24	2.500 (1.906-3.279)	2.52	2.7382 (2.178-3.533)	1.10	3.17	4.4048 (3.585-5.304)	1.80	2.90	10.4149 (8.424-12.991)	4.17	3.10	19.9098 (16.359-24.218)	7.69	3.17	70.4761 (57.355-84.866)	28.19
Fenvalerate	2.66	0.1458 (0.115-0.182)	2.63	0.4394 (0.359-0.538)	3.01	2.99	0.6363 (0.519-0.792)	4.36	2.87	3.7284 (3.088-4.522)	25.57	2.79	10.4149 (8.424-12.991)	71.43	2.95	41.5149 (33.902-51.198)	284.74
Decamethrin	2.80	0.6356 (0.513-0.790)	2.92	0.7859 (0.645-1.005)	1.24	2.75	1.3691 (1.086-1.767)	2.15	3.15	3.8986 (3.105-4.692)	6.13	2.04	8.1620 (6.174-14.070)	12.84	3.06	16.5561 (13.241-20.216)	26.05