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MONITORING OF CROSS RESISTANCE AMONG SEVERAL INSECTICIDES IN THE COTTON LEAFWORM, *Spodoptera littoralis* (BOISDUVAL)

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ABSTRACT

The pattern of cross resistance for 14 compounds representing the newest promising and main conventional groups of insecticides (spinosyn, spinosoid, avermectin, pyrethroids, carbamates, organophosphates, oxadiazines, nicotinoides, chitin synthesis inhibitors and chlorinated hydrocarbons) in spinosad and abamectin resistant selected strains were studied. Moreover, the cross-resistance of spinosad and abamectin insecticides in cypermethrin resistant strain and methomyl resistant strain were also studied. Spinosad dipping resistant strain (SDRS), showed clear cross resistance against spinetoram and abamectin where resistance factor (RF) values were 48.81 and 18.39, respectively**.** Negative cross-resistance was observed against seven of tested compounds i.e., fenvalerate, methomyl, chlorpyrifos-methyl, cyanophos, profenofos, indoxacarb and hexaflumuron. Values of RF for those compounds were 0.79, 0.80, 0.08, 0.98, 0.86, 0.47 and 0.76, respectively. Spinosad feeding resistant strain (SFRS) showed considerable cross-resistance against spinetoram, abamectin and profenofos where RF values were 74.90, 23.24 and 9.36, respectively. RF values for chlorpyrifos, hexaflumuron and endrin were around 2. Negative cross resistance was detected against the rest of tested compounds. Values of RF for only three insecticides (thiodicarb, methomyl and chlorpyrifos-methyl) revealed clear cross–resistance with abamectin dipping resistant strain (ADRS). The values of RF were 16.79, 14.90 and 10.04 fold, respectively. Cross resistance of the rest tested insecticides exhibited either slight cross-resistance levels or negative cross resistance. Using abamectin against adults from parent field strain (PS) revealed that no difference in susceptibility to abamectin between males and females. In cypermethrin dipping resistant strain (CDRS), spinosad had negative cross-resistance representing 0.019 fold as RF value. While it had low level of cross-resistance in methomyl dipping resistant strain (MDRS)(RF=3.44-fold).

INTRODUCTION

Knowledge of resistance type in certain resistant populations may offer valuable information to find new compounds to be used instead of those have lost their toxic effect against resistant populations. Some researchers have found cross resistance in several lepidoptrane species toward some insecticide classes (Mahmoud, 2005; Zhao *et al.,* 2002 and Miles and Lysandrou, 2002). In the present study, the cross resistance among fourteen conventional insecticides and spinosad, abamectin, cypermethrin and methomyl were investigated in four selected resistant strains of cotton leaf worm.

MATERIALS AND METHODS

1- Insecticides:

A- Bioinsecticides

a- Spinosyns

Spinosad (SC 24 %, Dow AgroSciences Co.)

b-Avermectins

Abamectin (EC 1.8 %, Roan Agrochemicals Co.)

B- Synthetic insecticides:

Cypermethrin (EC 20 %, Dow AgroSciences Co.)

(RS)-α-cyano-*3*-phenoxybenzyl(*1RS,3RS;1RS,3SR*)-*3*-(*2,2*-dichlorovinyl)-*2,2*-dimethyl cyclopropane carboxylate.

Methomyl (SP 90 %, DuPont Agricultural Co.)

S-methyl-*N*-(methyl carbamoyloxy)thioacetimidate

Chemicals used as surfactant

Triton X_{100} (100 % purity, BDH Chem, Ltd. Poole England)

2- Insects

- Parent field strain (PS)

The parent field strain of cotton leafworm, *S. littoralis* was brought as eggs and new hatches larvae from Alexandria university laboratory and kept away from insecticidal contamination in Plant Protection laboratory at Assiut University for two years to be stable. The strain was then divided into sub-strains to start the present study.

-Spinosad dipping resistant strain (SDRS)

This strain was obtained by selecting a part of the parent field strain with spinosad (SC, 24%) solution. Dipping of 4th instar larvae was followed for 25 generations.

-Spinosad feeding resistant strain (SFRS)

This strain was obtained by selecting a part of the parent field strain with spinosad (SC, 24%) solution. Feeding method to 4th instar larvae was followed for 23 generations.

-Abamectin dipping resistant strain (ADRS)

This strain was obtained by selecting a part of the parent field strain with abamectin (EC, 1.8 %) solution. Dipping of the 4th instar larvae was followed for 25 generations.

-Cypermethrin dipping resistant strain (CDRS)

This strain was obtained by selecting a part of the parent field strain with cypermethrin (EC, 20 %) solution. Dipping of the 4th instar larvae was followed for 32 generations.

-Methomyl dipping resistant strain (MDRS)

This strain was obtained by selecting a part of the parent field strain with methomyl (SP, 90 %) solution. Dipping of the 4th instar larvae was followed for 32 generations.

Field populations:

Five field populations collected from El-Behera, El-Minia, Assuit and El-Badary Egyptian cotton fields as egg masses and new hatched larvae during June and July 2005 season. The populations transferred to the laboratory and reared on fresh castor bean leaves to reach 4th instar larvae under optimum conditions (26± 2 C° and 65 ±5 % RH).

Selection methods

Whole- larval dipping method:

All insects used were maintained at 26± 2 C° and 65 ±5 % RH, under the normal daily light and dark. The larval dip technique (Babu and Santharam, 2002; Nayak and Chhibber, 2002 and Young *et al*., 2003) was carried out to build up the spinosad dipping resistant strain (SDRS), abamectin dipping resistant strain (ADRS), cypermethrin dipping resistant strain(CDRS) and methomyl dipping resistant strain (MDRS). Selection was carried out by using the 4th instar larvae (the mean weight of larvae = 40±5mg). At each generation, aqueous solution of the selected insecticide concentration which used in selection pressure plus 0.1 % triton x_{100} as a surfactant was prepared. This concentration equals the LC_{50} value of the previous selected generation. The larvae were dipped in the selection concentration for 5 seconds by metal net. The treated larvae were put in a large dry container that contained filter paper to dry the larvae. Then the dipped larvae were supplied with fresh castor leaves and put under the optimum conditions. After 24 hrs., dead larvae were separated and removed. However, the lived ones were distributed in clean jars (2 Kg), supplied with fresh untreated castor bean leaves and cared to get a new generation. Selection was carried out continuously through 25 generations for SDRS and ADRS. While For CDRS and MDRS, the selection was carried out for 32 generations.

Leaf dipping method

Leaf dip technique (Moulton *et al*., 2000 and Young *et al*., 2003) was used to build up the SFRS. Selection were carried out by the same technique mentioned above, except that the fresh castor leaves (instead of larvae) were dipped in the spinosad concentration for 5 seconds. Dipped leaves were put in a container with filter paper for 20-30 minutes to dry. After drying, the 4th instar larvae were supplied with the treated leaves for 24 hrs. The lived larvae were separated and cared, then supplied with fresh untreated castor bean leaves to get a new generation. Selection was carried out continuously for 23 generations. In both selection methods, about 15000-20000 larvae in each generation were selected.

Bioassay experiments

The same methods used in the selection pressure with some modification were used to determine the toxicity of insecticides.

Larval- dip bioassay:

Fourth instar larvae of *S. littoralis* at an average weight of 38-40 mg / larva were selected. Serial water aqueous solution of concentrations of the tested insecticide prepared+ triton x_{100}(0.1 %) were used for bioassay tests. Three replicates at least were used for each concentration using 10 larvae/ replicate. Larvae of each replicate were dipped in the tested concentration for 5 seconds and then transferred to Petri-dishes containing filter papers to dry. Same number of larvae for each replicate were similarly dipped in distilled water plus the surfactant as a control treatment.

The treated larvae were supplied with fresh castor leaves and incubated at 26± 2 temperature and 12:12 L:D and 65± 5 RH until recording the results. Mortality was counted 48 hrs after treatment. The larva was considered dead if no movement was detected when it was touched with a small brush. Results were corrected by Abbot's formula (Abbott, 1925) and LC_{50} and slope values were determined by a computerized probit analysis program. The toxicity of each insecticide was replicated 2 to 3 times.

Leaf -dip bioassay:

The same steps of the above mentioned bioassay except that the 4th instar larvae of CLW were fed on dried insecticide treated castor bean leaves for 24 hrs. The larvae were allowed to feed on untreated fresh castor bean leaves for another 24 hrs, then mortality was counted. Mortality percentages were corrected by Abbott's formula (Abbott, 1925) and LC_{50} and slope values were determined by a computerized probit analysis program. Each experiment was replicated 2 to 3 times.

RESULTS AND DISCUSSION

Cross-resistance in spinosad resistant CLW strains to various insecticides

Results in Table 1 show the cross resistance factors of fourteen tested insecticides representing different groups against spinosad dipping resistant strain (SDRS). Tested strain showed clear cross resistance against spinetoram and abamectin where resistance factor (RF) values were 48.81 and 18.39, respectively**.** Negative cross-resistance was observed against seven of tested compounds i.e., fenvalerate, methomyl, chlorpyrifos-methyl, cyanophos, profenofos, indoxacarb and hexaflumuron. Values of RF for those compounds were 0.79, 0.80, 0.08, 0.98, 0.86, 0.47 and 0.76, respectively. Low level of cross resistance was obtained against cypermethrin, chlorpyrifos, thiodicarb, imidacloprid and endrin whereas the RF values ranged from 1.19 to 4.29. The tested strain (SDRS) showed slope values ranged from 1.02 to 3.28 except for each of cyanophos and hexaflumuron where those were 0.58 and 0.47, respectively. Table 2 revealed that spinosad feeding resistant strain (SFRS) showed considerable cross-resistance against spinetoram, abamectin and profenofos where RF values were 74.90, 23.24 and 9.36, respectively. RF values for chlorpyrifos, hexaflumuron and endrin were around 2. Negative cross resistance was detected against the rest of tested compounds. Values of RF for cypermethrin, fenvalerate, chlorpyrifos-methyl, cyanophos, indoxacarb and imidacloprid were 0.02, 0.15, 0.72, 0.69, 0.16 and 0.47, respectively. Generally, slope values of LCp lines for all tested insecticides ranged between 1.26 and 3.43 except the chitin synthesis inhibitor, hexaflumuron (slope = 0.84). Comparing cross resistance between spinosad dipping resistant strain (Table 1) and spinosad feeding resistant strain (Table 2), it can be concluded that: First, regarding the only spinosoid insecticide tested spinetoram (mixture of two synthetic analogs of spinosyn J and spinosyn L), the result revealed that spinetoram exhibited the highest cross resistance in the two spinosad resistant strains (resistance factors were 48.81 and 74.90 fold in SDRS and SFRS, respectively) among all tested insecticides. However, the cross resistance values of spinetoram in the same strains were less than the resistance factor of the selected agent, spinosad (RF= 108 fold in SDRS and 86.85 fold in SFRS). The results suggest that the mechanism(s) of resistance to spinosad in the two spinosad resistant strains may be responsible to a great extent of the cross resistance to spinetoram. Second, interestingly, the cross resistance results of the second bioinsecticide abamectin in the two spinosad resistant strains showed that both strains had considerable high level of cross resistance (cross resistance factor =18.39 & 23.24 in SDRS and SFRS, respectively). Obtained results revealed that the two spinosad resistant strains of cotton leafworm had cross resistance to spinetoram and abamectin. So, it is worried about using these second generation of spinosyns (spinetoram) and abamectin, especially for controlling cotton leafworm spinosad resistant strains. Third, concerning the tested anticholinesterase insecticides, the carbamate (methomyl and thiodicarb) and organohosphate (chlorpyrifos, chlorpyrifos-methyl, cyanophos and profenophos) insecticides in the two spinosad resistant strains, the results showed that the OP profenofos was the only anticholinesterase insecticide which exhibited considerable cross resistance in SFRS (RF=9.36 fold), and negative cross resistance in SDRS (RF=0.86 fold). The rest of anticholinesterases tested (methomyl, thiodicarb, chlorpyrifos, chlorpyrifos-methyl and cyanophos) exhibited either low level or negative cross resistance in the two spinosad resistant strains (RF ranged from 0.08 to 4.50 fold). Fourth, out of the fourteen tested insecticides, eleven compounds representing two pyrethroids (cypermethrin and fenvalerate), two carbamates (methomyl and thiodicarb), three organophosphates (chlorpyrifos, chlorpyrifos-methyl and cyanophos), oxadiazine compound (indoxacarb), nicotinoide insecticide (imidacloprid), chitin synthesis inhibitor insecticide (hexaflumuron) and chlorinated hydrocarbons compound (endrin) had either low level of cross resistance or negative cross resistance in both spinosad resistant strains. The RF values of the same corresponding insecticides were 3.49, 0.79, 0.8, 1.32, 4.29, 0.08, 0.98, 0.47, 1.54, 0.76 and 1.19 in SDRS and were 0.02, 0.15, 3.90, 4.50, 1.33, 0.72, 0.69, 0.16, 0.47, 2.16 and 2.28 in SFRS, respectively. From the present results it might be recommend any of the eleven insecticides that showed low or negative cross resistance for controlling the two spinosad resistant strains of cotton leafworm. Kerns and Gaylor, 1992 suggested that the presence of variable resistance in pest field populations is beneficial to the grower because all populations remained susceptible to at least one insecticide. However, confirmatory bioassays are necessary to determine which insecticide bioassays are necessary to determine which insecticides will control a given pest population. Most of the present results were in agreement with certain published literatures (Wang *et al*., 2005; Zhao *et al,* 2002 and Mahmoud, 2005).

Cross-resistance in abamectin resistant CLW strain to various insecticides

Fourteen insecticides representing different groups which mentioned previously were tested against abamectin dipping resistant strain(ADRS)**.** Data in Table 2 show that values of RF for only three insecticides (thiodicarb, methomyl, chlorpyrifos-methyl) reveal clear cross–resistance with abamectin resistant strain. Their RR values were 16.79, 14.90 and 10.04 fold, respectively. On the other hand the cross resistance of the rest tested insecticides against the resistant strain exhibited slight cross-resistance levels and/or negative cross resistance. The RR values were 7.58, 6.80, 4.36, 3.47, 2.66, 1.05, 0.99, 0.39, 0.34, 0.16 and 0.11 for cypermethrin, endrin, chlorpyrifos, profenofos, spinosad, cyanophos, imidacloprid, spinetoram, fenvalerate, hexaflumuron and indoxacarb, respectively. The tested strain (ADRS) showed slope values ranged from 1.28 to 4.27 except for hexaflumuron (slope = 0.38). The study of cross resistance in abamectin resistant strain has been carried out by certain investigators (Wu *et al*., 2002; Zhang and He, 2001 and Rugg *et al.* ,1998). Comparing the cross resistance results in abamectin resistant strain and the two spinosad resistant strains in Tables 1&2, it can be concluded that: (1) The two spinosad resistant strains exhibited clear cross resistance against abamectin insecticide. The RF of abamectin in SDRS and SFRS were 18.39 and 23.24-fold, respectively. The opposite was not true, spinosad and spinetoram (mixtue of synthetic analogues of spinosyn J and spinosyn L) had slight tolerance or negative cross resistance with abamectin resistant strain. The RF values were 2.66 and 0.39 to spinosad and spinetoram, respectively. These results suggest that spinosad and spinetoram could be nominated as good insecticides to suppress any problem related to resistance of cotton leafworm toward abamectin but the abamectin could not used to solve a problem if cotton leafworm became resistant to spinosad. (2) It is considered good results that six from the fourteen tested insecticides had no cross resistance or negative cross resistance in abamectin resistant strain and the two spinosad resistant strains. Three insecticides were conventionals: The organophosphate chlorpyrifos and cyanophos; and fenvalerate from pyrethroid group. The other three insecticides were from new groups: indoxacarb (oxadiazines group) imidacloprid (nicotinoides group) and hexaflumuron (chitin synthesis inhibitors group). These results suggest that the mechanisms of resistance in the two spinosad resistant strains and abamectin resistant strain may have no effect on the toxicity of the above mentioned six insecticides.(3) Four tested conventional insecticides, chlorpyrifos cypermethrin, methomyl and thiodicarb exhibited negative or low cross resistance in the two spinosad resistant strains where RF values of the same corresponding insecticides were 0.7, 3.49, 0.8 and 1.1 fold, respectively, in SDRS and were 0.8, 0.02, 3.9 and 4.5 fold in SFRS, respectively. However, in ADRS, thiodicarb, methomyl and chlorpyrifos had clear cross resistance where RF values were 16.79, 14.90 and 4.36 fold, respectively. Cypermethrin exhibited slight cross resistance in ADRS (RF= 7.58 fold). Clear cross resistance to methomyl, thiodicarb and chlorpyrifos-methyl in ADRS and negative or low cross resistance of the same tested insecticides in the two spinosad resistant strains (SDRS&SFRS) were found in the present study. In addition, the two spinosad resistant strains exhibited clear cross resistance against abamectin insecticide, but the opposite was not true. Abamectin resistant strain had no cross resistance to spinosad. Finally, these results suggest that the mechanism(s) of resistance to spinosad in the two spinosad resistant strains of cotton leafworm seem to be differed than the mechanism(s) of resistance against abamectin in abamectin resistant strain of cotton leafworm.

Table (1).Toxicity and resistance factor of certain insecticides to spinosad dipping resistant strain (SDRS) of *S. littoralis* larval-dip method.

Insecticide	LC50 a	95 % Confidence limits Lower-Upper	Slope ± SE b	RF c
Spinetoram	7820.39	6516.99-9384.47	2.03±0.34	48.81
Abamectin	1552.80	1049.42-2242.06	1.90±.40	18.39
Cypermethrin	3.07	1.03-5.69	1.19±0.27	3.49
Fenvalerate	81.82	44.51-129.03	1.44±0.28	0.79
Methomyl	64.98	34.78-105.17	1.11±0.19	0.80
Thiodicarb	78.82	46.50-126.04	1.02±.18	1.32
Chlorpyrifos	21.87	16.23-35.71	2.08±0.49	4.29
Chlorpyrifosmethyl	1.04	0.63-1.92	1.43±0.24	0.08
Cyanophos	282.67	124.09-1286.85	0.58±0.16	0.98
Profenofos	5.24	2.37-11.79	1.95±0.23	0.86
Indoxacarb	0.64	0.40-1.00	1.33±0.25	0.47
Imidacloprid	7438.40	5789.08-9963.28	3.28±0.54	1.54
Hexaflumuron	562.17	129.99-1904.61	0.47±.16	0.76
Endrin	19.37	12.92-28.69	2.71±0.61	1.19

a, a.i. : active ingredient, μg ml-1
b, SE : standard error
c, RF : resistance factor =LC50 of the tested insecticide in resistant strain/ LC50 of the same
insecticide in parent field strain(table1)

Table (2).Toxicity and resistance factor of certain insecticides to spinosad feeding resistant strain (SFRS) of *S. littoralis* using leaf-dip techinque.

Insecticide	LC50 a	95 % Confidence limits Lower-Upper	Slope ± SE b	RF c
Spinetoram	9696.19	8080.16-11635.42	1.92±0.46	74.90
Abamectin	2873.44	174.05-9913959.26	1.75±0.28	23.24
Cypermethrin	0.35	0.11-0.70	1.26±0.21	0.02
Fenvalerate	27.03	15.04-41.18	1.38±0.23	0.15
Methomyl	275.89	84.69-436.07	1.48±0.41	3.90
Thiodicarb	135.53	42.72-371.02	3.36±0.45	4.50
Chlorpyrifos	18.37	15.15-22.39	3.43±0.50	1.33
Chlorpyrifosmethyl	146.02	30.64-331.40	2.08±0.32	0.72
Cyanophos	266.05	188.95-354.41	2.70±0.52	0.69
Profenofos	268.89	213.84-348.28	3.25±0.63	9.36
Indoxacarb	0.15	0.10-0.24	1.26±0.25	0.16
Imidacloprid	5178.27	3889.77-6746.54	1.82±0.30	0.47
Hexaflumuron	2658.54	1232.64-9322.29	0.84±.25	2.16
Endrin	35.01	27.58-45.92	3.18±0.48	2.28

a, a.i. : active ingredient, μg ml-1
b, SE : standard error
c, RF : resistance factor =LC50 of the tested insecticide in resistant strain/ LC50 of the same
insecticide in parent field strain(table 2)

Cross-resistance of CLW adults toward abamectin and spinosad in resistant strains

The toxicity (LD50 Values) for abamectin and spinosad insecticides against the resistant and parent adult strains in both sexes were determined by treating adults a topically ccording to the adult vial assay method (Plapp *et al*., 1987) with some modification. As exhibited in the present study, selected larvae showed resistance to the bioinsecticides abamectin and spinosad . It is very important to detect the resistance in adult stage.

Table (3):Toxicity and resistance factor of certain insecticides to abamectin dipping resistant strain (ADRS) of *S. littoralis* using larval-dip method.

Insecticide	LC50 a	95 % Confidence limits Lower-Upper	Slope ± SE b	RF c
Spinosad	431.00	90.95-771.50	1.28±0.35	2.66
Spinetoram	62.87	40.69-87.93	1.69±0.22	0.39
Cypermethrin	6.67	1.78-65.71	1.70±0.28	7.58
Fenvalerate	34.99	16.76-58.51	1.45±0.31	0.34
Methomyl	1208.74	1017.47-1438.76	4.19±0.62	14.90
Thiodicarb	999.69	454.38-1853.02	1.28±0.36	16.79
Chlorpyrifos	22.22	16.98-27.67	4.27±0.76	4.36
Chlorpyrifosmethyl	134.28	107.22-163.73	3.45±0.49	10.04
Cyanophos	304.57	193.40-478.80	1.51±0.27	1.05
Profenofos	21.22	3.70-83.81	1.77±0.28	3.47
Indoxacarb	0.15	0.07-0.22	1.53±0.36	0.11
Imidacloprid	4779.33	4004.23-5808.23	3.16±0.60	0.99
Hexaflumuron	118.67	0.23-4097.65	0.38±.19	0.16
Endrin	110.67	54.14-301.16	2.40±0.37	6.80

a, a.i. : active ingredient, µgml-1

b, SE : standard errorc, RF : resistance ratio= LC50 of the tested insecticides on the selected strain generation/ LC50

of the same insecticides on the parent field strain (table 1)

stronger in males than females and stronger in SDRS adults than in SFRS. The present study coincides strongly with certain studies (Roe *et al.*, 2000; Young *et al.,* 2003 and Brewer *et al.* 1990).

Regarding the abamectin insecticide, Table 4 show the LD50 values, slopes of LDp lines and resistance ratios of abamectin used against adults from parent field strain (PS) and from ADRS. Based on the LD50 values against the adults of the same strain, there were no differences in susceptibility to abamectin between males and females. When comparing the toxicity values for the same sex in the parent strain and abamectin resistant strain, the results revealed that slight differences were found in RR values in males and/or females toward abamectin, where RR was 1.61 fold for males compared with 1.28 fold for females against abamectin. The results suggest that the resistance gene(s) of abamectin insecticide in cotton leafworm were not able to express in adults and resistance could not be detected in abamectin adults. Gouamene-Lamine *et al*.(2003) reported that larval stages and adults of abamectin resistant strain(Ab-F) of Colorado potato beetle were significantly less sensitive to the toxic action of abamectin and for two tested analogues compared with susceptible strain (SS). This result indicated resistance of abamectin and two tested analogues expressed in the mature stage.

Table (4). Toxicity of abamectin to *S. littoralis* adults from abamectin dipping resistant strain(ADRS) and parent field strain(PS).

Insecticide	Treated adults form	Gender		
			Male	**Female**
Abamectin	**PS**	LD50 a 95% (C.L.) b	22.94 8.72-54.95	23.61 2.5-33.23
		Slope± SE c	3.55±1.96	1.97±0.93
	ADRS	LD50 a 95%(C.L.) b	36.88 3.85-61.74	37.33 18.55-58.47
		Slope± SE c	2.77±0.78	2.57±0.63
		RR d	1.61	1.28

a, a.i. : active ingredient, µg/g of adult body weight
b, C.L. : confidence limits
c, SE : standard error
d, RR : resistance ratio= LD50 of the resistant strain / LD50 of the parent field strain

Concerning spinosad insecticide, Table 5 shows the LD50 values, slopes of LDp lines concerning the adults of parent field strain (PS), SDRS and SFRS.

Table (5). Toxicity of spinosad to *S. littoralis* adults from parent field strain(PS), spinosad dipping resistant strain(SDRS) and spinosad feeding resistant strain(SFRS).

Insecticide	Treated adults form	Gender		
			Male	**Female**
Spinosad	**PS**	LD50 a 95% (C.L.) b	33.39 4.03-48.17	71.85 0.80-167.5
		Slope ± SEc	0.73±0.49	1.50±0.95
	SDRS	LD50 A 95% (C.L.) b	1212.66 106.42-1874.95	1297.66 509.35-2175
		Slope ± SEc	1.88±1.38	2.27±1.12
		RR d	36.31	18.06
	SFRS	LD50 A 95% (C.L.) b	298.17 122.39-6437.52	346.77 191.13-518.95
		Slope± SE c	1.87±0.82	2.14±0.71
		RR d	8.93	4.83

a, a.i. : active ingredient, µg/ g of adult weight
b, C.L. confidence limits
c, SE : standard error
d, RR : resistance ratio= LD50 of the resistant strain / LD50 of the parent field strain

The toxicity data in the three tested strains revealed that adult females exhibited higher LD50 values compared with males indicating that females were more tolerant than males against spinosad by 2.15, 1.07and 1.16 fold for parent field strain(PS), SDRS and SFRS, respectively. Concerning resistance ratios (Table 5), it is shown that the two sex adults in resistant strains exhibited considerable level of resistance, but adult males were able to build up resistance toward spinosad than the females in the two resistant strains. Adult males of SDRS and SFRS strains exhibited RR values of 36.31 and 8.93 fold, respectively, toward spinosad, while the same corresponding values for the females were only 18.06 and 4.34 fold, respectively. This means that RR in SFRS was half of RR values in SDRS. These results suggest that spinosad resistance gene(s) in the two selected strains of cotton leafworm larvae could express in adults, the expression might be

Cross resistance between cypermethrin and spinosad in cypermethrin dipping resistant strain (CDRS)

Data of cypermethrin resistant strain in Table 6 show that spinosad had negative cross-resistance with cypermethrin dipping resistant strain (CDRS) representing 0.019 fold as RF value. These results indicate that spinosad could be the effective insecticide in controlling cypermethrin resistant strains of cotton leafworm. The present results are supported by other studies. Miles and Lysandrou (2002) found that Lebanese field strain of CLW was 250-360 times less sensitive to cypermethrin compared with the susceptible strain. However, the same strain exhibited negative cross resistance to spinosad. They concluded that spinosad had potential as a resistance management tool due to its novel mode of action and negative cross-resistance with pyrethroids. Sayyed *et al.(*2005) found that the field populations of *P. xylostella* from Pakistan were highly resistant to deltamethrin (>500-fold) but had little or no resistance to spinosad. This confirmed that the mode of action of spinosad is unique. Data in Table 6 show that abamectin treated larvae had considerable tolerance in CDRS (RF=11.06-fold). The opposite was true, the RR of cypermethrin toward ADRS=8.67-fold (table 2).

Table (6). Toxicity and resistance factor of spinosad and abamectin to *S. Littoralis* 4th instar larvae from cypermethrin dipping resistant strain (CDRS) and methomyl dipping resistant strain(MDRS).

Treated larvae by dipping	Insecticide	LC50a	95 %Confidence limits LowerUpper	Slope ± SEb	RFc
Cypermethrin Dipping resistant strain	Spinosad	3.06	0.09-8.69	0.70±.22	0.019
	Abamectin	934.43	736.44-1188.49	3.18±0.60	11.06
Methomyl dipping Resistant strain	Spinosad	528.05	294.24-780.89	1.26±0.22	3.44
	Abamectin	642.74	543.70-759.87	2.93±0.45	7.61

a, a.i. : active ingredient, µg ml-1
b, SE : standard error
c, RF : resistance ratio= LC50 of the tested insecticides on the resistant strain of the same insecticides on the parent field strain.

Cross resistance between methomyl and spinosad in methomyl dipping resistant strain (MDRS)

The results in Table 6 showed that spinosad had low level of cross-resistance in methomyl resistant strain (MDRS)(RF=3.44-fold). The opposite was also true, methomyl showed negative cross resistance with SDRS (RF=

0.8-fold, Table 1). These results confirm that spinosad could be the effective insecticide in controlling methomyl resistant strains of cotton leafworm. The results in Table 6 revealed that abamectin had considerable cross resistance against methomyl resistant strain (RF=7.61-fold), the opposite was true; the RF value of methomyl in abamectin resistant strain was 14.90 fold. Wolfenbarger *et al*.(1997) found that field beet armyworm population had high resistance to methomyl after selection for seven generations in the laboratory. The resistant strain had high susceptibility toward emamectin benzoate (abamectin analogue).

Wu *et al*. (1998) reported that the chlorfluazuron resistant strain of diamondback moth (RR =23.78 fold) did not show corss resistance to cypermethrin, methomyl or abamectin. The above results suggested that there were cross resistance among abamectin, cypermethrin and methomyl insecticides in the cotton leafworm, so it is advised to avoid using abamectin insecticide as alternative pyrethroid and/or carbamate insecticide or avoid it to control pyrethroid and/or carbamte resistant strains.These results may suggest the cross role of detoxification enzymes in conferring resistance to abamectin, methomyl and cypermethrin but may be less in metabolizing spinosad insecticide.

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استقصاء المقاومة المشتركة فى دودة ورق القطن بين عدة مبيدات مختلفة

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تم دراسة المقاومة المشتركه لـ ١٤ مركب تتضمن مركبات جديده وأخرى تقليدية كمبيدات حشـرية (الاسبينوسـين ، الاسبينوسـويد ، الأفـرمكتين ، البيروثرويـدات ، الكربامـات ،المركبـات الفوسـفورية العضـوية ، الاوكسـادايزين ، نيكوتينويـدز ، مضـادات تكـوين الكيتـين ، المركبـات الكلورينيـة العضـوية) وذلك فى سلالة مقاومة للاسبينوساد واخرى مقاومه للابـامكتين . و قد تـم دراسـة المقاومـه المشتركه أيضـا للاسبينوساد و الأبـامكين فـى سـلالة مقاومـة للسيبرمثرين وأخـرى للميثوميل . وقد أظهرت السلالة المقاومة للاسبينوساد بـالغمر (SDRS) مقاومـة مشتركة واضـحة ضـد الاسـبينتورام والأبـامكتين حيـث كـان معامـل المقاومـة (R F) = ٤٨,٨١ ، ١٨,٣٩ علـى التـوالى . وقـد وجـدت مقاومــة مشـتركة سـلبية تجـاه سـبعه مــن المركبــات المختبــره وهــى : الفينفاليرات،الميثوميـــل،الكلوربيريفوس ميثيـــل، ســـيانوفوس، بروفينوفـــوس، اندوكســـاكارب ، هكسافلوميرون حيث كانت قيم الـ R F لهذه المركبات ٠,٧٩ ، ٠,٨٠ ، ٠,٠٨ ، ٠,٩٨ ، ٠,٨٦ ، ٠,٤٧ ، ٠,٧٦ علـى التـوالى . وقـد أظهـرت السـلاله المقاومـة للاسبينوسـاد بالتغذيـة (SFRS) مقاومة مشتركة الى حد ما ضد الاسبينتورام ، الابامكتين ، البروفينوفوس حيث كانت قيم الـ RF هى : ٧٤,٩٠ ، ٢٣,٢٤ ، ٩,٣٦ على التوالى . وكانت قيم الـ RF للكلوربيريفوس ، هكسـافلوميرون ، الاندرين قريباً من ٢ . وقد ظهرت مقاومة مشتركه سلبيه ايضاً ضد باقى المركبات المختبره . وفـى السلاله المقاومـة للأبـامكتين بـالغمر (A D R S) أوضـحت قيم الـ R F لثلاثة مركبات وهـى ثيوديكـارب ، ميثوميـل ، كلوربيريفـوس ميثيـل مقاومـة مشـتركة وكانـت ال RF مقدارها ١٦,٧٩ ، ١٤,٩٠ ، ١٠,٠٤ على التوالى . والمقاومة المشتركة لباقى المركبات المختبرة أظهرت اما مقاومـة مشتركه طفيفه أو مقاومة مشتركه سلبيه . وأظهـر أسـتخدام الأبـامكين ضـد الحشـرات الكاملـه مـن سلالة الأباء (P S) أنه لا توجد أختلافات فى الحساسية تجاه الأبامكتين بين الذكور والأناث . وفـى السلاله المقاومه للسيبرمثرين (CDRS) أظهر الأسبينوساد مقاومة مشتركه سلبية حيث كان الـ R F = ٠,٠١٩ بينما كانت قيمه الـ RF = ٣,٤٤ فى السلالة المقاومة للميثوميل (MDRS) .