

TOXICITY OF SPINOSAD AND ABAMECTIN COMPARED WITH SOME CONVENTIONAL INSECTICIDES AGAINST PARENT FIELD STRAIN OF COTTON LEAF WORM, *Spodoptera littoralis* (BOISD.)

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ABSTRACT

Fifteen insecticides from different chemical groups were tested against the 4th instar larvae of parent field strain of cotton leaf worm *Spodoptera littoralis* (Boisd.). Two bioassay methods were followed to determine the toxicity of all tested compounds, larval-dip and leaf-dip methods. In the larval-dip bioassay, the most effective insecticide was cypermethrin (LC50 = 0.88 ug /ml), while imidacloprid showed the lowest toxic effect (LC50 = 4838 ug/ml). The toxicity order of the tested compounds using larval-dip method was as follow: cypermethrin > indoxacarb > chlorpyrifos > profenofos > chlorpyrifos-methyl > endrin > thiodicarb > methomyl > abamectin > fenvalerate > spinetoram > spinosad > cyanophos > hexaflumuron > imidacloprid. In the leaf-dip bioassay, the indoxacarb was the most toxic compound (LC50 = 0.96 ug/ml), while imidacloprid was the least toxic one (LC50 = 10912 ug/ml). In this method, indoxacarb was more toxic than chlorpyrifos, endrin, cypermethrin, profenofos, thiodicarb, methomyl, spinosad, abamectin, spinetoram, fenvalerate, chlorpyrifos-methyl, cyanophos, hexaflumuron and imidacloprid by 14.35, 16.0, 22.78, 29.94, 31.36, 73.73, 106.11, 128.8, 134.84, 193.16, 211.91, 400.89, 1283, and 11367 fold, respectively. Slope values in both bioassay methods indicated that the insect population was relatively heterogenous in their susceptibility toward the tested insecticides. Toxicity of all tested insecticides was compared for the two methods of application.

INTRODUCTION

Insecticide resistance has been reported all over the world for almost of the insecticides used against insect pests (McManus *et al.*, 1994; Leplé *et al.*, 1995; Duan *et al.*, 1996; Xu *et al.*, 1996; Gatehouse *et al.*, 1997; Yeh *et al.*, 1997). Development of resistance in cotton leafworm to all categories of synthetic insecticides has been also recorded by many investigators such as Georghiou and Lagunes-Tejeda, 1991; Smaffe and Degheele, 1997; Muller-Cohn *et al.*, 1996 and El-Ghareeb, 1994. In Egypt, the cotton leafworm (CLW), *Spodoptera littoralis* (Boisduval) is a key polyphagous cotton pest. Its larvae feed not only on cotton but also attack more than 29 hosts from other crops and vegetables, and more than 60 different cultivated and wild plants (Gordon, 1961). The rate of CLW infestation can reach up to 50,000 egg-masses/acres, causing severe damage to leaves, buds, flowers and bolls (Metcalf, 1994). Farmers often use large quantities of insecticides and spray diversity of chemicals to control this insect. In addition to the life cycle of this insect without hibernation period, it has destructive feeding habits and its demonstrated ability to develop resistance to chemical insecticides. One of recommended strategies to manage resistance problem is using insecticides

with novel modes of action. In recent years, several natural plant products have been considered potential alternatives to conventional insecticides as natural means of pest control (Rice and Coats, 1994). Spinosad is a one of the new and highly promising insecticides. It is a microbial origin, macrocyclic lacton glycoside, derived from actinomycete bacterium species, *Saccharopolyspora spinosa* Mertz&Yao. This research was carried out for Studying the relative toxicity of spinosad and abamectin compared with some conventional insecticides on the 4th instar larvae of parent field strain of cotton leafworm using two bioassay techniques.

MATERIALS AND METHODS

Insecticides:

A- Bioinsecticides

a- Spinosyns

- Spinosad (SC 24 %, Dow AgroSciences Co.)

b-Avermectins

- Abamectin (EC 1.8 %, Roan Agrochemicals Co.)

B- Synthetic Insecticides:

a-Spinosoids

Spinetoram (SC 12%, Dow AgroSciences Co.)

b-Pyrethroids

- Cypermethrin (EC 20 %, Dow AgroSciences Co.)
- Fenvalerate (TG, > 90 % purity, Sumitomo Chem. Co.)

c-Carbamates

- Methomyl (SP 90 %, DuPont Agricultural Co.)
S-methyl-N-(methyl carbamoyloxy)thioacetimidate
- Thiodicarb (SC 50 %, Bayer Crop Science Co.)
3,7,9,13-tetramethyl-5,11-dioxa-2,8,14-trithia-4,7,9,12-tetra-azapentadeca-3,12-diene-6,10-dione

d- Organophosphates

- Chlorpyrifos (EC 48 % , Dow AgroSciences Co.)
O,O-diethyl O-3,5,6-trichloro-2-pyridyl phosphorothioate
- Chlorpyrifos-methyl (EC 50 %, Dow AgroSciences Co.)
O,O-dimethyl O-3,5,6-trichloro-2-pyridyl phosphorothioate
- Cyanophos (EC 50 % , Bayer Crop Science Co.)
O-4-cyanophenyl O,O-dimethyl phosphorothioate

- Profenofos (EC 72 % , Ciba-Geigy Co.)
O-4-bromo-2-chlorophenyl O-ethyl S-propyl phosphorothioate

e-Oxadiazines

- Indoxacarb (SC 15 % , DuPont Agricultural Co.) Methyl(S)-N-[7-chloro-2,3,4a,5-tetrahydro-4a-(methoxycarbonyl)indeno[1,2-e]-[1,3,4]oxadiazin-2-ylcarbonyl]4-(trifluoro-methoxy)carbonilate.

f- Nicotiniodes

- Imidacloprid (FL 24 % , Bayer Crop Science Inc.)
1-(6-chloro-3-pyridylmethyl)-N-nitroimidazolidin-2-ylideneamine

g-Chitin synthesis inhibitors

- Hexaflumuron (EC 5% , Dow AgroSciences Co.)
-[3,5-dichloro-4-(1,1,2,2-tetrafluoroethoxy)phenyl]-3-(2,6-difluorobenzoyl)urea

h-Chlorinated hydrocarbons

- Endrin (TG, > 90.0 % purity, Shell International Chem. Co.)
1,2,3,4,10,10-hexachloro-1R,4S,4aS,5S,6,7R,8R,8aR-octahydro-6,7-epoxy-1,4:5,8-dimethanonaphthalene
fenvaterate and endrin are repeatedly crystallized from a solvent system of acetone : n hexane).

. 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 concentration of the tested insecticide prepared+ triton \times_{100} (0.1 %) were used for bioassay tests. Three or four replicates were used to each concentration using 10 larvae per each replicate. The ten larvae were dipped in the tested solution for 5 seconds and then transferred to Petri-dishes containing filter papers to dry. Larvae of the same number and replicates were similarly dipped in solution of distilled water plus surfactant as a control treatment. Then the treated larvae were supplied with fresh castor leaves and incubated at 26 ± 2 temperature, 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 (Abbot, 1925); 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 were followed except that the 4th instar larvae of CLW were fed on dried insecticide treated castor bean leaves for 24 hrs. Then the larvae were allowed to feed on untreated fresh castor bean leaves for another 24 hrs, and the mortality was counted. Mortality percentages were corrected by Abbot's formula (Abbot, 1925); LC_{50} and slope values were determined by a computerized probit analysis program. Each experiment was replicated twice at least.

RESULTS AND DISCUSSION

A. Larval-dip bioassay

Table 1 show the LC_{50} values, lower and upper values of confidence limits and slope values of LC_p lines for the fifteen tested compounds against the 4th instar larvae of parent field strain of the cotton leafworm (CLW), *Spodoptera littoralis* (Boisduval) using larval-dip bioassay.

The data cleared that the most effective insecticide was cypermethrin ($LC_{50} = 0.88 \mu\text{g ml}^{-1}$), while imidacloprid showed the lowest toxic effect ($LC_{50} = 4838.42 \mu\text{g ml}^{-1}$). So cypermethrin was more potent than imidacloprid

by 5498 fold. The toxicity order of all tested compounds was as follow: cypermethrin > indoxacarb > chlorpyrifos > profenofos > chlorpyrifos-methyl > endrin > thiodicarb > methomyl > abamectin > fenvalerate > spinetoram > spinosad > cyanophos > hexaflumuron > imidacloprid. The slope values of the regression lines (Table 1) were fluctuated around 1.0 for almost tested insecticides. The least slope was (0.76) with chlorpyrifos-methyl and the highest one was (4.60) with fenvalerate. This indicates that the insect population was relatively heterogenous in their susceptibility toward tested insecticides by larval dip method.

Table 1. Toxicity of the tested insecticides to 4th instar larvae of the parent field strain (PS) of cotton leafworm, *S. littoralis* using larval-dip method.

S.	Insecticide	LC50 a	95 % Confidence limits		Slope ± SE b
			Lower	Upper	
1	spinosad	162.03	39.99	275.29	1.42±0.49
2	spinetoram	160.20	68.47	303.52	1.38±0.28
3	abamectin	84.46	34.15	203.67	1.25±0.38
4	cypermethrin	0.88	0.46	2.21	1.07±0.28
5	fenvalerate	103.75	87.62	123.24	4.60±0.82
6	methomyl	81.14	54.51	108.41	2.88±0.77
7	thiodicarb	59.55	44.05	92.35	1.94±0.39
8	chlorpyrifos	5.10	3.56	7.86	1.65±0.27
9	Chlorpyrifos methyl	13.37	3.04	27.77	0.76±0.18
10	cyanophos	289.24	233.23	358.63	2.61±0.51
11	profenofos	6.12	2.78	9.52	1.29±0.27
12	indoxacarb	1.35	0.23	2.38	0.93±.32
13	imidacloprid	4838.42	2618.65	13479.62	1.01±0.24
14	hexaflumuron	738.81	403.33	1159.97	1.04±0.17
15	endrin	16.28	10.69	26.17	1.68±0.31

a, a.i. : active ingredient, µg ml⁻¹

b, SE : standard error

B. Leaf-dipping bioassay technique

In the Table (2), data show the toxicity of the fifteen tested insecticides mentioned previously against the 4th instar larvae of parent strain by feeding-dip bioassay technique. The LC50 values indicate that the indoxacarb was the most toxic compound (0.96 Ug/ml), since it was more toxic than chlorpyrifos, endrin, cypermethrin, profenofos, thiodicarb, methomyl, spinosad, abamectin, spinetoram, fenvalerate, chlorpyrifos-methyl, cyanophos, hexaflumuron and imidacloprid with 14.35, 16.0, 22.78, 29.94, 31.36, 73.73, 106.11, 128.80, 134.84, 193.16, 211.91, 400.89, 1282.98 and 11367.43 fold, respectively. Slope values in the same table were more than 1.0 for most tested insecticides. The least slope was (0.67) with imidacloprid and the highest one was (4.64) with fenvalerate. This indicates that the insect population was relatively heterogenous in their susceptibility toward tested insecticides by leaf dip method.

Comparing the toxicity of both methods, according to LC50 values (Tables 1&2), it could be concluded that generally the tested insecticides can be divided into three groups. The first group is the most toxic compounds (cypermethrin, indoxacarb and chlorpyrifos). The second group is the moderate toxic compounds (profenofos, chlorpyrifos-methyl, endrin, thiodicarb, methomyl, abamectin, fenvalerate, spinosad and spinetoram). While the third group is the least toxic compounds (cyanophos, hexaflumuron and imidacloprid). The regression lines obtained by both methods (Tables 1& 2) showed that parent strain had high level of homogeneity toward fenvalerate and methomyl (4.6 and 2.88, respectively), while chlorpyrifos-methyl and indoxacarb showed low slope values (< 1) The same strain showed moderate homogeneity (4.6 > slopes >1.0) toward the rest tested compounds which means that the homogeneity was not the same in the tested population. This finding may be explained by expecting that there are at least two susceptible genotypes which are usually phenotypically different because complete dominance or complete receptivity is unusual (Tsukamoto, 1983). cypermethrin, chlorpyrifos,

Table 2. Toxicity of the tested insecticides to 4th instar larvae of the parent field strain (PS) of cotton leafworm, *S. littoralis* using leaf-dip method.

S.	Insecticide	LC50 a	95 % Confidence limits		Slope ± SE b
			Lower	Upper	
1	spinosad	101.87	30.51-194.17		1.27±0.31
2	spinetoram	129.45	78.15-223.38		1.49±0.32
3	abamectin	123.65	64.58-267.04		0.70±.13
4	cypermethrin	21.87	11.46-1989.97		1.17±0.49
5	fenvalerate	185.43	153.75-215.98		4.64±0.81
6	methomyl	70.78	43.59-91.11		2.88±0.75
7	thiodicarb	30.11	23.46-40.78		2.73±0.57
8	chlorpyrifos	13.78	11.12-18.13		3.37± 0.75
9	Chlorpyrifos methyl	203.43	137.37-295.99		2.17±0.34
10	cyanophos	384.85	288.69-560.93		2.21±0.45
11	profenofos	28.74	18.66-47.88		1.70±0.33
12	indoxacarb	0.96	0.38-1.55		1.73±0.42
13	imidacloprid	10912.73	3928.37-1685488.50		0.67± 25
14	hexaflumuron	1231.66	519.73-8537.30		0.78± 26
15	endrin	15.36	13.08-19.53		3.21 ±0.53

a, a.i. : active ingredient, µg ml⁻¹

b, SE : standard error

chlorpyrifos-methyl, profenofos and imidacloprid were more toxic in larval dipping method than feeding method by 24.85, 2.7, 15.22, 4.7 and 2.26 fold, respectively. Whereas, the toxicity differences between the two methods were less than two fold among the rest of tested compounds. Interestingly, spinosad and abamectin had a moderate position activity against the 4th instar larvae of cotton leafworm in both methods. These results coincide with that obtained by Zhao et al.(2004) who tested 15 insecticides to the tobacco caterpillar, *Spodoptera litura* (Fab.) in the laboratory. Gupta et al. (2005) assessed the toxicity of some new insecticides having novel modes of action

against second-instar (5-day-old) larvae of American bollworm *Helicoverpa armigera* (Hubner) using Potter tower spray method. Based on the LC50 values, the new insecticides, such as emamectin benzoate, indoxacarb, abamectin and spinosad were highly toxic to the tested insect. The most toxic compound was emamectin benzoate (SG 5%) while the least toxic one was betacyfluthrin (SC.2.5%). The present results showed that the toxicity of spinosad in the leaf-dip bioassay was more toxic than the larval-dip method by ~ 1.7 fold. Regarding to the second bioinsecticide tested, abamectin, data in (Table 1&2) showed that the toxicity of this compound in the larval-dip method was more toxic than the leaf-dip method by ~1.5-fold. It was observed that leaves treated with high concentration of abamectin were not preferable to the larvae. This observation suggests that abamectin has antifeedant effect (El-Malla et al., 2003 and Corbitt et al. (1985) who found that toxicity of abamectin on cabbage leaves sprayed with 4.5 ppm was more pronounced for 1st than 3rd instar larvae of *S. littoralis*, since 100% mortality was observed after 96 hr. Abamectin would therefore be expected to give a certain degree of foliar protection against this species. Some studies stated that the novel insecticides were more toxic than conventional insecticides. Adamczyk et al. (1999) reported that novel insecticides chlorfenapyr, methoxyfenozide, spinosad, and tebufenozide were more toxic than conventional insecticide thiodicarb toward the third instar larvae of fall armyworm, *S. frugiperda* using diet bioassay. In our study, indoxacarb was the most toxic compound using the leaf-dip method. Wing et al. (2000) reported that indoxacarb is a novel oxadiazine insecticide which has good field activity against number of lepidopteran pests, as well as certain homoptera and coleoptera.

Several species of lepidopteran larvae can rapidly metabolize the active ingredient after ingestion, but more slowly after topical treatment. This conversion is correlated with the appearance of neurotoxic symptoms. The mode of action of indoxacarb seems to be voltage dependent blocker of Na⁺-dependent compound action potentials when tested in a *Manduca sexta* larvae. Indoxacarb's inherent activity against Lepidoptera is comparable to the most potent insecticides ever commercialized. The high toxicity of pyrethroid insecticide cypermethrin in the present results was in agreement with certain studies on other lepidopteran insects (El-Ghareeb,1985; Ascher et al.,1986; Murugesan and Dhingra, 1995 and Brempong-Yeboah et al.,1984).

From the previous results it can be conclude that cypermethrin, indoxacarb and chlorpyrifos were the most toxic compounds. While cyanophos, hexaflumuron and imidacloprid were the least toxic ones. The rest of tested compounds were occupied a moderate position. According to slope values, the insect population tested was relatively heterogenous in their susceptibility toward tested insecticides by both techniques, larval-dip and leaf-dip bioassay.

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سمية مبيد الأسيبنوساد والإبامكتين مقارنة ببعض المبيدات التقليدية على دودة ورق القطن
حسام عزالدين - عبدا لرؤف محمد الغريب - عرفات محمد كامل السيد و
جمال عبد اللطيف محمد عبد الله
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تم اختبار سمية خمسة عشر مبيدا من مجموعات مختلفة ضد يرقات العمر الرابع لدودة ورق القطن للسلالة حساسة . تم استخدام طريقتين حيويتين لقياس سمية هذه المبيدات . الطريقة الأولى هي غمر اليرقات في محلول المبيد والطريقة الثانية هي تغذية اليرقات على أوراق معاملة بالمبيد . بالنسبة لطريقة غمر اليرقات أظهرت نتائج السمية أن مبيد السبيرمثرين هو أكثر المبيدات سمية (التركيز السام النصفى له ٠.٨٨ ميكروجرام/مل) بينما كان مبيد الاميداكلوبريد هو أقل المبيدات سمية (التركيز السام النصفى له ٤٨٣٨ ميكروجرام/مل) وكان ترتيب المبيدات تبعا لسميتها كالتالى :

السبيرمثرين ، اندوكساكارب ، كلوروبيروفوس ، بروفينوفوس ، كلوربيروفوس
مثيل ، الاندريين ، ثيوداكارب ، ميثوميل، أبامكتين ، فينفااليرات ، سبينتورام ، سبينوساد ،
سيانوفوس ، هكسافلورميرون ، أميداكلوربريد . أما فى طريقة تغذية اليرقات غلى أوراق
الخروج المعاملة بالمبيد كان مركب الاندوكساكارب هو أكثر المركبات سمية (التركيز
السام النصفى ٠.٩٦ ميكروجرام/مل) بينما مركب الاميداكلوبريد هو أقل المركبات سمية
(لتركيز السام النصفى له ١٠٩١٢ ميكروجرام/مل) وكان مركب الاندوكساكارب أكثر
سمية عن مركبات الكلوربيروفوس والاندرين والسبيرمثرين والبروفينوفوس والثيوداكارب
والميثوميل والأسيبنوساد والإبامكتين وأسبينتورام والفينفااليرات والكلوربيروفوس مثيل
والسيانوفوس والهكسافلورميرون والاميدا كلوربريد بزيادة ١٤٣٧ر ، ١٦ر ، ٢٢٧٨ر ،
٢٩٩٤ر ، ٣١٣٦ر ، ٧٣١٣ر ، ١٠٦١١ر ، ١٢٨٨ر ، ١٣٤٨٤ر ، ١٩٣١٦ر ،
٢١١٩١ر ، ٤٠٠٨٩ر ، ١٢٨٣ر ، ١١٣٦٧ر مرة بالترتيب . قيم للميل لخطوط السمية فى
كلتا الطريقتين أظهرت أن مجتمع الحشرات المستخدم فى تقدير السمية غير متجانس نسبيا .