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EFFICACY OF TWO BIOINSECTICIDES, ABAMECTIN AND SPINOSAD ON THE LARVAL STAGE OF LABORATORY AND FIELD STRAINS OF *Spodoptera littoralis* L. (Lepidoptera: Noctuidae).

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ABSTRACT

The present investigation was carried out to evaluate the activity of abamectin and spinosad on the development of *S. littoralis*. Pupation, pupal weight and adult emergence were significantly decreased and this decrease was dependent on both of insecticidal agent and its concentration. The obtained results clearly indicated that the field strain of *S. littoralis* exhibited low resistance towards abamectin and spinosad. The insecticidal activity of spinosad was considered as the most effective, and LC_{50} values reflected the sensitivity of 2^{nd} instar larvae than 4^{th} ones. In addition, the fecundity and fertility of the emerged adults significantly decreased, regardless the applied insecticide and the developmental stage bioassayed.

INTRODUCTION

The cotton leafworm, *Spodoptera littoralis* is considered one of the most destructive pests of cotton, vegetables and ornamental plants throughout the world. Synthetic insecticides are the most commonly agents used to control *S. littoralis* on cotton fields. However, *S. littoralis* have developed resistance to several of these insecticides. Therefore, there is a need to identify novel insecticides with unique mode of action and with minimal impact on associated biological control agents (Jones *et al.*, 2005).

Spinosad is a reduced-risk insecticide with a novel mode of action that provides as alternative to older groups of insecticides such as organophosphates, carbamates and pyrethroids (Clevenland *et al.*, 2002). The new insecticidal compounds include avermectins acting at gamma aminobutryic acid (GABA) and glutamate receptors in proximity to chloride channels (Bloomquist, 1996), and spinosad acting at novel site on the nicotinic acetylcholine receptor (Bloomquist, 1996 and Salgado, 1999). Abamectin instability as well as its low water solubility and tight binding to soil, prevent it from leaching into ground water or entering the aquatic environment (Lasota and Dybas, 1990).

The aim of the present work was to elaborate the variation of susceptibility in both of laboratory and field strains of *S. littoralis* larvae towards abamectin and spinosad. Also, the present study was carried out to evaluate the effects of different concentrations of abamectin and spinosad on the development of laboratory strain larvae. Fecundity and fertility of moths emerged from treated larvae were determined.

MATERIALS AND METHODS

Laboratory strain of the cotton leafworm, *Spodoptera littoralis* was obtained from a laboratory culture reared at Zoology Department, Faculty of Science, Menoufyia University. The larvae were grown on castor bean leaves, *Rincinus communis* at 28 ± 2°C and 60 ± 5% R.H. The emerged adults were fed on sugar solution (10%) and supplied with strips of soft tissue paper as substrate for egg deposition. The field strain of the cotton leafworm was collected from Menoufyia governorate as adults and the toxicological studies were adopted on their progeny (F1 generation). The commercially available bioinsecticides, Spinosad [240 g (AI)/ liter EC], and Abamectin [18g (AI)/liter EC] were obtained from the central laboratory of agricultural pesticides.

BIOASSAYS

Thirty larvae of 2nd and 4th instar larvae were allowed to feed for 48h. on leaves of *Rincinus communis* treated with different concentrations of bioinsecticide. The leaves presented were kept over a layer of sawdust (to absorb frass) in glass jars (500mg capacity). Mortality levels were determined and the insecticidal activity (LC_{50}) of both bioinsecticides was determined for both strains. Control experiments were conducted for each bioassay. Different treatments and control were replicated three times. Biological parameters such as pupation, pupal weight and adult emergence were determined for different treatments of laboratory strain. Resistant ratio (RR) for each insecticide was calculated after Mushtaq and Robert (2004) by dividing LC_{50} of field strain by LC_{50} of laboratory strain. The newly emerged moths treated as larvae were further observed for fecundity and fertility. Fecundity was estimated as the total number of eggs laid per female. Fertility was expressed as the number and percentage of hatched eggs.

STATISTICAL ANALYSES

LC_{50}; concentration-mortality relationship was determined using Probit analysis. The difference between means was assessed using one-way ANOVA analysis. The statistical analyses were carried out using the computer program SPSS (version 11.0).

Table (1): LC_{50} values of laboratory strain larvae of *S. littoralis* treated as 2nd and 4th instar larvae.

Insecticide	2nd					4th				
	LC_{50} (ppm)	Slope	Intercept	95% confidence Lower	95% confidence Upper	LC_{50} (ppm)	Slope	Intercept	95% confidence Lower	95% confidence Upper
Abamectin	86.58	0.008	-0.777	51.03	122.88	119.68	0.008	-1.026	90.19	161.25
Spinosad	54.40	0.014	-0.789	18.53	83.87	81.18	0.010	-.883	45.89	115.76

Table (2): LC_{50} values of field strain larvae of *S. littoralis* treated as 2nd and 4th instar larvae

Insecticide	2nd						4th					
	LC_{50} (ppm)	Slope	intercept	95% confidence Lower	95% confidence Upper	RR	LC_{50} (ppm)	Slope	Intercept	95% confidence Lower	95% confidence Upper	RR
Abamectin	173.12	0.006	-1.070	131.61	275.78	1.99	168.48	0.008	-1.358	151.40	191.18	1.40
Spinosad	92.00	0.010	-0.972	64.49	121.71	1.69	109.10	0.009	-1.047	77.29	150.15	1.34

Resistant ratio (RR) = LC_{50} for field strain / LC_{50} for laboratory strain

RESULTS

The response of larvae of both laboratory and field strains was demonstrated by Table 1 & 2 and Fig.1, through LC_{50} values of abamectin and spinosad. LC_{50} values indicated that spinosad was more toxic agent against 2nd and 4th instar larvae of *S. littoralis* followed by abamectin. Spinosad was 1.59- and 1.47-fold more toxic than abamectin against 2nd and 4th instar larvae of laboratory strain, respectively. LC_{50} values reflected the higher sensitivity of 2nd instar larvae than 4th ones (Table 1 &2 and Fig. 1). Based on LC_{50}, toxicity of abamectin and spinosad had decreased against larvae of field strain (Table 2). Resistant ratio was 1.99- and 1.69 – fold for 2nd instar larvae, and 1.40- and 1.34-fold for 4th instar larvae, respectively for abamectin and spinosad (Table 2).

The effect of abamectin and spinosad on the development of 2nd instar larvae of the laboratory strain was represented by Table (3). This insecticidal activity illustrated by the significant decrease in pupation, pupal weight and adult emergence, and spinosad was the most effective. The corresponding figures for the 4th instar larvae indicated that, the effects of the bioinsecticides were lower than those obtained with 2nd instar larvae (Table 4). However, these findings showed that the 4th larval instar was more resistant than 2nd ones toward the tested insecticides.

Results obtained in Table 5 & 6 clearly indicated that there is a significant decrease in the fecundity and fertility of the emerged adults treated as 2nd or 4th instar larvae. The fecundity of the resulting moths is calculated as the number of deposited eggs per female and as a percentage of control. The fecundity as percentage of control was 85.87 and 35.14 for abamectin; 78.59 and 22.48 for spinosad, at 25 and 150 ppm, respectively, for adults developed from 2nd instar larvae (Table 5). The corresponding values reported in females treated as 4th instar larvae were slight more, indicating resistance of 4th instar larvae than 2nd ones (Table 5 & 6). On the other hand, the fertility of deposited eggs was affected as the number of eggs hatched significantly decreased in a concentration-dependent manner (Table 5 &6). This decrease was more pronounced with spinosad than abamectin, regardless the adults treated as 2nd or 4th instar larvae.

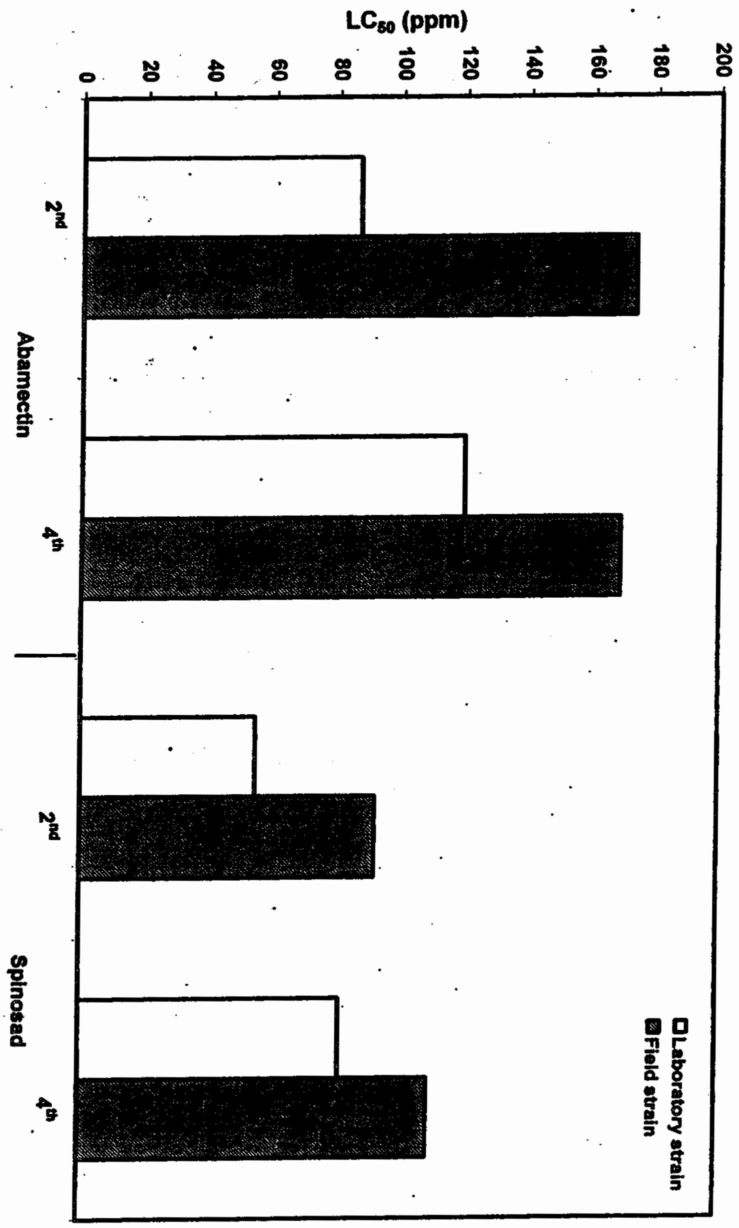

Fig. (1): LC_{50} values of two bioinsecticides (abamectin and spinosad) against 2nd and 4th instar larvae of both laboratory and field strain of *Spodoptera littoralis*.

Table (3): Effect of Abamectin and Spinosad on the development of *S. littoralis* treated as 2nd instar larvae

Concentration (ppm)	Pupation (%) Mean ± SD	Pupation 95% confidence Lower	Pupation 95% confidence Upper	Weight of one pupa (mg) Mean ± SD	Weight 95% confidence Lower	Weight 95% confidence Upper	Adult emergence (%) Mean ± SD	Adult emergence 95% confidence Lower	Adult emergence 95% confidence Upper
Control	89.99± 6.66	73.43	106.55	245.33 ± 5.03	232.83	257.83	92.74 ± 3.17	84.85	100.64
Abamectin									
25	64.44 ± 3.84	54.88	73.99	229.00 ± 4.58	217.61	240.38	77.59 ± 2.50	71.36	83.81
50	56.66 ± 8.81	34.76	78.56	219.33 ± 3.78	209.92	228.73	72.36 ± 3.23	64.32	80.39
100	43.33 ± 6.67	26.76	59.89	200.66 ± 8.14	180.43	220.89	59.49 ± 5.44	45.97	73.01
150	31.11 ± 10.18	5.81	56.40	192.00 ± 8.18	171.66	212.33	54.99 ± 13.63	21.11	88.87
F value	26.32			35.63			14.00		
P value	0.000			0.000			0.000		
Spinosad									
25	55.55 ± 5.09	42.90	68.20	**219.00 ± 15.09**	**181.94**	**256.50**	70.08± 4.69	58.42	81.74
50	47.77 ± 5.10	35.12	60.42	200.66 ± 14.18	**165. 41**	**235.91**	64.85 ± 3.64	55.80	73.90
100	24.44 ± 5.09	11.79	37.09	**181.00 ± 10.14**	**155.78**	**206.21**	45.76 ± 3.75	36.43	55.08
150	15.55± 5.12	2.90	28.20	**169.66 ± 13. 42**	**136.30**	**203.02**	35.55 ± 3.85	25.98	45.11
F value	86.57			**18.54**			99.95		
P value	0.000			**0.000**			0.000		

df between groups = 4 df within groups = 10
P < 0.01 = high significant

Table (4): Effect of Abamectin and Spinosad on the development of *S. littoralis* treated as 4[th] instar larvae

Concentration (ppm)	Pupation (%)			Weight of one pupa (mg)			Adult emergence (%)		
	Mean ± SD	95% confidence Lower	Upper	Mean ± SD	95% confidence Lower	Upper	Mean ± SD	95% confidence Lower	Upper
Control	91.10 ± 5.09	78.45	103.75	239.33 ± 4.50	228.13	250.53	95.09 ± 2.18	89.68	100.51
Abamectin									
25	76.66 ± 6.66	60.10	93.22	225.00 ± 13.22	192.13	257.86	84.22 ± 3.70	75.01	93.42
50	67.77 ± 5.09	55.12	80.42	212.66 ± 6.42	196.69	228.63	80.25 ± 1.45	76.64	83.85
100	59.99 ± 6.66	43.43	76.55	203.00 ± 16.70	161.50	244.49	71.99 ± 3.13	64.21	79.76
150	37.77 ± 5.10	25.12	50.42	199.66 ± 9.50	176.05	223.27	65.05 ± 4.41	54.09	76.01
F value	35.51			6.63			39.94		
P value	0.000			0.007			0.000		
Spinosad									
25	65.55 ± 8.39	44.70	86.39	220.33 ± 5.03	207.83	232.83	74.73 ± 1.88	70.03	79.42
50	59.99 ± 6.66	43.43	76.55	204.66 ± 10.01	179.78	229.54	68.65 ± 3.61	59.68	77.62
100	33.33 ± 6.67	16.76	49.89	192.33 ± 7.50	173.68	210.97	48.05 ± 16.16	7.90	88.20
150	25.55 ± 6.93	8.31	42.78	170.00 ± 8.54	148.77	191.22	33.96 ± 5.74	19.70	48.23
F value	44.62			38.12			26.85		
P value	0.000			0.000			0.000		

df between groups = 4 df within groups = 10

• $P < 0.01$ = high significant

Table (5): Effect of Abamectin and Spinosad on the fecundity and fertility of adults treated as 2[nd] instar larvae

Concentration (ppm)	Fecundity: No. of eggs / female, Mean ± SD	95% confidence Lower	95% confidence Upper	As % of* control	Fertility: No. of eggs hatched, Mean ± SD	95% confidence Lower	95% confidence Upper	% of hatched eggs
Control	1043.33 ± 60.27	893.59	1193.07		803.33 ± 47.25	685.93	920.72	76.99
Abamectin								
25	896.00 ± 137.57	554.23	1237.76	85.87	653.00 ± 90.20	428.91	877.08	72.87
50	770.00 ± 72.80	589.13	950.86	73.80	466.00 ± 66.12	301.74	630.25	60.51
100	600.33 ± 100.06	351.76	848.90	57.53	298.33 ± 48.50	177.85	418.81	49.69
150	366.66 ± 84.00	157.97	575.35	35.14	121.00 ± 19.67	72.13	169.86	33.00
F value	23.03				63.46			
P value	0.000				0.000			
Spinosad								
25	820.00 ± 96.94	579.16	1060.83	78.59	590.33 ± 64.84	429.25	571.40	71.99
50	680.66 ± 100.61	430.71	930.62	65.23	400.00 ± 55.01	263.32	536.67	58.76
100	510.00 ± 53.73	376.52	643.47	48.88	230.00 ± 47.03	113.16	346.83	45.09
150	234.66 ± 12.50	203.60	265.72	22.48	48.33 ± 9.29	25.25	71.41	20.59
F value	53.86				111.71			
P value	0.000				0.000			

***Fecundity calculated as % of control** = $\frac{\text{No. of eggs / female in treatment}}{\text{No. of eggs / female in control}} \times 100$

df between groups = 4 **df within groups = 10**

P < 0.01 = high significant

Table (6): Effect of Abamectin and Spinosad on the fecundity and fertility of adults treated as 4th instar larvae

Concentration (ppm)	Fecundity: No. of eggs / female Mean ± SD	95% confidence Lower	95% confidence Upper	As % of control *	Fertility: No. of eggs hatched Mean ± SD	95% confidence Lower	95% confidence Upper	% of eggs hatched
Control	1025.33 ± 57.76	881.84	1168.81		810.33 ± 102.00	556.94	1063.71	79.03
Abamectin								
25	868.00 ± 36.71	776.79	959.20	84.65	618.00 ± 106.88	352.47	883.52	71.19
50	760.66 ± 63.84	602.06	919.26	74.18	475.00 ± 54.67	339.18	610.81	62.44
100	599.00 ± 133.63	267.04	930.95	58.42	329.66 ± 35.92	240.43	418.89	55.03
150	430.00 ± 42.33	324.84	535.15	41.93	133.00 ± 23.06	75.70	190.29	30.93
F value	28.26				38.14			
P value	0.000				0.000			
Spinosad								
25	828.00 ± 20.42	777.27	878.72	80.75	585.00 ± 43.03	478.09	691.90	70.65
50	680.00 ± 64.83	518.93	841.06	66.32	410.66 ± 23.71	351.75	469.57	60.39
100	513.00 ± 36.04	423.46	602.53	50.03	240.33 ± 37.87	146.25	334.41	46.84
150	211.00 ± 21.65	157.20	264.79	20.57	47.00 ± 6.55	30.71	63.28	22.27
F value	148.42				92.09			
P value	0.000				0.000			

*** Fecundity calculated as % of control =** $\frac{\text{No. of eggs / female in treatment}}{\text{No. of eggs / female in control}} \times 100$

df between groups = 4 **df within groups = 10**

P < 0.01 = high significant

DISCUSSION

Extensive studies have been conducted to support the safety of agricultural uses of abamectin to man and environment. Abamectin is highly unstable to light and has been shown to photodegrade rapidly on plant and soil surfaces and in water following agricultural applications (Lasota and Dybas, 1990). Spinosad is a highly effective bioinsecticide against a broad range of agriculturally important insect pests and this agent has an excellent environmental and mammalian toxicological profile (Romi *et al.*, 2006).

LC_{50} values reported in the present study showed that spinosad was the most effective against larvae of *S. littoralis* compared with abamectin. The LC_{50} values obtained in the present investigation were 92.00 and 173.12 for spinosad and abamectin, respectively against 2nd instar larvae. Similarly, it was found that spinosad has potential for the control of the cigarette beetle, *Lasioderma serricorne* F. and the tobacco moth, *Ephestia elutella* (Hiibner) in stored tobacco, since 100% control of both pests could be achieved at 50 mg/kg (Blanc *et al.*, 2004). Also, it was found that spinosad (incorporated with artificial diet) was toxic to neonate larvae of gypsy moth, *Lymantria dispar* (L.) with LC_{50} of 20 mg/liter (Wanner *et al.*, 2002).

The present data demonstrated that larvae of *S. littoralis* (field strain) collected from Menoufyia governorate, exhibited low level of resistance to abamectin and spinosad, where the resistant ratio (RR) ranged from 1.34 to 1.99. Similarly, most field populations of diamondback moth, *Plutella xylostella* were susceptible to spinosad, but populations from Thailand and Hawaii showed high levels of resistance (Zhao *et al.*, 2002). Also, the magnitude of resistance induced in the horn fly, *Haematobia irritans* ranged from < 3-fold with ivermectin to 1470-fold with permethrin (Byford *et al.*, 1999). Similar results were achieved by Huang and Subramanyam (2004), who reported that the field strain of rice moth, *Corcyra cephalonica* (Stainton), in Weslaco, Texas was highly susceptible to spinosad at 0.5 and 1mg/kg. In addition, a similar study reported that LD_{50} and LD_{95} values of spinosad for Indian meal moth, *Plodia interpunctella* (Hubner) field strain was 1.7-times greater than value for corresponding laboratory strain (Huang *et al.*, 2004). A field population of *Plutella xylostella* L. from Pakistan was found to be highly resistant to deltamethrin (> 500-fold) but had low or no resistance to spinosad and abamectin, when compared with a susceptible laboratory population (Sayyed *et al.*, 2005).

From the results obtained in the present work, one can conclude that spinosad and abamectin affected the development of treated larvae, as pupation, pupal weight and adult emergence significantly decreased. These effects were dependent on concentration and developmental stage bioassayed. Similarly, spinosad at 0.5 and 1 mg/kg leads to a reduction in larval survival of rice moth, *Corcyra cephalonica,* as well as egg-to-adult emergence was decreased (Huang and Subramanyam, 2004). Also, a spry application of emamectin at concentration 25mg AI/litre in a cotton field resulted in over 90% suppression of *Helicoverpa armigera* and *Spodoptera littoralis* larvae (Ishaaya *et al.*, 2002). 100% mortality was detected by spinosad in both larval and adult stages of *Ceratothripoides claratris,* a major thrips pest on tomatoes, regardless of the concentration tested (Premachandra *et al.*, 2005).

The present results showed that different concentrations of abamectin and spinosad significantly decreased reproduction of *S. littoratlis* developed from treated larvae. Field trials clearly detected that spinosad at a concentration of 10 ppm inhibited the reproduction of *Aedes aegypti* for the entire 22-week period of the first trial (Bond *et al.,* 2004). In addition, ovicidal properties of spinosad have been reported for lepidopteran species (Bret *et al.,* 1997). At 20 ppm of spinosad, the average number of adult progeny produced by each braconid female, *Chelonus insularis* was reduced by about 70% compared to the control (Penagos *et al.,* 2005). Emara and Younes (2006) came to the same findings on the effects of abamectin and spinosad on the larvae of flesh fly, *Parasarcophaga aegyptiaca,* who reported the highest insecticidal activity of spinosad than abamectin.

CONCLUSION

Based on the low values of resistant ratio of abamectin and spinosad against the field strain of *S. littoralis,* these results suggest that spinosad and abamectin are potentially potent compounds for control of *S. littoralis.* Also, abamectin and spinosad might be a promising candidate for future cotton leafworm control, as the present results obviously demonstrated a significant decrease on development and reproduction of *S. littoralis.* However, the net results and findings reported here clearly consistent with the previous results obtained by Williams *et al.* (2004), who came to conclusion that spinosad formulation resulted in a good and promising levels of control for *Spodoptera frugiperda.*

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