A. Cotton

Spodoptera littoralis (Boisd.)

Pectinophora gossypiella Saunders

Earias insulana Boisd

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INTRODUCTION

The present study was planned to evaluate the Bioenhancer compound in combinations with chemical insecticides and/or bio-insecticides against major cotton insect pests under Egyptian field and lab conditions. In addition, the adverse influence of these combinations on the natural enemies associated with these pests was also estimated.

MATERIALS AND METHODS

Pesticides used:

1. **Bioenhancer:** It is an insect feeding stimulant and attractant. It contains 35% active ingredients (disaccharides, hydrolyzed starch, whey and vegetable oil) and 65% inert ingredients. Application rate was 5% /feddan (feddan = acre = 0.4 hectare).

2. Chemical insecticides:

- ♦ Diazinox KZ: Diazinox 40% WP, 4 EC, 14% granules. Formulation: (Organic phosphate insecticide), O-O- diethyl-O-(2- isopropyl-6- methyl-5 pyrimidinly) phosphorothioate. Application rate of 6 Kg/feddan. In laboratory, it was used at concentrations of 10, 20, 40, 80 and 160 ppm.
- Reldan: (Chlorpyrifos-methyl) 2 EC, 25% WP, 1% G, 6 1b/gal oil. Formulation: O, O-dimethyl O-(3, 5, 6-trichloro-2-pyridimyl) phosphorothioate). Application rate of 0.005-0.75, a.i./A. In laboratory, it was used at concentrations of 0.025, .05, 0.1, 0.2, 0.4 and 0.8 ppm.
- ♦ **Dursban:** 48% EC. Application a rate of 1 liter / feddan. In laboratory, it was used at the concentrations 0.05, 0.1, 0.2, 0.4 and 0.8 ppm.

3. Bioinsecticides:

- ◆ Xentari: (Selective bacterial insecticide) *Bacillus thuringiensis* subsp. *aizawai* 35.000 Diamond back moth Units/ mg. Application rate was 454g / feddan. In the laboratory, it was used at concentrations of 5x10⁵,10x10⁵, 20x10⁵, 40x10⁵, 80x10⁵ and 160x10⁵ Diamond back moth Units.
 - ♦ Agerin: Bacillus thuringiensis 32000 International Units/mg. Application rate was 250g/ feddan. The utilized concentrations, in laboratory, were $10x10^5$, $20x10^5$, $40x10^5$, $80x10^5$ and $160x10^5$ I.U. for S.littoralis larvae. In case of P. gossypiella and E. insulana concentrations became $5x10^4$, $10x10^4$, $20x10^4$, $40x10^4$, $80x10^4$ and $160x10^4$ I.U..

Laboratory Experiments:

Procedure: Spodoptera littoralis (Boisd.), Pectinophora gossypiella Saunders and Earias insulana Boisd. were reared under the laboratory conditions, at a constant temperature of $27\pm1^{\circ}$ C and $65\pm5\%$ R.H. S. littoralis were reared using the technique described by **Abdel Hakim (1996)** and **Ibrahim (1974)**. P. gossypiella and E. insulana rearing technique was described by **Abd El-Hafez** et al. (1982).

Parts of castor leaves and cotton green bolls were dipped in each compounds and left till dry, then offered to 4th S. littoralis, 1st instar P. gossypiella, and E. insulana larvae.

The following procedures were followed in all experiments:

- 1- Three replicates of ten larvae each into a cup (6x7.5cm) were fed on potato leaves contaminated with bioenhancer and bioinsecticides for a period of 48 hours. After treatment, the surviving larvae were fed on untreated foods till pupation. Mortality was recorded daily. Also, the percentage of pupation and emerged adults were observed.
- 2- Before introducing the larvae to treated food, they were starved for six hours in order to obtain rapid simultaneous ingestion of the offered food.
- 3- The control tests were conducted using foods dipped in water only and left to dry.

Statistical analysis:

The LC₅₀ was determined by using Finney (1952) and corrected according to Abbott's formula (1925).

Field experiments:

Experiment (1): Planting date was 24 Mars 2000 with Cotton variety "Giza 88".

Target Pests: S. littoralis, P. gossypiella and E. insulana

Design: An area of about a feddan was chosen and divided into 24 equal plots in randomized complete blocks. Each plot was about $42m^2$ (6m x 7m). The plots were specified for 8 treatments with 3 replicates and the untreated (control). Treatments included the Bioenhancer, Agerin, Biofly, the chemical insecticides (Reldan for leafworm & Dursban for bollworms) and their combinations with Bioenhancer (½ Bioenhancer+½ Agerin, ½ Bioenhancer+½ Biofly and ½ Bioenhancer+½ each of the chemical insecticide).

Procedure: The cumulative damage caused by *S. littoralis* larvae was estimated by scoring the damage (0 to 5) of each of 100 randomly chosen leaves in each treatment, according to the size of eaten part of the leaf. The rate of infestation was then, calculated according to the formula given by **Kasopers (1965)**. As for the damage caused by bollworms, 50 green cotton bolls were randomly chosen from each treatment and inspected for any symptoms of infestation, and the percentage of infested bolls subsequently calculated. For predators, samples were taken by 5 randomly double sweeping net strokes/plot (10-strokes/ treatment). The collected predators were transferred to the laboratory for identification and counting.

Spray applications:

Different pesticides were applied by means of 20L. knapsack sprayer using a total volume of 200 L/feddan. Different treatments were applied in bi-weekly interval in the second experiment.

Statistical analysis:

Data were statistically analyzed by ANOVA and mean values were separated by the least significant difference (L.S.D.) procedure (Snedecor and Cochran, 1980) at P = 5%. An estimate for percent reduction for each treatment was calculated using Henderson's formula (Henderson & Tilton, 1955).

RESULTS AND DISCUSSION

Laboratory experiments

Table (1) shows that the LC₅₀ values of 4th instar *S. littoralis* larvae were 0.00, 70.25x10⁵ I.U., 0.062 ppm for bioenhancer, agerin (after 72 hours of treatment) and reldan (after 24 hours of treatment) alone. While these values were 40.96x10⁵ I.U. and 0.056 ppm for the combinations of

different agerin and reldan concentrations with 5% of bioenhancer, respectively. For newly hatched P. gossypiella and E. insulana larvae, the LC₅₀ values were 0.00&0.00, $10.72 \times 10^4 \& 8.51 \times 10^4 \text{ I.U.}$, 0.081&0.095 ppm, 6.72×10⁴&4.63×10⁴ I.U. and 0.057&0.068 ppm for bioenhancer; agerin; dursban; agerin + bioenhancer and dursban + bioenhancer, respectively.

Obtained results indicated that bioenhancer was exhibited a high degree of efficiency against target lepidopterous larvae when combined with chemical insecticide and with bioinsecticides. But when it was used alone, it had no obvious effects.

These results agree with those of **Naguib** *et al.* (1994) who indicated that *E. insulana* larvae were more susceptible to bio-compounds than *P. gossypiella*. While *P. gossypiella* larvae were more susceptible to Esfenvalerate as insecticide than *E. insulana* larvae.

Field experiments

1- Effect of bioenhancer and combinations on target pests

♦ Cotton leafworm and bollworms

Cotton leaves damaged by the cotton leafworm were significantly higher in the control compared with the treatments. Reduction in the pests damage (monthly) reached 12.87, 22.40, 17.49, 34.98, 42.16, 30.42 and 26.35% for bioenhancer, agerin, biofly, reldan, ½ bioenhancer + ½ reldan, ½ bioenhancer + ½ agerin and ½ bioenhancer + ½ biofly, respectively. In case of bollworms, reldan and other combined treatments were significantly different from the untreated control. Neither bioenhancer treatment was significantly different from either agerin and biofly treatments or the untreated control. The damage of bollworms (monthly) was reduced by 18.54, 38.76, 29.22, 61.81, 69.09, 56.76% and 50.86 at different treatments, respectively (Table, 2 & Fig. 1).

2- Effect of bioenhancer and its combinations on predators

♦ In cotton field

The predaceous species collected during the period of the experiment were six coleopterous; Cocinella undecimpunctata, Cydonia vicina var. nilotica Muls and Scymnus spp. (interruptus Goeze, syriacus Mars. and globossus var. pieceus Ws.) (Cocinellidae) and Paederus alfierii Koch (Staphylinidae); two hemipterous Orius spp. (albidipennis Reut. and laevigatus Fieb.) (Anthocoridae); one neuropterous Chrysoperla carnea Steph. (Chrysopidae).

The (monthly) mean number of predators collected from bioenhancer, agerin, biofly, reldan, ½ bioenhancer + ½ reldan, ½ bioenhancer + ½ agerin and ½ bioenhancer + ½ biofly treated plots, were 12.98, 12.25, 11.39, 7.22, 8.56, 12.59 and 11.78 individuals. Correspondent, number in the control was 13.53 predators. Obtained results showed that bioenhancer and the bioinsecticide had the least harmful effect on the entomophagous insect populations. Bioenhancer, agerin and biofly treatments were insignificantly different from either their combined treatments or the untreated control. While significant difference between control or bioenhancer treatment and the chemical insecticides alone or in combination with bioenhancer were found.

The safety of bacterial bio-insecticide on different predatory species was previously reported by McCutcheon et al. (1990), Samy (1999). Also, the effect of insecticides on predaceous insects was discussed by Abo-Elghar et al. (1985) indicated that Coccinella and Chrysoperla tolerated to the insecticidal treatments compared with Scymnus and Paederus, while Farag et al. (1989) indicated that insecticides highly affected Scymnus spp., followed by Orius spp.

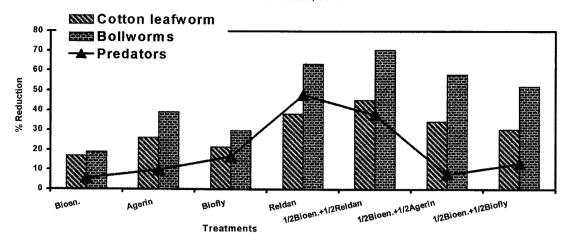
CONCLUSION

Finally, it could be concluded that in laboratory tests, bioenhancer had no obvious effects when used alone. On the contrary, in the field application, it relatively succeeded to control pests alone indirectly, probably because of its low harmful effect on the entomophagous insects. Besides, bioenhancer had a high degree of efficiency against lepidopterous larvae when combined with bio-and chemical insecticides, in both lab. and field applications.

Table (1): Comparative toxicity of bioenhancer, bioinsecticides (after 72 hours of treatments), chemical insecticides(after 24 hours of treatments) and combinations of bioenhancer with different insecticides against S. littoralis, P. gossypiella, and E. insulana larvae.

Treatments	LC ₅₀	Slope
S. littoralis		
Bioenhancer	0.00	0.00
Agerin	10.72 x 10 ⁴	1.18
Reldan	0.081	1.82
Agerin+ Bioenhancer	6.72 x 10 ⁴	1.17
Reldan+ Bioenhancer	0.057	1.74
P. gossypiella Bioenhancer	0.00	0.00
Agerin	10.72 x 10 ⁴	1.18
Dursban	0.081	1.82
Agerin + Bioenhancer	6.72 x 10 ⁴	1.17
Dursban+ Bioenhancer	0.057	1.74
E. insulana		
Bioenhancer	0.00	0.00
Agerin	8.51 x 10 ⁴	1.20
Dursban	0.095	1.69
Agerin + Bioenhancer	4.63 x 10 ⁴	1.42
Dursban+ Bioenhancer	0.068	1.76

Fig.(1): Percent reduction in cotton leafworms damage, bollworms and associated predatory species at different treatments, in Egypt cotton fields, 2000.



Months	% of ave	% of average damage in different treatments										
	Control	Bioen- hancer	Agerin	Biofly	Reldane or Dursban	1/2 Bioen.+ 1/2 Reldane or Dursban	Contract to the Contract of th	1/2 Bioen.4 1/2 Biofly				
Cotton Le	afworm	2 1				1						
June	25.36	24.86	22.00	23.50	19.00	17.17	20.80	21.60				
July	34.74	29.47	26.03	27.93	22.63	19.76	23.56	24.81				
Aug.	43.86	36.23	32.64	34.34	25.95	23.18	27.96	30.16				
Overall	36.99	31.43	28.00	29.78	23.42	20.72	24.86	26.43				
% Reduction	n	-17.05	-26.09	-21.40	-38.19	-45.32	-34.38	-30.28				
L.S.D.	4.3019		<u> </u>			1						
Bollworm	S				· ··							
July	9	6.5	4.5	6	3.5	3	3	4.5				
Aug.	30	24	18	20.5	9.5	7.5	11.50	13.25				

Sept.	50	42	32	36.5	21	17	24	26
Overall	29.75	24.13	18.13	20.88	10.88	8.75	12.50	14.25
% Reduction		-18.90	-39.08	-29.83	-63.45	-70.59	-57.98	-52.10
L.S.D.	11.6428	· ·						

Table (3): Effect of bioenhencer and its combinations on the average numbers of predators counted in different treatments in Egypt cotton fields, season 2000.

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Months	Control	Bioen- hancer	Agerin	Biofly	Reldane or Dursban		1/2 Bioen.+ 1/2 Agerin	½ Bioen. + ½ Biofly
July	19.50	18.75	17.84	16.67	11.00	12.17	18.17	17.00
Aug.	11.25	10.84	10.25	9.34	5.50	6.83	10.42	10.00
Sept.	9.84	9.34	8.67	8.17	5.17	6.67	9.17	8.33
Mean	12.96	12.29	11.75	10.88	6.79	8.12	12.04	11.33
% Reduc	tion	-5.59	-9.76	-16.48	-47.84	-37.61	-7.52	-13.00
L.S.D.	3.9602			1		<u> </u>	I	

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- ♦ Reldan: (Chlorpyrifos-methyl) 2 EC, 25% WP, 1% G, 6 1b/gal oil. Formulation: O, O-dimethyl O-(3, 5, 6-trichloro-2-pyridimyl) phosphorothioate). Application rate of 0.005-0.75, a.i./A. In laboratory, it was used at concentrations of 0.025, .05, 0.1, 0.2, 0.4 and 0.8 ppm.
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Laboratory Experiments:

Procedure: Sesamia cretica Led. were reared under the laboratory conditions, at a constant temperature of 27±1°C and 65±5% R.H. S. cretica were reared using the technique described by **Abdel Hakim (1996)** and **Ibrahim (1974)**.

Parts of maize plants were dipped in each compound and left till dry then offered to 2^{nd} instar S. cretica larvae.

The following procedures were followed in all experiments:

1- Three replicates of ten larvae each into a cup (6x7.5cm) were fed on potato leaves contaminated with bioenhancer and bioinsecticides for a period of 48 hours. After treatment,

the surviving larvae were fed on untreated foods till pupation. Mortality was recorded daily. Also, the percentage of pupation and emerged adults were observed.

- 2- Before introducing the larvae to treated food, they were starved for six hours in order to obtain rapid simultaneous ingestion of the offered food.
- 3- The control tests were conducted using foods dipped in water only and left to dry.

Statistical analysis:

The LC₅₀ was determined by using Finney (1952) and corrected according to Abbott's formula (1925).

Field experiments:

Planting date was April 28th 2000 with Maize variety "Giza 2".

Target Pest: S. cretica

Design: An area of about half feddan was chosen and divided into 18 equal plots in randomized complete blocks with 3 replicates per treatment. Treatments consisted of an untreated control, Bioenhancer, the Bioinsecticide (Xentari), the chemical insecticides (Diazinox) and the combinations ($\frac{1}{2}$ Bioenhancer + $\frac{1}{2}$ Diazinox and $\frac{1}{2}$ Bioenhancer + $\frac{1}{2}$ Xentari) and again two weeks later.

The experimental unit plot was equivalent to 1/100 feddan i.e. 42m². Every plot consisted of 10 rows with 25 hills for seeds separated by 25 cm and 70cm apart.

Spray applications:

Different pesticides were applied by means of 20L. knapsack sprayer using a total volume of 200 L/feddan. Different treatments were applied in bi-weekly interval in the second experiment.

Procedure: At each treatment, random samples of 50 maize plants were taken regularly every week. The population of collected *S. cretica* larvae was counted in the laboratory. Direct counts of predators on 50 plants were also made weekly.

Statistical analysis:

Data were statistically analyzed by ANOVA and mean values were separated by the least significant difference (L.S.D.) procedure (Snedecor and Cochran, 1980) at P = 5%. An estimate for percent reduction for each treatment was calculated using Henderson's formula (Henderson & Tilton, 1955).

RESULTS AND DISCUSSION

Laboratory experiments

Table (1) shows the LC₅₀ values of 2^{nd} instar *S. cretica* larvae treated with bioenhancer; xentari; diazinox; bioenhancer + xentari and bioenhancer + diazinox, reached 0.00, $25.60x10^5$ D.U., 20.20 ppm, $14.57x10^5$ D.U. and 13.18 ppm, respectively.

Obtained results indicated that bioenhancer was exhibited a high degree of efficiency against target lepidopterous larvae when combined with chemical insecticide and with bioinsecticides. But when it was used alone, it had no obvious effects.

Field experiments

1- Effect of bioenhancer and combinations on target pests

♦ The corn borer

The population of *S. cretica* larvae reduced significantly in diazinox alone and its combination with Bioenhancer compared with the control. The two B.t. applications alone or in combination with Bioenhancer were numerically best although none of them were significantly different from each of the other treatments (Table, 4). The (monthly) reduction in *S. cretica* larvae were 33.96, 47.83, 80.62, 66.75 and 86.13 % for bioenhancer, xentari, diazinox, ½ bioenhancer + ½ xentari and ½ bioenhancer + ½ diazinox, respectively (Fig.,2).

Our findings agree with those of **Fédière** et al. (1997) who showed that a sharp reduction in S. cretica larval population (75 to 7/ 100 plants) when using the chemical insecticide. **Samy** (1999) indicated that the B. thuringiensis formulation Ecotech, Dipel and Thuricide HP. against S. cretica exhibited infestation reduction of 82.99, 54.78 and 56.50%, respectively.

Effect of bioenhancer and its combinations on predators in maize field

Eight predaceous species were collected in this study, six were coleopteran species: Coccinella undecimpunctata, and Scymmus spp. (interruptus Goeze, syriacus Mars., bipunctatus Klug. and punctillum WS.) (Coccinellidae) and Paederus alfireii (Staphilinidae) and two were the hemipterous species: Orius spp. (albidipennis Reut. and laevigatus Fieb.) (Anthocoridae).

The (monthly) mean number of predaceous species associated with *S. cretica*, were 11.78, 11.11, 10.33, 5.00, 10.89 and 5.67 individuals for control, bioenhancer, xentari, diazinox, ½bioenhancer + ½xentari and ½bioenhancer + ½ diazinox, respectively (Table, 5). These numbers were insignificantly lower than those counted in the control after bioenhancer, xentari and bioenhancer + ½ xentari treatments. But insecticides and their combinations had significantly effects compared to control. Predaceous species were reduced (monthly) by 5.69, 12.31, 57.56, 7.56 and 51.87 individuals from the control, at different treatments, respectively (Table, 5 & Fig.2).

The safety of bacterial bio-insecticide on different predatory species was previously reported by McCutcheon et al. (1990), Samy (1999). Also, the effect of insecticides on predaceous insects was discussed by Abo-Elghar et al. (1985) indicated that Coccinella and Chrysoperla tolerated to the insecticidal treatments compared with Scymnus and Paederus, while Farag et al. (1989) indicated that insecticides highly affected Scymnus spp., followed by Orius spp.

CONCLUSION

Finally, it could be concluded that in laboratory tests, bioenhancer had no obvious effects when used alone. On the contrary, in the field application, it relatively succeeded to control pests alone indirectly, probably because of its low harmful effect on the entomophagous insects. Besides, bioenhancer had a high degree of efficiency against lepidopterous larvae when combined with bio-and chemical insecticides, in both lab. and field applications.

Table (1): Comparative toxicity of bioenhancer, bioinsecticides (after 72 hours of treatments), chemical insecticides(after 24 hours of treatments) and combinations of bioenhancer with different insecticides against *S. cretica* larvae.

Treatments	LC ₅₀	Slope
S. cretica		
Bioenhancer	0.00	0.00
Xentari	25.60×10^5	1.27
Diazinox	20.20	1.15
Xentari+ Bioenhancer	14.57×10^5	1.83
Diazinox+ Bioenhancer	13.18	1.65

Fig.(2): Percent reduction of *S. cretica* larvae and associated predatory species at different treatments in Egypt maize fields, 2000.

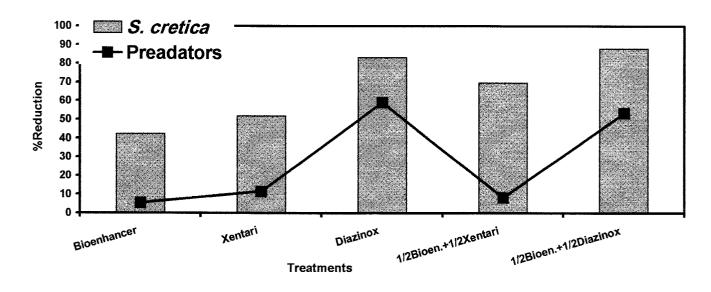


Table (4): Average number of S. cretica larvae/ 50 plants in untreated and treated plots, in Egypt maize fields, 2000.

Months	Average numbers S. cretica larvae / 50 plants									
	Control	Bioenhancer	Xentari	Diazinox		1/2 Bioen.+ 1/2 Diazinox				
May	76.00	55.00	45.50	18.50	29.50	13.00				
June	27.50	14.00	9.00	1.75	5.25	1.50				
July	1.00	0.00	0.00	0.00	0.00	0.00				
Mean	33	20.75	15.88	5.5	10	4				
%Reduction		-42.42	-51.89	-83.33	69.70	-87.87				
L.S.D. 5%	20.0275									

Months	Control	Bioen- hancer	Xentari	Diazimox	1/2 Bioenhancer + 1/2 Xentari	1/2 Bioenhancer + 1/2 Diazimox
May	19.00	17.67	16.33	7.67	17.00	8.33
June	10.00	9.67	9.33	4.33	9.67	5.00
July	6.34	6.00	5.33	3.00	6.00	3.67
Mean	13.21	12.50	11.71	5.42	12.13	6.17
%Reduction		-5.32	-11.34	-58.98	-8.17	-53.30
L.S.D. 5%	4.3012	<u>. t</u>	1	<u> </u>		