COMBINED USE OF *TRICHOGRAMMA EVANESCENS* WEST. AND ENTOMOPATHOGENS FOR CONTROLLING *PECTINOPHORA GOSSYPIELLA* (SOUND.) AND *EARIAS INSULANA* (BOISD.) IN COTTON FIELDS

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INTRODUCTION

Cotton is a crop of great economic importance in Egypt. This plant is susceptible to infestations by several insect pests. Low yield of cotton is mainly related to damage in bolls caused by the pink bollworm *Pectinophora gossypiella* (Sound.) and the spiny bollworm *Earias insulana* (Boisd.).

As biological control is an effective component in modern integrated pest management, the insecticidal activities of *Bacillus thuringiensis* against the pink and spiny bollworms have been widely investigated, *e.g.* El-Husseini & Afifi (1981a & 1981b); Salama *et al.* (1983) and El-Lebody *et al.* (2003). Field trials involving combinations of nuclear Polyhedrosis virus (NPV) or granulosis viruses (GV) with *B. thuringiensis* have been reported by Takchev (1987) and Moawad *et al.* (1998).

Mass-production and release of *Trichogramma* egg parasitoids is regarded as a promising approach to reduce egg hatching and subsequent crop damage by cotton bollworms (Hassan, 1989; Li, 1994; Wajnberg & Hassan, 1994 and Smith, 1996). Principles of parasitoid release in the design of a program were summarized by Smith (1996).

The present work aims to evaluate the efficacy of the egg parasitoid *T. evanescens* alone and in combination with four bioagents of entomopathogens namely, Profect®, Protecto®, Viroset® and Virotecto® for the control of the two bollworms *P. gossypiella* and *E. insulana*.

MATERIAL AND METHODS

1. The egg parasitoid *Trichogramma evanescens*

T. evanescens was obtained from a well established culture maintained at the laboratories of Plant Protection Research Institute. Eggs of the moth *Sitotroga cereallela* Olivier were used as alternative host eggs for rearing the egg parasitoid as described by Sakr (2003). According to this method, host eggs were glued to cards measuring 15.5 x 5.5 cm, approximately 2000 eggs/ card, which were offered to the parasitoid in an environmental cabinet set at 27±1°C, 16 L: 8D photoperiod and 60-70 RH%.

2. Entomopathogens

Four commercial bioagents obtained from Plant Protection Research Institute, Giza, Egypt were evaluated for their efficacy in the control of *P. gossypiella* and *E. insulana* in the field, Table (I).

TABLE (I)
Tested microbial bioagents

Entomopathogens	Commercial name	Recommended rate
Bacillus thuringiensis kurstaki	Protecto®	300 gm/ feddan
Nuclear polyhedrosis virus (NPV)*	Viroset®	300 gm/ feddan
B. thuringiensis + NPV	Profect ®	300 gm/ feddan
Granulosis virus (GV)**	Virotecto®	300 gm/ feddan

*NPV- *Spodoptra littoralis*
**GV-*Phthorimaea operculella*

3. Field experiment

The field experiments were carried out at Shalkan, Qalyubia Governorate in an area of 1400m^2 cultivated with the cotton variety Giza 85. The experiments were conducted during two successive agricultural seasons 2004 and 2005. The area was divided into 10 plots each measuring 140 m^2 and one treatment was used as a control. Each plot was divided into two replicates, each measured 70 m^2. Similarly, a plot was left untreated as a control.

The following experiments were conducted with the beginning of the appearance of the cotton boll, *i.e.* at 1st of July.

3.1. Release of *T. evanescens*

Egg cards bearing *S. cereallela* parasitized eggs were hung on a branch of the cotton plants, one card per plot. Each card carried approximately 2000 parasitized eggs. Every 10 days, these cards were removed and replaced by new ones making a total of 8 treatments.

3.2. Treatment with bioagents

Each of the bioagents was sprayed at the recommended concentration (Table 1) using a hand sprayer, starting from the appearance of the cotton bolls. Spraying was repeated at a 10-days interval making a total of 8 treatments per plot.

3.3. Combined effect of *T. evanescens* and bioagents

An experiment was conducted using the joint effect of the egg parasitoid and a bioagent. Egg cards, were hung on cotton plants as described before. After 10 days, they were removed and the cotton plants were sprayed with one of the bioagents. This procedure was repeated, *i.e.* alternate release of the parasitoid and spraying of a bioagent making a total of four treatments with 10-days intervals.

Cotton bollworm infestation

Five days following release of the egg parasitoid or spraying by bioagents, 100 cotton bolls were randomly collected from each treated plot. This procedure was repeated every 5 days. The collected cotton bolls were transferred to the laboratory and investigated for infestation by the spiny and pink bollworms. Percentage of reduction in infestation was calculated according to formula of Henderson and Tilton (1955).

RESULTS AND DISCUSSION

Trend of infestation by the two cotton bollworms in untreated plots (control) was relatively similar in the two successive cotton agricultural seasons, 2004 and 2005. However, percentage infestation was higher in the former season and more by the spiny bollworm *E. insulana* than *P. gossypiella*. Infestation by the cotton bollworms in the untreated plot were first detected during the third week of July reaching 8.0 and 9.25% for *P. gossypiella* and *E. insulana* in 2004 and 6.5 and 8.25% in 2005 for the respective mentioned insect species. Infestation gradually increased during cotton boll maturation to reach the highest percentages during the last week in August and the 3rd week of September, reaching 25.0 and 18.5% in

2004 and 14.0 and 13.75% in 2005 for *P. gossypiella* and 15.0 and 24.5% in 2004 and 22.0 and 25.75% in 2005 for *E. insulana*, respectively.

The release of the parasitoid *T. evanescens* (8 times at a 10-days interval) during the cotton boll maturation led to the lowest rate of infestation (Tables 2 & 3). Prior to cotton harvest in September 2004, percentage infestation by *P. gossypiella* and *E. insulana* was 2.50 and 3.0%, respectively. Meanwhile, in the year 2005 a lower infestation percentage was found, in the second and fourth of August and third week of September (2%) for *P. gossypiella.* In case of *E.* insulana, it was recorded in the second and fourth week of August (3.5%) (Table 3). This was followed by the joint treatments by the parasitoid with the Profect®; as it reduced infestation for *P. gossypiella* and *E. insulana* to 3.0 and 6.5% in 2004 and 3.25 and 5.0% in 2005. Also, a relatively low rate of infestation was found when the parasitoid was released four times alternatively with 4 sprays by Protecto®, Viroset® or Virotecto®. This effect was more apparent for the control of *P. gossypiella* as infestation was reduced to 5.0, 7.25 and 5.75% at harvest, *i.e.* 3rd week of September in 2004 season and 3.75, 4.0 and 4.75% at the end of 2005 to the respect mentioned treatments. Meanwhile, infestation percentage for *E. insulana* was 9.0, 8.50 and 8.75% at the end of 2004 and 5.50, 7.75 and 7.75% at the end of 2005 for the joint effect of *T. evanescens* release with, Protecto®, Viroset® or Virotecto® respectively. On the other hand, due to the cross reactivity of insect viruses against insect pests, it could be noticed that both NPV and GV represented some effects against pink and spiny bollworms (Srinivasan & Kennedy, 2003)

Similar results were reported by Abu-Nasr *et al.* (1983) using *B. thuringiensis* subsp. *thuringiensis* and subsp. *finitimus* for the control of *P. gossypiella* and *E. insulana* in cotton field.

The two bacterial treatments reduced the number of infested bolls by 42-45%. El-Heneidy *et al.* (2004) released the egg parasitoid, *T. evanescens* for controlling the pink and spiny bollworms in cotton fields that showed significant reductions in the percentages of bollworms infestation. Mansour (2004) found that the rate of damage caused by the cotton bollworms *P. gossypiella* and *E. insulana* was 29.0, 28.38, 26.25 and 20.63% in treatments by *B.t., T. evanescens, T. evanescens* with *B.t.* and chemical insecticides in 2001 and 22.75, 21.88, 19.63 and 29.13 in 2002 for the four treatments, respectively. The infestation was significantly higher in the control than in any of the treatments.

A moderate gradual low rate in infestation by *P. gossypiella* was observed when the virus or *Bacillus* bioagents, Protecto®, Viroset®, Virotecto® and Profect®

TABLE (II)

Efficiency of *Trichogramma evanescens* and commercial bioagent products against *Pectinophora gossypiella* and *Earias insulana* at Shalakan (Qalyubia Governorate) during 2004 cotton agricultural season.

Treatments	% cotton boll infestation										% Reduction
	P. gossypiella					*E. insulana*					
	3rd week of July	2nd week of August	4th week of August	3rd week of September	Mean ±S.D	3rd week of July	2nd week of August	4th week of August	3rd week of September	Mean ±S.D	
Trichogramma	2.25	3.5	2.25	2.50	2.62±0.7^{g}	4.75	2.0	3.0	3.0	3.18±1.60^{j}	83.93
Trichogramma + Profect	3.25	4.5	5.0	3.0	4.43±1.23^{f}	6.50	4.25	3.75	6.5	5.25±1.77^{i}	74.57
Trichogramma + Protecto	3.25	5.75	6.0	5.0	5.0±1.41^{e}	8.25	6.75	7.75	9.0	7.93±1.28^{g}	61.21
Trichogramma + Viroset	3.5	7.5	8.25	7.25	6.62±2.15^{d}	8.50	8.50	7.25	8.50	8.18±1.55^{g}	55.60
Trichogramma + Virotecto	3.5	6.75	8.50	5.75	6.12±1.99^{d}	8.0	5.5	5.25	8.75	6.88±2.06^{h}	61.00
Profect	4.0	7.25	8.25	6.25	6.43±1.75^{d}	8.25	6.25	7.50	10.75	8.19±2.04^{g}	56.14
Protecto	5.0	10.75	10.25	8.75	8.69±2.65^{b}	8.75	7.25	8.50	12.75	9.31±2.44^{f}	41.09
Viroset	5.0	11.25	17.0	10.75	11.0±4.92^{c}	9.0	10.75	7.50	14.75	10.50±3.26^{e}	29.63
Virotecto	4.0	11.75	10.75	9.5	9.0±3.24^{b}	9.5	8.25	13.0	15.75	11.62±3.32^{d}	32.51
Control	8.0	18.50	25.0	18.5	17.5±6.99^{a}	9.25	12.0	15.0	24.50	15.18±6.52^{c}	
					F value 8.50**					F value 7.73**	

Means followed by the same letter in the same column are not significantly different

TABLE (III)

Efficiency of *Trichogramma evanescens* and commercial boagent products against *Pectinophora gossypiella* and *Earias insulana* at Shalakan (Qalyubia Governorate) during 2005 cotton agricultural season.

Treatments	% cotton boll infestation										% Reduction
	P. gossypiella					*E. insulana*					
	3rd week of July	2nd week of August	4th week of August	3rd week of September	Mean ±S.D	3rd week of July	2nd week of August	4th week of August	3rd week of September	Mean ±S.D	
Trichogramma	3.0	2.0	2.0	2.0	2.25±0.68^{g}	4.0	3.50	3.50	4.25	3.81±0.83g	81.82
Trichogramma + Profect	3.25	3.25	4.0	3.25	3.44±0.72^{f}	4.50	5.75	5.0	5.0	5.06±0.99f	71.95
Trichogramma + Protecto	3.25	4.75	3.75	3.75	3.87±0.95^{de}	5.75	4.0	6.0	5.50	5.31±1.49f	66.34
Trichogramma + Viroset	6.25	5.0	4.25	4.0	4.88±1.20^{d}	4.50	7.75	8.75	7.75	7.18±1.93e	60.20
Trichogramma + Virotecto	4.25	4.75	4.75	4.75	4.62±0.88^{cd}	6.0	6.75	7.50	7.75	7.0±1.03e	61.65
Profect	4.5	5.0	5.25	5.75	5.12±0.88^{c}	7.0	8.75	8.5	9.75	8.50±1.36d	50.06
Protecto	5.25	6.0	7.75	6.25	6.31±1.40^{b}	7.0	9.75	11.5	15.25	10.87±3.34cb	43.30
Viroset	5.0	7.50	6.25	8.0	6.68±1.74^{b}	7.5	10.0	14.50	14.50	11.63±3.32b	32.86
Virotecto	7.75	5.25	6.0	5.75	6.18±2.42^{b}	3.75	8.50	14.00	13.75	10.0±4.93c	46.60
Control	6.5	10.5	14.0	13.75	11.19±3.46^{a}	8.25	17.5	22.0	25.75	18.37±7.02a	
					F value 4.56**					F value 6.49**	

Means followed by the same letter in the same column are not significantly different

were tested alone, *i.e.* 8.75, 10.75, 9.5 and 6.25% in the third week of September 2004 compared to 18.5% in the control and 6.25, 8.0, 5.75 and 5.75% at the end of 2005, respectively. Relatively moderate low rate in infestation by *E. insulana* was recorded when Protecto®, Viroset®, Virotecto® and Profect® were used, *i.e.* 12.75, 14.75, 15.75 and 10.75% in the end of 2004 (24.50% in the control) and 15.25, 14.50, 13.75 and 9.75% at the end of 2005 (25.75% in the control), respectively.

A high reduction in infestation was observed when the parasitoid *T. evanescens* was used alone; 83.93 and 81.82% in 2004 and 2005, respectively, followed by joint treatments with the parasitoid + Profect® bioagent reaching reduction of 74.57 and 71.95 in 2004 and 2005, respectively. The use of the parasitoid *T. evanescens* was considered more effective for the control of the cotton bollworms as their eggs were exposed to the parasitoid for a period of 4 days (egg incubation period). Furthermore, newly hatched larvae of *P. gossypiella* were exposed for only a few hours to the bioagent before larvae penetrate the cotton boll.

The statistical analysis showed a highly significant difference between treatments compared with control in 2004 and 2005 seasons.

Combination between nuclear polyhedrosis virus (NPV) or Granulosis viruses (GV) and *B.t.* have been reported by Takchev (1987) who found that, the mixture of Bitoxibacllin (*B.t.* subsp. *thuringiensis*) and a virus preparation (Virin-GVcp) gave 70% technical effectiveness against *Cydia pomonella* on apple. Also, Moawad *et al.* (1998) indicated that, the application of both *B.t.* and GV mixture gave such better results than both of them separately against potato tuber moth, *Phthorimaea operculella* (Zeller) in potato cultivations. Mesbah *et al.* (2003) found that the reduction of boll infestations was 36.5, 41.7 and 25.4% in seasons 1999, 2000 and 2001, respectively using the *Trichogrammatoidea bactrae* for controlling the spiny and pink bollworms. El-Heneidy *et al.* (2004) used *T. evanescens* for controlling the pink and spiny bollworms; the reduction ranged from 48.6-55.5% and 16.4-21.7% when the parasitoid was released early during the flowering stage or a few weeks later during the boll growth stage. Mansour (2004) used *B.t., T. evanescens, T. evanescens* with *B.t.* and chemical insecticides for controlling cotton bollworms. The difference in infestation rate was significantly higher in the control than in any of the treatments.

SUMMARY

Efficacy of the parasitoid *T.* evanescens and four commercial entomopathogenic bioproducts were evaluated for the control of the pink bollworm *P.*

gossypiella and the spiny bollworm *E. insulana* in cotton during the two seasons 2004 and 2005. The lowest rate of bollworm infestation was recorded when *T. evanescens* was released during the cotton boll growth and maturation. Percentage infestation by *P. gossypiella* and *E. insulana* were 2.50 and 3.0% at the end of 2004 season, respectively and 2.0 and 4.25 at the end of 2005 for the respective mentioned insect species. This was followed by the joint treatments by the parasitoid with Profect® bioagent (*B.t.* + NPV) as it reduced infestation by *P. gossypiella* and *E. insulana* to 3.0 and 6.5% in 2004 and 3.25 and 5.0% in 2005, respectively.

A relatively low rate of infestation was found when the parasitoid release was combined with Protecto® (*B.t.*), Viroset® (NPV), or Virotecto® (GV), *i.e.* 5.0, 7.25 and 5.75% at the end of 2004 season and 3.75, 4.0 and 4.75% at the end of 2005 for *P. gossypiella*, respectively.

Meanwhile, infestation percentage of *E. insulana* was 9.0, 8.50 and 8.75% in 2004 and 5.50, 7.75 and 7.75% at the end of 2005 to the respect mentioned bioagents. Meanwhile, spraying by the bioagents, Protecto®, Viroset®, Virotecto® or Profect® singulary resulted infestation percentage of 8.75, 10.75, 9.5 and 6.25% in 2004 and 6.25, 8.0, 5.75 and 5.75% in 2005, respectively for *P. gossypiella.* Infestation percentage for *E. insulana* was 12.75, 14.75, 15.75 and 10.75% at the end of 2004 and 15.25, 14.50, 13.75 and 9.75% when Protecto®, Viroset®, Virotecto® or Profect® were used, respectively.

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