Egyptian Journal of Biological Pest Control, 19(1), 2009, 41-48

Interaction Between the Entomopathogenic Fungus, *Beauveria bassiana* and some Insecticides Against the Whitefly, *Bemisia tabaci* (Genn.) (Homoptera: Aleyrodidae)

Ibrahim A. A.; H. H. Shalaby and H. M. El-Saadany
Plant Protection Research Institute, Agric. Res. Centre, Giza, Egypt

(*Received: March 22, 2009 and Accepted: April 13, 2009*)

ABSTRACT

In vitro inhibition of 4 different insecticides Cord, Imaxi, Orizon and Achook on the entomopathogenic fungus, *Beauveria bassiana* growth was tested. Three concentrations of each insecticide; recommended, half and 1\4 of recommended concentrates (RC) were used. There was no inhibition of mycelial growth with Imaxi at the three concentrates. On the other hand, Orizon and Achook had the same effect (100%) at 1/2RC and 1/4RC, but at RC it exhibited 50% mycelial growth. However Cord exhibited 50% mycelial growth at both RC and 1/2RC, while at 1/4RC mycelial growth was approximate 75%. In addition, the efficacy of tested combinations (Biovar plus each of Cord, Imaxi, Orizon and Achook) was evaluated under greenhouse conditions on cucumber against the whitefly, *Bemesia tabaci* (Genn.) at the recommended and half recommended rates. The combinations of the bioinsecticide: Biovar 4% WP at recommended concentration, (200 gm/100 L.) plus Imaxi 35%EC at its half recommended rate ½RC (37.5 cm^3/100L.); Biovar 4% WP at its RC plus Orizon 11%EC at its ½RC (25 cm^3/100L.) and Biovar 4% WP at its RC plus Achook 0.15%EC at its ½RC (93.7/100L.) achieved significant control of the whitefly, *B. tabaci* by decreasing the application rates of pesticides and obtaining good protection with less residuals. Also, the co-toxicity resulted from adding the Biovar to the Cord induced an irregular joint action against *B. tabaci* stages and resulted in dominated antagonistic effect. An additive effect was dominated in case of the addition of Biovar to Imaxi, Orizon and Achook by which the total mortality was higher than the mortality of summation of each compound separately. Thus, the joint action of two compounds in the mixtures was more effective against the tested insect than each compound alone.

Key words: Interaction, *Beauveria bassiana*, insecticides, *Bemesia tabaci*.

INTRODUCTION

Microbial control in integrated pest management (IPM) programs is an important factor for the reduction of pest population densities. Use of selective pesticides in combination with entomopathogens, increases the efficacy of the control and reduces the use of required insecticides (Oliveira *et al.* 2003). In addition, IPM is an efficient model for reducing the severe impact of chemical pesticides on ecosystem.

The entomopathogenic fungus, *Beauveria bassiana* (Bals.) Vuill. is a potential biocontrol agent occurred naturally. It attacks a large number of insect pests. Recent advances in production and application of insect pathogenic fungi have resulted in improvements in large-standing whitefly mycoinsecticide products based on *Paecilomyces fumosoroseus* and *B. bassiana.* These products have the capacity to suppress and, in some instances to provide good control of whiteflies in both greenhouse and field crops (Faria and Wraight, 2001).

Insecticides may have antagonistic or synergistic effect on the potentiality of *B. bassiana* and may disrupt natural epizootic. Fedorinchik, 1974 reported that under epizootic conditions, it is possible to enhance effectiveness through joint action of a pathogen and compatible insecticides to reduce not only the cost of production, but also the contamination of the environment. It was also shown that sub-lethal concentrate of an insecticide would physiologically make the insect weak up to a desired degree which made it much more susceptible to the attack of the entomopathogens.

The present study aims to open an area for using entomopathogens together with compatible insecticides as multiple mortality factors against target insect pest.

MATERIALS AND METHODS

Laboratory experiments:

Isolation of *Beauveria bassiana*:

B. bassiana was isolated in pure culture on Czapek Dox agar medium (CDA) from infected adults of whitefly collected from Sharkia Governorate, Egypt (Ibrahim, 2006).

Production of the entomopathogenic fungus *B. bassiana*:

Conidia were produced on agar Petri-dishes of semi-synthetic CDA medium, consisted of sucrose 20.0 g/L, K_2HPO_4 1.0 g/L, KNO_3 2.0 g/L, yeast extract 2.0 g/L, KCL 0.5 g/L, $MgSO_4$ $7H_2O$ 0.5 g/L, $FeSO_4$ $7H_2O$ 0.002 g/L, agar-agar 20.0 g/L, up to 1000 ml distilled H_2O. The medium was prepared and adjusted pH (5.5-6.5). Petri-dishes were inoculated with *B. bassiana* and incubated for two weeks at 25 ± 1°C & 50 – 60 % RH. At the end of incubation period, the conidia were harvested from the surface of the culture directly by scraping with sterile solution of 0.01 % tween-80. The conidia were separated by filtration through sterilized glass-wool. The resulted suspension was counted according to the hemocytometer counts technique.

Insecticides:

Four common commercial formulations of insecticides, in use against whitefly in Egypt; the organophosphorus compound, Profenofos (Cord 72%EC); Imidacloprid (Imaxi 35% SC); Acetamiprid+ Abamectin (Orizon 11% EC); the botanical insecticides, Azadirachtin 1500 ppm (Achook 0.15%) were used. These compounds were tested as growth inhibitors for *B. bassiana* by poison food technique on CDA in three replicate; at recommended concentrate (RC), half of the recommended concentrate (1/2 RC) and 1/4 of the recommended concentrate (1/4RC).

Each 100 ml portion of the medium was dispensed into a 250 Erlenmeyer conical flask and autoclaved at 121°C for 20 minutes and then cooled to about 45°C. Stock solutions of the pesticides were prepared in sterilized distilled water and incorporated into each flask to provide different levels of concentrates. Each flask was shaken well and poured into sterilized Petri-plates (90 mm). A medium without insecticides served as a control. Each plate was inoculated with 1 x 10^7 conidiospores from 12 days old culture of *B. bassiana*. The inoculated plates were incubated at 25±1°C. After 15th days of incubation, the growth of *B. bassiana* colony in the Petri-plates treated with different insecticides at different concentrates was recorded.

Greenhouse experiments:

This work was carried out under greenhouse conditions at Central Laboratory for Agricultural Climate, Giza, Egypt. Greenhouse with an area of 540 m^2 was cultivated by cucumber, *Cucumis sativus* L. variety Alfred to evaluate the efficacy of combining certain compounds with *B. bassiana* against the whitefly, *B. tabaci* on cucumber.

Tested compounds:

1- The bioinsecticide, *B. bassiana* (Biovar 4% WP) produced by Plant Protection Research Institute, Egypt.
2-The organophosphorus compound, Profenofos (Cord 72%EC), produced by El Helb Company, Egypt. Chemical name: O-(4-bromo- 2- chlorophenyl) –5-ethyl-S-Propyl phosphoro thioate.
3- Imidacloprid (Imaxi 35% SC), Rotam Co., Egypt. Chemical name: 1-[(6-chloro-3-pyridinyl) methyl]-*N*-nitro-2-imidazolidinimine
4- Acetamiprid+ Abamectin (Orizon 11% EC), EgyChem Co., Egypt. Chemical name: Acetamiprid; N-[(6-chloro-3-pyridinyl) methyl]-N'-cyano-N-methylethanimidamide Abamectin; 5-*O*-demethylavermectin A_{1a} (i) mixture with 5-*O*-demethyl-25-de (1-methylpropyl)-25-(1-methylethyl) avermectin A_{1a}
5-The botanical insecticides, Azadirachtin 1500 ppm (Achook 0.15%), The Egyptian Co. for Agricultural Development, Egypt.

Treatment procedures:

Experimental area (540 m^2) was divided into 69 equal plots (each of $7m^2$), arranged in complete randomized design with three replicates for each treatment. All agricultural practices were applied in this experiment except the studied variant.

Application concentrations:

1- Cord at 187.5 cm^3 and 93.7 cm^3/100L.
2- Imaxi at 75 cm^3 and 37.5 cm^3/100L.
3- Biovar at 200 gm. and 100 gm. /100L.
4- Orizon 11% EC at 50 cm^3 and 25 cm^3/100L.
5- Achook at 187.5 cm^3 and 93.7 cm^3/100L.
6- Combinations of the chemical compounds used:
6. a- Biovar + Cord at 200 gm.+187.5cm^3
 Biovar + Cord at 200 gm. +93.7cm^3
 Biovar + Cord at100 gm. +93.7cm^3
6. b- Biovar + Imaxi at 200 gm. + 75cm^3
 Biovar + Imaxi at 200 gm. +37.5cm^3
 Biovar + Imaxi at 100 gm. +37.5cm^3
6. c- Biovar + Orizon at 200 gm. + 50cm^3
 Biovar + Orizon at 200 gm. + 25cm^3
 Biovar + Orizon at 100 gm. + 25cm^3
6. d- Biovar + Achook at 200 gm. + 187.5cm^3
 Biovar + Achook at 200 gm. + 93.7cm^3
 Biovar + Achook at 100 gm. + 93.7cm^3
7- Control, water used without any insecticides.

By using a knapsack sprayer (20 liters) cucumber plants were treated on April 16th, 2008. Inspection of the plants was carried out one day before spraying and after 1, 3, 5 and 7 days from application to evaluate the effect of different treatments on the numbers of adults and immature stages except eggs of *B. tabaci*.

Direct counts (3 replicates) were inspected at random for whitefly adults (each replicate of 20 leaves, representing upper, middle and lower levels of the plant). Samples were placed in paper bags and transferred to the laboratory to investigate the whitefly immature stages (except eggs). Obtained data were statistically analyzed at 5% probability level according to Snedecor and Cochran (1967).

Percentage of population reduction (% mortality) was calculated according the equation of Hinderson and Tilton (1955) as follow:

$$\text{Reduction (\% mortality)} = \left[\left(\frac{C_b X T_a}{C_a X T_b}\right) - 1\right] X\ 100$$

Where:

T = number of alive insect individuals in treatment.

C = number of alive insect individuals in control.

a = number of alive insect individuals after treatment.

b = number of alive insect individuals before treatment.

While, to evaluate the effect of different pairs of insecticides used here, the following equation was used:

$$\text{Co-toxicity factor} = \frac{\text{Observed \% mortality} - \text{Expected \% mortality}}{\text{Expected \% mortality}} X\ 100$$

This factor was used to differentiate the results into three categories. A positive factor of 20 or more meant potential, a negative factor of 20 or more meant synergistic, and any intermediate value (*i.e.,* between -20 and +20) was considered only additive effect . The expected mortality for the mixture of 2 insecticides was the sum of the expected mortalities of each of dosage used in the combination, (After, Mansour *et al.*, 1966).

RESULTS AND DISCUSSION

1- Effect of the chemical insecticides on *B. bassiana* growth:

Data showed that there was no inhibition of mycelial growth with Imaxi at the three concentrates. This result agree with Anderson and Roberts (1983) who discussed that the formulations obtained in either wettable or in flowable forms caused no inhibition of fungal growth but often increases colony numbers, whereas emulsifiable concentrate formulation showed inhibition of *B. bassiana* germination and Vilas-Boas and Alves (1989) who reported that no inhibition of viability with some insecticides. On the other hand, Orizon and Achook had the same effect at 1/2RC and 1/4RC. But at RC they exhibited 50% mycelial growth. However Cord exhibited 50% mycelial growth at both RC and 1/2RC while at 1/4RC mycelial growth was approximate 75%.

Compatibility in vitro of a mixture of *B. bassiana* isolates Bb9205 and Bb9002, and three insecticides, Diazinon, Isazofos and Metacrifos at RD, 1/2 RD and 1/4 RD, were studied by Marin *et al.* (2000). They showed lowest to heavy growth inhibition due to the differences of the isolates of *B. bassiana*, kind and dosages of insecticides. They demonstrated that there were differences among isolates of a same fungus, which poses the necessity of selecting isolates more resistant to pesticides in IPM programs. These differences of mycelial growth may be due to strain variations.

Yue and Wen (2005) studied compatibility of seven strains of *B. bassiana* with some insecticides. They found that the insecticides, Bt WP, Imidacloprid 10% WP, Spinosad 2.5% SC and the *Dendrolimus* virus WP were compatible with the strains of Bb7001, Bb7004, and Bb8001. But other insecticides exhibited inhibitory effects on the germination of their conidia especially the insecticides Atabron (Chlorfluazuron) 5% EC, Abamectin 2.0% EC and Cascade (flufenoxuron) 5% DC/ml.

Puzari *et al.* (2006) showed that Cypermethrin followed by Deltamethrin, Alphamethrin and Phosphamidon at RC could be tolerated well growth inhibition of <50 percent.

2- Efficacy of combining *Beauveria bassiana* with pesticides against *Bemisia tabaci* (Genn.) infesting cucumber in greenhouse.

2.I. Efficacy of the tested compounds alone:

2.I.1. Whitefly adults:

Results in (Table 1) showed that the average number of the whitefly adults per 20 leaves was noticeably reduced (with some exceptions with Biovar and Achook at ½RC) during the following 3 days after treatment to reach (16.0, 20.0), (10.0, 12.0), (6.0, 12.0), (6.0, 14.0) and (14.0, 20.0 adults/20. leaves) on the 3rd day for the two application rates {recommended (RC) & half recommended (½RC)} of Cord at (187.5 cm^3, 93.7 cm^3/100L.), Imaxi at (75 cm^3, 37.5 cm^3/100L.), Biovar at (200 gm., 100 gm. /100L.), Orizon 11% EC at (50 cm^3, 25 cm^3/100L.) and Achook at (187.5 cm^3, 93.7 cm^3/100L.), respectively. The corresponding reduction rates of adults were (72.4, 67.2), (84.3, 80.3), (89.7, 97.3), (89.1, 75.9) and (75.9, 63.7 adults/20. leaves) for the various treatments, respectively. However, the number of adults was gradually increased on the 5th and 7th day after zero time and amounted (28.0, 62.3), (22.0, 28.0), (20.0, 22.0), (18.0, 40.0) and (24.0, 26.0 adults/20 leaves) at the 7th day after application for the two application rates of various treatments, respectively. However, the number of adults was gradually increased on the 5th and 7th day after zero time and amounted (28.0, 62.3), (22.0, 28.0), (20.0, 22.0), (18.0, 40.0) and (24.0, 26.0 adults/20 leaves) at the 7th day after application for the two application rates of various compounds, respectively.

The average reduction rates of whitefly adult population after the investigation periods (residual effect) reached (67.8, 63.0), (81.5, 75.4), (84.8, 74.5), (84.8, 66.3) and (76.6, 66.7%) for

Table (1): Efficacy of certain pesticides on *Bemisia tabaci* (Genn.) adults infesting cucumber in greenhouse.

Treatments	Rate /100L	Mean number of adults per 20 leaves and reduction rates at indicated days post treatment					
		Pre-spray	1	Residual effect			Average
				3	5	7	
Biovar 4% WP	200 gm	40.0	20.0 (66.7)	16.0 (72.4)	24.0 (63.6)	28.0 (67.4)	22.0* (67.8)
	100 gm	42.0	32.0 (49.2)	20.0 (67.2)	28.0 (59.6)	34.0 (62.3)	28.5* (63.0)
Cord 72% EC	187.5cm^3	44.0	10.0 (84.8)	10.0 (84.3)	12.0 (83.5)	22.0 (76.7)	13.5* (81.5)
	93.7cm^3	42.0	14.0 (77.8)	12.0 (80.3)	16.0 (76.9)	28.0 (69.0)	17.5* (75.4)
Imaxi 35%EC	75cm^3	40.0	6.0 (90.0)	6.0 (89.7)	8.0 (87.9)	20.0 (76.7)	10.0* (84.8)
	37.5cm^3	40.0	10.0 (83.3)	12.0 (79.3)	20.0 (69.7)	22.0 (74.4)	16.0* (74.5)
Orizon 11%EC	50cm^3	38.0	6.0 (89.5)	6.0 (89.1)	8.0 (87.2)	18.0 (78.0)	9.5* (84.8)
	25cm^3	40.0	14.0 (76.7)	14.0 (75.9)	20.0 (69.7)	40.0 (53.5)	22.0* (66.3)
Achook 0.15% EC	187.5cm^3	40.0	16.0 (73.3)	14.0 (75.9)	12.0 (81.8)	24.0 (72.1)	16.5* (76.6)
	93.7cm^3	38.0	20.0 (64.9)	20.0 (63.7)	20.0 (68.1)	26.0 (68.2)	21.5* (66.7)
Control	--	40.0	60.0	58.0	66.0	86.0	67.5

L.S.D. = 12.7 *= Significant at 5% level.

% Reduction rates are given in brackets

Table (2): Efficacy of certain pesticides on *Bemisia tabaci* (Genn.) immature stages infesting cucumber in greenhouse.

Treatments	Rate /100L	Mean number of individuals per 20 leaves and reduction rates at indicated days post treatment					
		Pre-spray	1	Residual effect			Average
				3	5	7	
Biovar 4% WP	200 gm	92.0	28.0 (73.1)	22.0 (80.2)	28.0 (75.6)	32.0 (72.5)	27.5* (76.1)
	100 gm	102.0	40.0 (65.4)	36.0 (70.8)	40.0 (68.5)	46.0 (64.3)	40.5* (67.9)
Cord 72% EC	187.5cm^3	102.0	10.0 (91.3)	10.0 (91.9)	12.0 (90.6)	22.0 (82.9)	13.5* (88.5)
	93.7cm^3	112.0	30.0 (76.3)	24.0 (82.3)	40.0 (71.3)	48.0 (66.1)	35.5* (73.2)
Imaxi 35% EC	75cm^3	106.0	6.0 (95.0)	6.0 (95.3)	8.0 (93.9)	20.0 (85.1)	10.0* (91.4)
	37.5cm^3	104.0	20.0 (83.0)	22.0 (82.5)	40.0 (69.1)	50.0 (62.0)	33.0* (71.2)
Orizon 11%EC	50cm^3	100.0	6.0 (94.7)	8.0 (93.4)	12.0 (90.4)	20.0 (84.2)	11.5* (89.3)
	25cm^3	84.0	20.0 (79.0)	20.0 (80.3)	26.0 (75.1)	40.0 (62.3)	26.5* (72.6)
Achook 0.15% EC	187.5cm^3	100.0	24.0 (78.8)	20.0 (83.4)	30.0 (75.9)	40.0 (68.4)	28.5* (75.9)
	93.7cm^3	102.0	40.0 (65.4)	40.0 (67.5)	42.0 (66.9)	50.0 (61.2)	43.0* (65.2)
Control	--	106.0	120.0	128.0	132.0	134.0	128.5

L.S.D. = 13.5 *= Significant at 5% level.

% Reduction rates are given in brackets.

the bioinsecticide, *B. bassiana* (Biovar 4% WP), the organophosphorus compound, Profenofos (Cord 72%EC), Imidacloprid (Imaxi 35% SC), cetamiprid+ Abamectin (Orizon 11% EC) and the botanical insecticides, Azadirachtin 1500 ppm (Achook .15%) respectively. It was found that, values of the general mean of adults showed significant reduction for the various treatments at the two application rates as compared with the control.

Statistically, the data revealed no significant differences in the general means of adults between RC and ½RC of the above treatments with Biovar, Cord, Imaxi Orizon and Achook. The previous results indicated that the efficacy of the tested compounds could be arranged in descending order as follows: Imaxi, Orizon, Achook and Bivar which was the least effective one.

2.1.2. Whitefly immature stages:

Obtained data concerning immature stages, revealed that the tested compounds were more effective on the whitefly immature stages than adults. The average numbers of immature stages per 20 leaves were obviously reduced during the following 3 days after treatment for Biovar, Cord, Imaxi, Orizon and Achook. Conversely and the numbers of immature stages were gradually increased during the 5th and 7th day after zero time (Table 2).

The average reduction rates (residual effect) of the immature stages populations recorded (76.1, 67.9), (88.5, 73.2), (91.4, 71.2), (89.3, 72.6) and (75.9, 65.2%) for the two application rates (RC & ½RC) of various compounds, respectively. Values of the general mean of immature stages were significantly lower for all treatments than control. Statistically, the data revealed significant differences in the general means of whitefly immature stages between RC and ½RC of the above treatments under study except for the Biovar treatment which did not reveal a significant difference.

In conclusion, the Neonicotinoid compound, Imaxi was the most effective insecticide against the whitefly, *B. tabaci* stages followed by Orizon, Cord, Achook and the bioinsecticide, Biovar.

3- Efficacy of combining *Beauveria bassiana* with certain pesticides:

The effects of mixtures of the bioinsecticide, Biovar 4% WP at (recommended, 200 and half recommended concentration, 100 gm/100 L.) plus Cord (Profenofos) 72% at its RC & ½RC (187.5 & 93.7 cm^3/100L.), Imaxi 35%EC at its RC & ½RC

Table (3): Effect of various combinations of pesticides on *Bemisia tabaci* (Genn.) adults infesting cucumber in greenhouse.

Treatments	Rate /100L	Mean number of adults per 20 leaves and reduction rates at indicated days post treatment					
		Pre-spray	1	Residual effect			Average
				3	5	5	
Biovar 4% WP + Cord 72% EC	200gm +187.5cm^3	42.0	2.0 (96.8)	8.0 (86.9)	12.0 (82.7)	20.0 (81.0)	10.5* (83.5)
	200gm +93.7cm^3	40.0	8.0 (86.7)	16.0 (72.4)	20.0 (69.7)	28.0 (72.0)	18.0* (71.4)
	100 gm +93.7cm^3	44.0	8.0 (87.9)	14.0 (78.1)	20.0 (72.5)	32.0 (70.9)	18.5* (73.8)
Biovar 4% WP + Imaxi 35%EC	200 gm +75cm^3	38.0	0.0 (100.0)	4.0 (92.7)	8.0 (87.2)	16.0 (83.2)	7.0* (87.7)
	200 gm +37.5cm^3	40.0	0.0 (100.0)	8.0 (86.2)	12.0 (81.8)	20.0 (80.0)	10.0* (82.7)
	100 gm +37.5cm^3	42.0	2.0 (96.8)	10.0 (83.6)	16.0 (76.9)	30.0 (71.4)	14.5* (77.3)
Biovar 4% WP + Orizon 11%EC	200 gm +50cm^3	38.0	10.0 (82.5)	8.0 (85.5)	10.0 (84.1)	26.0 (72.6)	13.5* (80.7)
	200 gm +25cm^3	44.0	10.0 (84.8)	2.0 (96.9)	6.0 (91.7)	24.0 (78.2)	10.5* (88.9)
	100 gm +25cm^3	40.0	10.0 (83.3)	6.0 (89.7)	8.0 (87.9)	22.0 (78.0)	11.5* (85.2)
Biovar 4% WP + Achook 0.15%EC	200 gm +187.5cm^3	40.0	10.0 (83.3)	6.0 (89.7)	10.0 (84.8)	20.0 (80.0)	11.5* (84.8)
	200gm +93.7cm^3	36.0	8.0 (85.2)	16.0 (69.3)	20.0 (66.3)	12.0 (86.7)	14.0* (74.1)
	100gm +93.7cm^3	42.0	10.0 (84.1)	8.0 (86.9)	12.0 (82.7)	20.0 (81.0)	12.5* (83.5)
Control	--	40.0	60.0	58.0	66.0	100.0	71.0

L.S.D. = 14.1 *= Significant at 5% level.
% Reduction rates are given in brackets.

Table (4): Effect of various combinations of pesticides on *Bemisia tabaci* (Genn.) immature stages infesting cucumber in greenhouse.

Treatments	Rate /100L	Mean number of individuals per 20 leaves and reduction rates at indicated days post treatment					
		Pre-spray	1	Residual effect			Average
				3	5	7	
Biovar 4% WP + Cord 72% EC	200gm +187.5cm^3	90.0	36.0 (72.7)	2.0 (98.6)	10.0 (93.1)	18.0 (87.5)	16.5* (93.1)
	200gm +93.7cm^3	100.0	40.0 (72.7)	16.0 (89.9)	20.0 (87.5)	24.0 (85.0)	25.0* (87.5)
	100 gm +93.7cm^3	102.0	42.0 (71.9)	20.0 (87.6)	30.0 (81.7)	40.0 (75.6)	33.0* (81.6)
Biovar 4% WP + Imaxi 35%EC	200 gm +75cm^3	120.0	40.0 (77.2)	0.0 (100.0)	6.0 (96.9)	18.0 (90.7)	16.0* (95.8)
	200 gm +37.5cm^3	106.0	40.0 (74.2)	4.0 (97.6)	10.0 (94.1)	20.0 (88.2)	18.5* (93.3)
	100 gm +37.5cm^3	104.0	40.0 (73.7)	28.0 (83.0)	30.0 (82.0)	36.0 (78.4)	33.5* (81.1)
Biovar 4% WP + Orizon 11%EC	200 gm +50cm^3	80.0	28.0 (76.1)	24.0 (81.0)	26.0 (79.7)	28.0 (78.2)	26.5* (79.7)
	200 gm +25cm^3	82.0	32.0 (73.4)	24.0 (81.5)	26.0 (80.2)	26.0 (80.2)	27.0* (80.7)
	100 gm +25cm^3	100.0	32.0 (78.2)	24.0 (84.8)	26.0 (83.8)	24.0 (85.0)	26.5* (84.6)
Biovar 4% WP + Achook 0.15% EC	200 gm +187.5cm^3	102.0	48.0 (67.9)	44.0 (72.7)	44.0 (73.1)	54.0 (67.0)	47.5* (70.9)
	200gm +93.7cm^3	104.0	48.0 (68.5)	48.0 (70.8)	46.0 (72.4)	56.0 (66.4)	49.5* (69.9)
	100gm +93.7cm^3	100.0	48.0 (67.2)	26.0 (83.6)	30.0 (81.3)	32.0 (80.1)	34.0* (81.6)
Control	--	86.0	126.0	136.0	138.0	138.0	134.5

L.S.D. = 14.3 *= Significant at 5% level.
% Reduction rates are given in brackets.

(75 & 35.5 cm^3/100L.), Orizon 11%EC at its RC & ½RC (50 & 25 cm^3/100L.) and Achook 0.15%EC at its RC & ½RC (187.5 & 93.7) against the whitefly stages are presented in (Tables 3 & 4).

Results of the whitefly adults and immature stages (except eggs), revealed that the average numbers of *B. tabaci* decreased clearly during the following 3 days after spraying as compared with control and amounted (8.0, 16.0, 14.0), (4.0, 8.0, 10), (8.0, 2.0, 6.0) and (6.0, 16.0, 8.0 adults/20 leaves) for adults and (2.0, 16.0, 20), (0.0, 4.0, 28.0), (24.0, 24.0, 24.0) and (44.0, 48.0 and 26.0 individuals/20 leaves) for immature stages on the 3rd day for the mixtures of Biovar plus Cord, Imaxi, Orizon and Achook, respectively. Thereafter, the values of the average numbers of *B. tabaci* adults and immature stages increased gradually from the 5th days after spraying for the various combinations, respectively.

The average reduction rates of *B. tabaci* population during the period of investigation reached (83.5, 71.4, 73.8), (87.7, 82.7, 77.3), (80.7, 88.9, 85.2) and (84.8, 74.1, 83.5%) for adults and (93.1, 87.5, 81.6), (95.8, 93.3, 81.1), (79.7, 80.7, 84.6) and (70.9, 69.9, 81.6%) for immature stages for the various combinations, respectively. Values of the general mean of *B. tabaci* showed significant decreases in all combinations as compared with the control. In addition, there were insignificant differences between the general mean of *B. tabaci* adults for the mixtures; Biovar plus Cord, Imaxi, Orizon and Achook at (RC + RC), (RC + ½RC) and (½RC + ½RC) for each combination separately. Regarding the immature stages, there were no significance differences in the general means for the combinations of Bivar plus Cord and Biovar plus Imaxi at (RC + RC) and (RC + ½RC) and there was a significant difference at (RC + RC) and (½RC + ½RC). Also, the three combinations (RC + RC), (RC + ½RC) and (½RC + ½RC) of Biovar plus Orizon and Achook did not match the significance level in the general mean of *B. tabaci* immature stages.

Results obtained revealed that, the combination of Biovar plus Orizon was the most effective mixture against *B. tabaci* adult population followed by Biovar plus Imaxi, Biovar plus Achook and Biovar plus Cord. Whereas, efficacy of the tested mixtures could be arranged in descending order as follow: Biovar plus Imaxi, Biovar plus Cord, Orizon and plus Achook.

These results are in agreement with the findings of Mayoral *et al.* (2006) who stated that the efficacy of Naturalis alone at 200 ml/hl was lower than that of the chemical Imidacloprid-based reference product of the bioinsecticide applied at the same concentrate in tank mixture with an adjuvant, and of the biocontrol agent alone at 300 ml/hl. However, when the product was applied at 200 ml/hl in tank mixture with the adjuvant and alone at 300 ml/hl, its efficacy was always comparable to that of the chemical standard. Also, Oliveira *et al.* (2003) stated that, the use of Alpha-Cypermethrin and Thiamethoxam formulations in coffee IPM programs for a *B. bassiana* inoculum conservation strategy are recommended, since these products were compatible with the entomopathogenic fungus *B. bassiana* (CG 425), an important natural control agent of the coffee berry borer, *Hypothenemus hampei*.

Co-toxicity resulted from addition of Biovar to Cord, Imaxi, Orizon and Achook against *Bemisia tabaci* adults and immature stages are presented in (Tables 5, 6, 7 and 8),

Results in (Table 5) show that the type of joint action obtained from the addition of the bioinsecticide at its recommended RC and half recommended rate ½RC to Cord at the two application rates varied and induced an irregular joint action against *B. tabaci*, indicating a dominated antagonistic effect against adults and additive effect against immature stages. At the same time, results obtained from addition of Biovar at RC & ½RC to Imaxi and to Orizon at their two application rates, showed regular joint action where, the additive effect for the total mortality of the combination was higher than the mortality of summation of each compound separately, thus, the joint action of the two compounds in the mixtures was more effective against the tested insect than the compound alone. Also, an irregular joint action obtained from addition of Biovar at RC to Achook at its two rates indicated a dominated antagonistic effect; conversely the addition of Biovar at ½RC to Achook at its ½RC indicated an additive joint action (Table 6 & 7).

There were also antagonistic pairs as well as other pairs producing only additive effect when jointly applied. In addition, co-toxicity resulted from addition of the LC_{50} of Malathion (145 ppm) to Neemazal at (50 – 4000 ppm) for *Rhizopertha dominica* showed an additive effect at all tested concentrations. In case of mixtures of Sumithion

Table (5): Co-toxicity resulted from addition of Biovar at 200 and 100gm/100L to Cord at two rates for controlling *Bemisia tabaci* (Genn.).

Application rate (cm^3/100 L.)	% Mortality after 3 days post treatment						Co-toxicity factor		Type of joint action	
	Biovar alone		Cord alone		Biovar+ Cord					
	Expected mortality				*Observed mortality*					
	A	I	A	I	A	I	A	I	A	I
At 200gm/100L										
187.5	72.4	73.1	84.3	91.9	86.9	98.6	-13.1	-1.4	d	d
93.7			80.3	82.3	72.4	89.9	-27.6	-10.1	a	d
At 100gm/100L.										
93.7	67.2	70.8	80.3	82.3	78.1	87.6	-21.9	-12.4	a	d

Table (6): Co-toxicity resulted from addition of Biovar at 200 and 100gm/100L to Imaxi at two rates for controlling *Bemisia tabaci* (Genn.).

Application rate (cm^3/100 L.)	% Mortality after 3 days post treatment						Co-toxicity factor		Type of joint action	
	Biovar alone		Imaxi alone		Biovar + Imaxi					
	Expected mortality				*Observed mortality*					
	A	I	A	I	A	I	A	I	A	I
At 200gm/100L										
75	72.4	73.1	89.7	95.3	92.7	100.0	-7.3	0.0	d	d
37.5			79.3	82.5	86.2	97.6	-13.8	-2.4	d	d
At 100gm/100L.										
37.5	67.2	70.8	79.3	82.5	83.6	83.0	-16.4	-17.0	d	d

a: antagonistic effect (-20 or more)
d: additive effect (between –20 & +20)
A: adults I: immature stages

Table (7): Co-toxicity resulted from addition of Biovar at 200 and 100gm/100L to Orizon at two rates for controlling *Bemisia tabaci* (Genn.).

Application rate (cm^3/100 L.)	% Mortality after 3 days post treatment						Co-toxicity factor		Type of joint action	
	Biovar alone		Orizon alone		Biovar + Orizon					
	Expected mortality				*Observed mortality*					
	A	I	A	I	A	I	A	I	A	I
At 200gm/100L										
75	72.4	73.1	89.1	93.4	85.5	81.0	-14.5	-19.0	d	d
37.5			75.9	80.3	96.9	81.5	-3.1	-18.5	d	d
At 100gm/100L.										
37.5	67.2	70.8	75.9	80.3	89.7	84.8	-10.3	-15.2	d	d

Table (7): Co-toxicity resulted from addition of Biovar at 200 and 100gm/100L to Orizon at two rates for controlling *Bemisia tabaci* (Genn.).

Application rate (cm^3/100 L.)	% Mortality after 3 days post treatment						Co-toxicity factor		Type of joint action	
	Biovar alone		Orizon alone		Biovar + Orizon					
	Expected mortality				*Observed mortality*					
	A	I	A	I	A	I	A	I	A	I
At 200gm/100L										
75	72.4	73.1	89.1	93.4	85.5	81.0	-14.5	-19.0	d	d
37.5			75.9	80.3	96.9	81.5	-3.1	-18.5	d	d
At 100gm/100L.										
37.5	67.2	70.8	75.9	80.3	89.7	84.8	-10.3	-15.2	d	d

Table (8): Co-toxicity resulted from addition of Biovar at 200 and 100gm/100L to Achook at two rates for controlling *Bemisia tabaci* (Genn.).

Application rate (cm^3/100 L.)	% Mortality after 3 days post treatment						Co-toxicity factor		Type of j oint action	
	Biovar alone		Achook alone		Biovar+ Achook					
	Expected mortality				*Observed mortality*					
	A	I	A	I	A	I	A	I	A	I
At 200gm/100L										
187.5	72.4	73.1	75.9	83.4	89.7	72.7	-10.3	-27.3	d	a
93.7			63.7	67.5	69.3	70.8	-30.7	-29.2	a	a
At 100gm/100L.										
93.7	67.2	70.8	63.7	67.5	86.9	83.6	-13.1	-16.4	d	d

a: antagonistic effect (-20 or more)
d: additive effect (between –20 & +20)
A: adults   I: immature stages

plus Neemazal, an additive effect was obtained at higher concentrations (500, 1000 ppm) but at 50, 100 and 250 ppm an antagonistic effect was obtained, (El-Lakwah, 1997).

Furlong and Groden (2001) stated that no synergistic interaction was detected when larvae of *Spodoptera littoralis* were fed on leaf discs treated with sublethal concentrates of Imidacloprid 24 h after application of *B. bassiana* conidia to larvae. However, a synergistic interaction was detected when larvae were fed on leaf discs treated with Imidacloprid and sprayed with *B. bassiana* conidia 24 h later. Although sublethal concentrates of both Imidacloprid and the Triazine insect growth regulator (IGR) Cyromazine prolonged the duration of the second instar, only Imidacloprid interacted with *B. bassiana* to produce a synergistic response in larval mortality. Additionally, Shalaby (2004) stated that, results obtained from addition of botanical compound, Achook at (200 and 100 cm^3/100L.) to the IGR, Admiral at its two application rates (300 cm^3 & 150 cm^3) showed an irregular joint action where, induced an additive effect only with Achook at high application rate 200 cm^2/100L against *B. tabaci* stages.

Generally, the bioinsecticide, Biovar in combination of the certain insecticides (Imaxi, Orizon and Achook) at their half recommended application rates could be successfully used for controlling the whitefly as it reduces the used rates of insecticides and consequently less hazards and problems to the environment.

REFERENCES

Anderson, T.E. and Roberts, D.W. 1983. Compatibility of *Beauveria bassiana* isolates with insecticide formulations used in Colorado potato beetle (Coleoptera: Chrysomelidae) control. J. Econ. Entomol., 76: 1437-1441.

El-Lakwah, F. A. M. 1997. Toxicity of the botanical insecticide, Neemazal-W (powder contains 10% Azadirachtin) alone and in mixtures with two insecticides against the lesser grain borer, *Rhizopertha dominica* (F.). Annals of Agric. Sci., Moshtohor, 35(4): 2583-2594.

Faria, M. and Wraight, S.P. 2001. Biological control of *Bemisia tabaci* with fungi. Crop production. 20 (9): 767-778

Fedorinchik,N.S. 1974. Biological control method for plant protection, pp. 163-307. In: E.H.Chunsakova, G.V. Gusev, N.S. Fedorinchik (Eds): Moscow, Kolos (Russian).

Furlong, M. J. and Groden, E. 2001. Evaluation of synergistic interactions between the Colorado potato beetle (Coleoptera: Chrysomelidae), the pathogen *Beauveria bassiana* and the insecticides, Imidacloprid, and Cyromazine. J. Econ. Entomol., 94(2): 344-356.

Hinderson, C. F. and Tilton, E. W. 1955. Test with acaricides against the brown white mite. J. Econ. Entomol., 48: 157-161.

Ibrahim, A.A. 2006. Action of certain microbes on cotton leaf worm. Ph.D. Thesis, Faculty of Science, Al-Azhar University, 171 pp.

Mansour, N. A.; El-Defrawi, M. E.; Toppozada, A. and Zeid, M. 1966. Toxicological studies on the Egyptian cotton leafworm, *Prodenia litura.* VI. Potential and antagonism of organophosphorus and carbamate insecticides. J. Econ. Entomol., 59(2): 307-311.

Marin, M.P., Bustillo, P.A.E. and Rivera, M.A. 2000. Compatibility in vitro and effect of mixtures of three insecticides in the viability of *Beauveria bassiana.* Revista Colombiana de Entomologia, 26 (3/4): 125-130.

Mayoral, F; Benuzzi, M. and Ladurner, E. 2006. Efficacy of the *Beauveria bassiana* strain ATCC 74040 (NaturalisReg.) against whiteflies on protected crops. Bulletin-OILB/SROP. 2006;

29(4): 83-88.

Oliveira, C. N.; Neves, P. M. O. J. and Kawazoe, L. S. 2003. Compatibility between the entomopathogenic fungus *Beauveria bassiana* and insecticides used in coffee plantations. Scientia-Agricola. 2003; 60(4): 663-667.

Puzari, K.C., Hazarika, L.K., Dutta, P. and Das, P. 2006. In vitro inhibition of *Beauveria bassiana* (Bals.) Vuill. growth by different commonly used insecticides in rice. J. Biological Control, 20 (1): 51-56.

Shalaby, H. H. 2004. Studies on the efficiency of some new pest control measures against certain pests of common bean. Ph.D. Thesis, Faculty of Agric., Zagazig Univ. 334 pp.

Snedecor, G. W. and Cochran, W. G. 1967. Statistical Methods, Sixth edition, Iowa State College Press, Ames. Iowa, USA.

Vilas-Boas, A.M. and Alves, S.B. 1989. Pathogenicity of *Beauveria* spp. and its effects of association with Monocrotophos on *Castnia licus* (Drury, 1970) (Lepidoptera: Castinidae). Annals Society of Entomology of Brazil,17: 305-332.

Yue, L.Z. and Wen, Z.Q. 2005. Relative virulence of isolates of *Beauveria bassiana* to the potato tuber moth (*Phthorimaea operculella*) and their biological compatibility with ten insecticides. Plant Protection. 31 (3): 57-61.