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Effect of Two Bioprepartions of *Bacillus thuring*ensis upon the Potato Tuber Moth (PTM), *Phthorimaea operculella* (Zeller) (Lepidoptera: Gelechiidae)

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ABSTRACT

The present investigation was carried out to evaluate the efficiency of two biopreparations of *Bacillus thuringensis* (MYX_{104} and MYX_{833}) on certain biological parameters of the potato tuber moth *Phthorimaea operculella* (Zeller) under laboratory conditions.

The results cleared that the tested biopreparation had a delayed effect on the fitness components of the insect. In general, all the treatments achieved a significant shortened moth longevity of both sexes, prolongation of larval and pupal durations, severe reduction in the number of resulted pupae, great increase of the rate of malformed pupae, and sharp decrease of the rate of emerged moths according to the tested concentration of each biopreparation. Moreover, the consequently reared generations after parent's one treatment that indicated so lowered viability of developing individuals and decreased numbers of emerged unviable weak and sterile moths followed by final termination of the life cycle, which revealed as distinct failure of (F_3) development due to the former treatment of (P) one with the lowest tested concentration of each of MYX_{104} & MYX_{833} (25 ppm). That failure could be attributed to the cumulative effect of induced recessive lethal genes in both influenced sexes along the extended period of the following generations post- treatment of parents one, causing apparent drastic effects, which appeared at the beginning of (F_3). The results also proved that MYX_{104} was more effective than MYX_{833} on the investigated parameters of the fitness component of the potato tuber moth.

INTRODUCTION

Potato, *Solanum tuberosum* L. (Solanaceae) is widely produced food crop worldwide. In Egypt, potato ranks the second crop after cotton as a cash crop. Potato plants and tubers are attractive to many pests. The potato tuber moth (PTM), *Phthorimaea operculella* (Zeller) (Lepidoptera: Gelechiidae) is a main pest of stored potato tubers and infests potatoes outdoors in the worm climates. It is considered to be the most common serious insect- pest of potato plants and other solanaceous crops (Arnone *et al*., 1998; Soeriaatmadja, 1988; Lal, 1990 and Nasseh and Al-Furassy, 1992). The insect is considered as a serious pest of potato tubers in storage and the storage losses vary from 40 to 80% depending on stores condition, potato cultivar and locality (Zaghloul *et al*., 1992); losses can be 100% within 90 days.

The use of chemical insecticides for controlling this insect- pest is undesirable hence, the idea of integrated pest management (IPM) is not a new one; and accepted as being as a control strategy in which a variety of biological, chemical, genetic and cultural control measures are combined to give stable long term pest control.

IPM is the planned integration of a range of techniques to minimize pesticides effects on the environment. Therefore, the beneficial microorganisms as biological control agents have been used for controlling many injurious economic insect-pests (Mesbah, 1985, Mesbah *et al*., 1990 a, b, 1994 and Omer *et al*., 1992). Biological pesticides based on the soil microbe, *B. thuringensis* Berliner, are becoming increasing by important in pest management. *B. thuringensis* is highly toxic to target pests, but is harmless to human, most beneficial insects and other nontarget organisms (Federici, 1999)

Bacillus thuringensis (B.t) was highly effective against the pest when applied at 10-25 °C. Treatment of larvae with preparations of *B.t* at 9-12°C showed that they were as active as chemical insecticides (Baklanova *et al*., 1990).

Efficiency of mixture of *B. thuringensi*s and granulosis virus and extra irrigation for integrated control of *P. operculella* was shown to be equally, and

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in some cases, more efficient than conventional insecticides (Salah *et al*., 1994).

Therefore, the present work has been conducted to evaluate the efficiency of two biopreparations of *B. thuringensis*, MYX_{104} and MYX_{833} upon the potato tuber moth (PTM), *P. operculella*; to attain new promisable results that can be added and the useful towards the control of potato tuber moths.

MATERIALS AND METHODS

Stock culture of the potato tuber moth:

Infested potato tubers were collected and kept with clean tubers in cubic foot cages. On its floor a thin layer of saw dust (3 cm thickness) was spread. Cages were covered with muslin. Rearing of the insect, *P. operculella* was successfully performed for several generations under laboratory higrothermic conditions of 24± 2 ^{o}C and 70 ± 5 % R.H. according to Assem (1966).

The used materials:

Bio-preparation of *Bacillus thuringiensis*. MYX_{104} and MYX_{833}

MYX_{104}: Active ingredient is CrylAc delta endotoxin of *B. thuringiensis* kurstaki encapsulated in Killed *Psuedomonas fluorescens* (15 %) and inert ingredients (85%), Mycogen corporation EPA Est. No. 53219-CA – 01-5501 oberlin Drive Net contents: 1 quart San Diego, CA 92121.

The MYX_{833} is containing *B. thuringiensis* aizawi Cry 1 Ca biopreparations of *B.t.* (MYX_{104} and MYX_{833}) were kindly obtained from Proffessor Dr.Walid Elyassaki, Ain-shams University, Fac. of Science, Entomology Dept, Cairo, Egypt.

Procedure of laboratory bioassay of MYX_{833} and MYX_{104} against PTM

Series laboratory bioassays were conducted to test both the tested biopreparations of *B. thuringiensis* (MYX_{833} and MYX_{104}) against the pest. The bioassay tests employed four progressive concentrations of each bio-preparation i.e., 25, 50, 100 and 500 ppm. Treatments were done in 3 replicates, a fixed weight of potato tubers (500 g) was placed in 1 litre jar, after had been dipped in each prepared progressive dilution for 1 minute and left till complete dryness. Later, two couples of the emerging adults (2♀+ 2♂) of stock culture were released in the jar, and provided with a hanged piece of soaked cotton pad in 5% sucrose solution.

The revealed effects of the tested progressive concentrations of each evaluated biopreparation on the fitness components of the treated parents generation and the following ones, were observed and recorded.

RESULTS AND DISCUSSION

Effect of certain tested biopreparations of *B. thuringensis* (MYX_{104} and MYX_{833}) on the rate of emergening of *Phthorimaea operculella* moths in consequent generations post parents treatment.

In general, both of the evaluated *B. thuringensis* biopreparations-MYX_{104} and MYX_{833} upon *P. operculella* (Zeller.) showed the direct and/or the delayed effects on the fitness components of the treated parents generation and the consequently raised offspring from F_1 to F_2, compared to the untreated control which could concisely explained by the decreased periods of moths longevity; increased durations of developing larvae, the reduced number of developed normal pupae, versus the increased number of deformed ones, and the sharp reduced rate of incipient moths of both sexes.

Effect of the tested concentrations of *B. thuringensis* biopreparation- MYX104:

Data recorded in Table 1 show the effect of the tested dilutions of *B. thuringensis* biopreparation-MYX_{104} on the rate of the resulted pupae and emerged moths from the exposed immatures of parent's generation to the treated tubers. The results showed the reduction of the resulted pupae according to the effect of the used concentrations. The high collected number of resulted pupae from the untreated control ranged from 55 to 75 with a mean number of 65.33 ± 5.78 pupae. That number was greatly decreased as MYX_{104} concentrations increased from 25 ppm (17.66 ± 2.40 pupae) up to 500 ppm (8.33 ± 0.88 pupae).

Concerning the dose effect of MYX_{104} biopreparation on the rates of moths emergence, the compartive highest average number of emerged moths (61.67 ± 5.24), in untreated control was faced with lowered rates in the different performed treatments of MYX_{104}. The increase of MYX_{104} concentration gradually decreased the mean numbers of the emerged moths from 15.33 ± 2.33 at 25 ppm to 4.67 ± 1.76 at 500 ppm. In general, the differences between the mean numbers of emergenced moths were highly significant compared with the control.

Moreover, the incipient moths post parents (P) treatment by tested MYX_{104} concentrations or/and control one were daily collected, sexed and separated. Later, these resulted moths of each treatment were allowed to copulate together on clean untreated potato tubers and each of three couples treatment was observed. The fitness components of resulting (F_1) progeny were inspected. The included results shown in Table 1 explain that the numbers of resulted (F_1) pupae

Table 1. Effect of certain evaluated concentrions of *Bacillus thuringiensis* biopreparation –MYX$_{104}$ on the fitness components of parents (P), F_1, F_2 and F_3 generations of *Phthorimaea operculella*

Concentration (ppm)	P			F1			F2			F3		
	No. of resulted Pupae	Moths emergence		No. of resulted pupae	Moths emergence		No. of resulted pupae	Moths emergence		No. of resulted pupae	Moths emergence	
	$\bar{x} \pm s\bar{x}$	No. $\bar{x} \pm s\bar{x}$	% $\bar{x} \pm s\bar{x}$	$\bar{x} \pm s\bar{x}$	No. $\bar{x} \pm s\bar{x}$	% $\bar{x} \pm s\bar{x}$	No. $\bar{x} \pm s\bar{x}$	No. $\bar{x} \pm s\bar{x}$	% $\bar{x} \pm s\bar{x}$	No. $\bar{x} \pm s\bar{x}$	No. $\bar{x} \pm s\bar{x}$	% $\bar{x} \pm s\bar{x}$
25	17.66±2.40 (13-21)***	15.33±2.33 (11-19)	86.44±2.03 (48.2190.48)	13.33±1.67 (10-15)	9.3±0.88 (8-11)	72.22±10.60 (53.33-90)	10.66±1.20 (9-13)	6.67±0.88 (5-8)	49.52±13.90 (23-70)	Nil	Nil	Nil
50	15.0±1.73 (12-18)	12.33±1.45 10-15	82.22±1.11 (80.0±83.33)	7.33±0.88 6-9	5.33±0.88 (4-7)	71.96±3.22 (66.67-77.78)	3.0±1.16 (1-5)	0.58±0.33 (0-1)	6.67±6.67 (0-20)	-	-	-
100	9.0±2.08 (6-13)	6.33±1.45 (4-9)	70.3±2.46 (66.67-75)	0.66±0.33 (0-1)	Nil	Nil	-	-	-	-	-	-
500	8.33±0.88 (7-10)	4.67±1.76 (2-8)	52.86±14.92 (28.57-80)	Nil	Nil	Nil	-	-	-	-	-	-
Control	65.33±5.78 (55-75)	61.67±5.24 (52-70)	94.44±0.62 (93.33-95.4)	62.33±6.23 (50-70)	57.66±5.46 (47-65)	92.64±0.86 (91.05-94.0)	62.23±6.69 (50-73)	59.67±6.07 (49-70)	95.88±1.2 (93.75-98.0)	59.67±6.74 (47-70)	56.33±6.12 (45-66)	94.53±0.64 (93.5595.74)
LSD	9.70	8.94	21.57	10.62	11.18	22.20	13.78	12.26	30.90	-	-	-
F(Cal.)	60.62**	69.57**	5.59*	74.37**	81.24**	3.42	65.70**	84.55**	24.97**	-	-	-

$\bar{x}$ = Mean number and s $\bar{x}$ = Standard devisition

P= Parents , F1= First generation , F2= Second generation and F3= Third generation

*= Significant **= highly significant and ***= Average

were greatly reduced than those of parents generation; according to the different conducted MYX_{104} treatments.

The mean numbers of resulted F1 pupae ranged between 13.33 ± 1.67 for the concentration of 25 ppm and 0.00 for 500 ppm in comparison to 62.33 ± 6.23 in untreated control, versus 17.66 ± 2.40 and 8.33 ± 0.88 for the same concentrations respectively in (P) generation.

Sequently, the highest number of emerged moths (57.66 ± 5.46) was observed for the untreated control, versus the lowered number of emerged ones in the formerly running treatments. It was found that the, treatment with MYX_{104} at concentrations of 100 & 500 ppm, completely inhibited the reproductive potential of both sexes of first generation (F1) even at comparison to the lower tested concentrations of 25 and 50 ppm (Table 1).

Moreover, the few number of 5.33 ± 0.88 of the emerged moths after treatment with the lower concentration of 50 ppm was unviable, weak and sterile and most of them died before the completion of the F_2 progeny. Generally, MYX_{104} gave highly significant effect on the studied biological characters of that insect – pest. Later, the fewer numbers of the emerged moths (6.67 ± 6.67) resulted from the lowest concentration treatment (25 ppm) and untreated control (59.67 ± 6.07) of 2nd generation were collected, sexed and separated. Then, they were paired and released on the clean tubers in adopted treatments of 3rd generation. Three couples were also used per treatment for studying the delayed effects on the fitness components of their progeny and the finally resulted alive pupae and emerged moths.

The utmost reduced numbers of the resulted (F_2) pupae according to MYX_{104} treatment of (P) were averaged from 10.66 ± 1.20 to 3.0 ± 1.16 pupae at 25 ppm up to 50 ppm; respectively. In check untreated the mean number of the resulted pupae was 62.23 ± 6.69 (Table 1).

Consequently, that delayed effect of MYX_{104} was reflected on the decreased rates of (F_2) emerged moths, in comparison to control treatment, which gave the highest mean number of emerged moths (59.67 ± 6.07).Moreover, that higher dose of MYX_{104} (50ppm), decreased the mean number of the emerged moths of the second generation (0.58 ± 0.33) (Table 1) compared with P & F_1 generations. The results also showed high significant effect of this dose on most of the studied biological characters whereas the incipient moths were inactive and sterile.

Also, it is worth to show the final failure of (F_3) development even at the lowest initiated treatmental concentration of MYX_{104} (25ppm). That could be attributed to the cumulative effect of induced recessive lethal genes in both influenced sexes along the following developing generations of (P), (F_1) and (F_2); causing unprofitable effects that appeared at the beginning of 3rd generation (Table 1).

Effect of the tested concentrations of *B. thuringiensis* biopreparation– MYX_{833}

Effect on parents generation (P):

Identical to the above-cited results of MYX_{104} biopreparation the included results in Table 2, explain similar effects of the tested dilutions of MYX_{833} biopreparation on the fitness components due to exposing the of parents generation to tubers treated with different concentrations of MYX_{104}.

From Table 2 the results indicated that larval feeding on tubers treated with MYX_{833} reduced the number of resulted pupae according to the used concentrations. The number of resulted pupae from untreated control ranged from 55 to 75 with a mean of 65.33± 5.78 pupae , while that number was greatly reduced due to the increase of the tested MYX_{833} concentrations from 25 ppm (40.33± 3.18 pupae) to 500 ppm (13.67 ±1.20 pupae).

Concerning the dose effect of MYX_{833} on the rates of moths incipiency, control treatment gave the highest mean number of the emerged moths (61.67 ±5.24) followed by lower rates of moths emergence in the other initiated MYX_{833} treatments. Whereas, the increase of MYX_{833} concentration gradually decreased the mean numbers of emerged moths from 31.67 ±3.18 at 25ppm to 7.33± 0.88 at 500 ppm. In general, the calculated rates of moths emergence were highly significantly differed from control. (Table 2).

1st *generation* (F_1):

The data in Table 2 declare the greatly reduced numbers of resulted pupae than that of parents generation; according to the different conducted MYX_{833} treatments.

The mean numbers of the resulted pupae were ranged between 31.0±2.65 and 9.33±0.88 for 25ppm and 500 ppm treatmental concentrations in comparison to 62.33±6.3 in untreated control (Table 2), versus 40.33±3.18; 13.67±1.20 and 65.33±5.78, respectively in (P) generation (Table 2).

Comparatively, the highest mean of the emerged moths (57.66±5.46) was observed for the untreated control, versus the lower means of emerged ones in the formerly run (P) treatments. Herein, at the

Table 2. Effect of evaluated concentration of *Bacillus thuringiensis* biopreparation –MYX$_{833}$ on the fitness components of parent P, F$_1$, F$_2$ and F$_3$ generations of *Phthorimaea operculella*

Concentration (ppm)	P			F_1			F_2			F_3		
	No. of resulted pupae	Moths emergence		No. of resulted pupae	Moths emergence		No. of resulted pupae	Moths emergence		No. of resulted pupae	Moths emergence	
	$\bar{x} \pm s\bar{x}$	No. $\bar{x} \pm s\bar{x}$	% $\bar{x} \pm s\bar{x}$	$\bar{x} \pm s\bar{x}$	No. $\bar{x} \pm s\bar{x}$	% $\bar{x} \pm s\bar{x}$	$\bar{x} \pm s\bar{x}$	No. $\bar{x} \pm s\bar{x}$	% $\bar{x} \pm s\bar{x}$	$\bar{x} \pm s\bar{x}$	No. $\bar{x} \pm s\bar{x}$	% $\bar{x} \pm s\bar{x}$
25	40.33±3.18 (35-46)***	31.67±3.18 (28-38)	78.49±4.25 (70-82.86)	31.0±2.65 (27-36)	25.0±1.73 (22-28)	80.86±1.63 (77.78-83.33)	18.0±1.53 (15-20)	5.0±0.58 (4-6)	27.66±1.17 (26.3-30)	Nil	Nil	Nil
50	27.33±1.45 (25-30)	17.67±0.88 (16-19)	64.67±1.02 (63.33±66.64)	22.33±1.45 (20-25)	14.33±0.88 (13-16)	64.36±3.16 (59.09-70)	8.33±1.20 (6-10)	1.0±0.58 (0-2)	12.22±6.19 (0-20)	-	-	-
100	16.67±1.45 (14-19)	12.67±0.88 (11-14)	77.7±10.36 (57.89-92.86)	13.0±1.73 (10-16)	9.33±033 (9-10)	73.91±8.28 (62.5-90)	Nil	Nil	Nil	-	-	-
500	13.67±1.20 (12-16)	7.33±0.88 (6-9)	55.02±9.31 (37.50-69.23)	9.33±0.88 (8-11)	5.0±0.58 (4-6)	55.64±11.16 (36.36-75)	Nil	Nil	Nil	-	-	-
Control	65.33±5.78 (55-75)	61.67±5.24 (52-70)	94.44±0.62 (93.33-95.4)	62.33±6.23 (50-70)	57.66±5.46 (47-65)	92.64±0.86 (91.05-94.0)	62.23±6.69 (50-73)	59.67±6.07 (49-70)	95.88±1.2 (93.75-98.0)	59.67±6.74 (47-70)	56.33±6.12 (45-66)	94.53±0.64 (93.55-95.74)
LSD	9.89	8.90	20.59	10.13	8.22	20.24	13.92	12.23	12.82	-	-	-
F(Cal.)	44.98**	59.53**	5.27*	43.37**	65.77**	4.99*	51.23**	86.08**	144.45**	-	-	-

$\bar{x}$ = Mean number and s$\bar{x}$ = Standard devisition

P= Parents , F1= First generation , F2= Second generation and F3= Third generation

*= Significant ,**= highly significant and ***= Average

concentrations of MYX_{833} increased the mean numbers of the emerged moths were decreased in first generation than parents one. Also, the few number of the emerged adults after treatment with the higher concentrations of 100 and 500 ppm were unviable, weak, sterile and died before the reproduction of the F_2 progeny (Table 2). Generally, MYX_{833} gave highly significant effect on all the studied biological characters of that insect- pest. The present results are in agreement with those reported by Khan et al. (2005) who found that MYX_{833} was more toxic against 3rd instar larvae of a susceptible Diamond back moth laboratory strain (*Plutella xylostella*) than the biopreparation MYX_{104} giving Lc_{50} of 2.63 and 3.26 mg a.i /l , respectively.

2nd generation (F_2)

Similarly, the emerged moths from each treatmental concentration and untreated control of 1st generation were daily collected , sexed and separated. Then, they were paired and released on the clean tubers in adopted treatments of 2nd one. Three couples were used per treatment for studying the moths emergency of resulting (F_2) progeny of each cross.

Identically, the presented results in Table 2 showed that the resulted (F_2) moths from paired untreated sexes lived longer period. Also, results indicated the utmost reduced numbers of resulted (F_2) pupae according to the former MYX_{833} treatments in comparison to P and F_1 generations giving average mean numbers of 18.0 ± 1.53 to 8.33 ±1.20 pupae at 25 ppm up to 50 ppm, respectively. In untreated control, the mean number of the resulted pupae was 62.33 ± 6.69.

Consequently; the delayed effect of former (P) treatment by MYX_{833} was reflected on the decreased rates of (F_2) emerged moths, compared to control treatment, which gave the highest mean number of emerged moths (59.67 ± 6.07) . The delayed effect of the high dose of MYX_{833} (50 ppm), to great extent, decreased the mean number of the emerged moths of second generation, in comparison to P and F_1 generations. The results also showed high significant effect of this dose on most of the studied biological characters, whereas the incipient moths were inactive and sterile (Table 2). Remarkably, treatment with MYX_{104} at concentrations of 100 and 500 ppm, completely inhibited the reproductive potential of both sexes of second generation in comparison to the lower tested concentration of 25 and 50 ppm (Table 2).

3rd generation (F_3)

From Table 2 it is worth to show the final failure of (F_3) development at the initiated lower concentration of MYX833 (25 ppm) and that could be attributed to the cumulative effect of induced recessive lethal genes in both influenced sexes along the following developing generations of (P), (F_1) and (F_2); causing unprofitable effects 3rd generation.

The above-mentioned results of testing *B.t* biopreparations (MYX_{104} and MYX_{833}) against *P. operculella* to a great extent, are in agreement with those earlier suggested by many workers, who studied and evaluated the used biopreparations of *B. thuringiensis* for controlling the potato tuber moth.

Herein, Arx *et al.* (1987) stated that the control of *P. operculella* was achieved by applying permethrin or *B. thuringiensis* at the beginning of the storage period .

Moreover, Arx and **Gebhardt** (1990) observed the standard application of Thuricide HP® [*B.t.* sub sp. *Kurstaki*] at 200 mg / kg potatoes) on the life table parameters of *P. operculella* .Survival from egg to adult emergence was lowest (0.4%) and no reproduction occurred when larvae were fed on tubers treated with Thuricid HP® .

Studying the persistence of tested *B.t* biopreparations, Salama *et al*. (1995) showed that *Bacillus thuringiensis* sub sp. *Kurstaki*, HD-1 protected potato tuber when applied inside stores for periods upto 255 days, particularly at high doses. Similarly, Das *et al*., (1998) stated that *B. thuringiensis* was effective in controlling *P. operculella* in stored potatoes.

Kroschel and Kock (1996) decleared that *B. thuringiensis*/sand mixture was extremely effective to larvae in tubers, with success rate of 96%. However, the effect must to be attributed to *B.t*. since sand alone was only 21.6% effective in reducing the development of adults.

In a field experiment in 1995 in Egypt, Bekheit *et al*. (1997) determined that *B. thuringiensis* reduced the infestation level by *P. operculella* in potatoes up to 82.5-95%.

In a laboratory evaluation of different *B. thuringiensis* sub species, Puntambokar *et al*. (1997) revealed that *B.t* sub sp *Kurstaki* (NCIM 2514) at 108 spores/ml concentration caused more than 85% mortality to neonate larvae of the lepidopteran insects *Spodoptera litura* and *P. operculella* .

Maowad *et al*. (1998) stated that the exposures to *B. thuringiensis* (*B.t*) and granulosis virus (GV), significantly, reduced pest infestation, larval population and yield loss compared to the control.

Also dust formulations of *B. thuringensis* (*B.t*) gave good protection of potato tubers from infestation by the

potato moth, *P. operculella* (Hamilton and Macdonald, 1990).

Moreover, the efficiency of microbial control by *B. thuringensis* was explained in the works of Jansens *et al*.(1995) and Kroschel *et al*. (1996).Also, Hernandez *et al* . (2005) stated that the isolated *B. thuringiensis* from 116 samples collected in high altitude potato-growing areas in Bolivia were found to be most toxic against *S. exigua* and *P. operculella* and these isolated B.t differed in their efficacy against different insect-pests due to their serotyping, crystal morphology, protein profile, and cry gene content.

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الملخص العربي

تأثير نوعين من التجهيزات الحيوية لبكتريا الباسيللس ثيورينجينسيس (MYX_{104}،MYX_{833}) على دودة درنات البطاطس

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أجري هذا البحث بدف تقييم تأثير التركيزات المختبرة مـن التجهيزات الحيوية لبكتريا الباسيللس ثيورينجينسيس (MYX_{833} و MYX_{104}) علي فراشات درنات البطاطس المعاملة تحت الظـروف المعملية وقد أظهرات النتائج تأثيرات متماثله مباشرة أو ممتدة المفعول لكل من المركبين الحيويين المختبرين حيث أدت كـل المعـاملات المستخدمة الى حدوث النقص التدريجي المعنوي في فترة حياه الحشرة الكاملة لكلا الجنسين وزيادة فترة زمن الطور اليرقـي. كـذلك زادت معدلات الإنخفاض الحاد في عدد العذاري والفراشات الناتجة حسب التركيز المختبر لكلا من التجهيزتين الحيويتين المستخدمتين.

كما وضحت التأثيرات الممتدة المفعول في الأجيال المتتالية مـن بعد معاملة جيل الأباء حيث انخفضت حيوية جميع الأطوار المختلفة وخاصة الحشرات الكاملة، والتي أعقبها التوقف النهائي لدورة حياة الحشرة والفشل الواضح للنمو والتطور في بداية الجيل الثالث نتيجة معاملة جيل الأباء بالتركيز المنخفض (٢٥ جزء في المليون) من كل منس الـ MYX_{104}، MYX_{833}. وهذا الفشل الواضح يمكـن أن يعزى إلى حدوث التأثير التراكمي للجينات المتنحية المميتة في كـلا الجنسين وعلى مدى فترات تطور ونمو تلك الأجيال المتتالية بعـد جيل الأباء المعامل مسببة ظهور تأثيرات ضارة، ظهـرت جليـا في بداية الجيل الثالث، وقد أتضح أن تجهيزه الـ MYX_{104} كانـت أكثر تأثير وفعالية على المقاييس التي تم دراستها لمكونات الموائمـة للحشرة أكثر من نظيرتا MYX_{833}