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EFFECT OF CERTAIN COMMERCIAL PREPARATIONS OF *BACILLUS THURINGIENSIS* ON LEAF MINER *LIRIOMYZA BRYONIAE* (KALTENBACH) PUPAE (DIPTERA – AGROMIZIDAE)

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Abstract

Laboratory studies were conducted to determine the effects of five preparations of *Bacillus thuringiensis* (Bt) on leaf miner pupae. Bt formulated as Bactospeine was the most effective (LC50 value 6.2 x 10^7 IU/L) on the pest followed by Dipel, while thuricide 64LV was the least potent as it showed the highest LC50 value (99.5 x 10^7 IU/L). These results suggest a useful role of Bt formulations Bactospeine and Dipel in control measures against leaf miner on vegetable crops.

INTRODUCTION

The polyphagous fly *Liriomyza bryoniae* is a major economic pest of vegetable crops such as tomato and cucurbits in greenhouse and outdoor in many countries (Ledium and Helyer, 1985; MacDonald, 1991; Mikenberg 1988). Kuwait practices protected environment agriculture (PEA) system to produce vegetable crops. Greenhouse conditions that promote intensive growth and productivity of vegetable have encouraged higher insect activities. The injury threshold for leaf miners on greenhouse tomatoes is relatively high (about 10 mines per plant). Damage is caused by larvae mining into leaves and petioles. The plant's photosynthetic ability is often greatly reduced as the chlorophyll-containing cells are destroyed. Severely infested leaves may fall exposing plant stems to wind action and flower buds and developing fruit to scald. In young plants and seedlings mining may cause considerable delay in plant development leading to plant loss (Converse 1933 and Anon 1994). Chemical insecticides are currently used to control this insect pest, but development of resistance as well as environmental concerns restrict their use as efficient control agent.

Insect control methods using bioinsecticides have emerged as powerful alternatives to conventional chemical insecticides (Sommerville, 1978; Aronson *et al.*, 1986; Hofte and Whiteley, 1989). Biological insect control methods are environmentally safer and more economical (Kirschbaum 1985). During the past several years, some *Bacillus thuringiensis* (Bt) strains have been successfully exploited for the control of specific target pest. More than 182 species of insects have been found to be susceptible to Bt-based bioinsecticides (Dean 1984), no information is available on the effect of Bt-based bioninsecticides on leaf miner. This study was conducted to determine the efficacy of some Bt-formulations against leaf miner pest under laboratory conditions.

MATERIALS AND METHODS

Laboratory Test: *Liriomyza bryoniae* pupae were collected from their host plants inside greenhouses. The insects were reared on tomato and eggplant seedlings raised under laboratory conditions as described by (Minkenberg, 1998; Oscar and Minkenberg, 1988).

A number of leaf miner pupae at the same age were transferred into petri dishes containing filter papers treated with the target bioinsecticides. Treatments were applied by the dipping technique. Mortality counts were recorded three days after treatment. Healthy pupae were barrel shaped, straw coloured and had obvious contents. Any pupae that were mishappen, dark-coloured or obviously dried husks were classified as dead. An alternative method of assessing the mortality on the basis of adult emergence was abandoned because of the low rate of successful adult emergence in the controls, which was typically only 50% (MacDonald 1991). Control not treated with pesticides were treated with tap water only using the same technique. Nine pupae were used in each treatment and three replicates were established for every treatment. The data were corrected for natural mortality according to Abbott's formula (1925).

The bioinsecticides preparations used were Thuricide 48 LV, i.e., *Bt* subspecies *Kurstaki* preparation, potency 12,000 IU/mg; Thuricide 64 LV, i.e., *Bt*, subspecies *Kurstaki*, potency 16,000 IU/mg; Thuricide HPC, i.e., Bt Berliner, potency 4,000 IU/mg; Dipel Es, i.e., *Bt* var *Kurstaki*, potency 17,600 IU/mg; and Bactospeine, i.e., *Bt* var *Kurstaki*, potency 17,000 IU/mg; and Bactospeine, i.e., *Bt* var *Kurstaki*, potency 16,000 IU/mg.

Greenhouse Test: Cultures of *L. bryoniae* were reared on tomato seedlings inside a greenhouse in PAAF area. The experiment was performed in triplicate and each replicate contained eight plants. Infested seedlings were sprayed twice with the required dose of the Bactospeine (11.16 x 10^7 I.U./L) toxin. Two plants were chosen randomly from each replicate and the number of surviving insects (all stages found on the whole plant) were counted before and after spraying. Another infested seedlings were conducted as control and sprayed only with water. The percentages of reduction were calculated as described by Handerson and Tiliton (1955).

$$\% \text{ Reduction} = \frac{1 \text{ Control before application}}{\text{Control after application}} \times \frac{\text{Treatment after application}}{\text{Treatment before application}} \times 100$$

RESULTS AND DISCUSSION

The data in Table 1 present the effects of various Bt-based bioinsecticides on the mortality rates of leaf miner pupae. The highest bioinsecticides concentration (128 x 10^7 I.U./L) used against leaf miner pupae resulted in 66.7%, 55.6%, 77.8%, 88.9% and 92.6% mortality for Thuricide 48 LV, Thuricide 64 LV, Thuricide HPC, Dipel and Bactospeine, respectively. In case of the lowest bioinsecticide concentration (4 x 10^7 I.U/L), the mortalities were 11.1%, 0.0%, 22.2%, 22.2% and 44.4% for Thuricide 48 LV, Thuricide 64 LV, Thuricide HPC, Dipel and Bactospeine, respectively, Table 1. These results suggest that at higher concentration, Dipel and Bactospeine produced the highest effects on the mortality rates of leaf miner pupae, followed by Thuricide HPC, Table 1. Data presented in Table 1 indicate also, the LC50 values of the tested Bt bioinsecticides against leaf miner pupae. Bactospeine showed the lowest LC50 value (i.e., 6.2 x 10^7 IU/L) of the tested preparations, it was followed by Thuricide HPC (12.5 x 10^7 IU/L) and Dipel (20.0 x 10^7 IU/L), whereas Thuricide 64 LV showed the highest LC50 value, Table 1. It was noticed that the sublethal doses of the tested commercial bionisecticides caused very clear abnormalities when tested against tomato leaf miner pupae. These abnormalities appeared not only in the same generation, but also in the further progeny. The abnormalities led to the death of the insects, Table 1.

The data in Table 2 show the percentages of reduction for all stages of the leaf miner insects (larvae, pupae and adults) before and after spraying with the commercial bioinsecticide Bactospeine. The percentage of reduction for leaf miner insects after the first spray calculated with Equation by Handersona and Tiliton (1955) was 73.2% after the first spray and increased to 89.3% after the second spraying. This study has demonstrated that Bt-based bioinsecticides (e.g., Bactospeine) is capable of inducing death in leaf miner insects. Similar observations on another dipterous insect, *Mansonia bon-*

neae (Diptera: Culicidae) have been reported by Chang *et al.* (1990). These workers demonstrated the toxicity of three Bt-based bioinsecticides against larvae of *M. bonneae* in simulated field studies. A possible explanation for the mortality rates and latent effects found in pupae could be the contact action of the tested formulation as explained by Radwan *et al.*, 1984 and Al-Shayji *et al.*, 1998. Another explanation for the mortality rates of the larvae could be through the penetration of the toxin to the internal tissues of the leaf and then to the larvae midgut. The inactivated protoxin will be proteolytically converted to activated toxin which interacts with the epithelial cells of the midgut of the susceptible insects. Evidence suggests that the toxin generates pores in the cell membrane, thus disturbing the osmotic balance. Consequently, the cells swell and lyse. The larva stops feeding and then dies (Harvey *et al.*, 1983; Haider and Ellar, 1987 a,b). Thus, the present data and the results reported (Chang *et al.* 1990) suggest the potential applications of Bt-bioinsecticides as an alternative control measure to reduce the population of dipterous insects including leaf miner on vegetable crops.

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Table 1. Effect of some commercial bioinsecticides on the mortality rates of leaf miner pupae

Bioinsecticides Concentration (IU/L)	Thuricide 48 LV	Thuricide 64 LV	Thuricide HPC	Dipel	Bactospeine
128×10^7	66.7±9.1a	55.6±9.1a	77.8±5.7a	88.9±5.7a	92.6±10.5a
64×10^7	44.4±9.1b	66.7±9.1b	44.4±5.2b	77.8±5.7b	88.9±5.7a
32×10^7	33.3±0c	33.3±9.1c	66.7±0b	66.7±9.1c	77.8±9.1b
16×10^7	22.1±0d	22.2±9.1d	55.6±9.1c	44.4±9.1d	66.7±0c
8×10^7	11.1±0e	11.1±5.2e	44.4±9.1d	33.3±0e	55.6±9.1d
4×10^7	11.1±0e	0.0f	22.2±0e	22.2±0f	44.4±5.7e
Control	0.0f	0.0f	0.0f	0.0g	0.0f
LC50 Values (IU/L)	80.0×10^7	99.5×10^7	12.5×10^7	20.0×10^7	6.2×10^7

- Mortality percentages were adjusted by Abbott's formula.
- Means of three replicates ± SD.
- Means in a column followed by the different letters are significantly different at a 0.05 level of probability (LSD).
- Means in a column followed by the same letters are not significantly different at a 0.05 level of probability (LSD).

Table 2. Effect of the commercial bioinsecticides bactospeine on leaf miner insects

Bioinsecticide	Percentage of reduction	
	After First Spraying	After Second Spraying
Bactospeine	73.2	89.3

Rate of application = 11.16×10^7 I.U./L

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تأثير بعض مستحضرات الباسلاس ثورنجينسس التجارية على خادرات صانعات الأنفاق في أوراق الخضر *Liriomyza bryoniae* (Kaltenbach) (Diptera : Agromizidae)

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تم إجراء الدراسات المخبرية لتحديد تأثير خمسة مستحضرات من الباسلاس ثورجينسس على طور الخادرة لحشرة صانعات الأنفاق.

وجد أن المستحضر المسمى باكتوسبين كان الأكثر فعالية ضد هذه الحشرة (LC50 value 6.2×10^7 IU/L) يليه مستحضر الدايابيل، بينما وجد أن الثوريسايد 64 LV كان الأقل فعالية حيث أظهر أعلى تركيز لقتل نصف عدد الحشرات (LC50 value 99.5×10^7 IU/L).

وقد بينت هذه النتائج أن مستحضرات الباسلاس ثورنجينسس (الباكتوسبين والدايابيل) يمكن أن تلعب دوراً مفيداً في مكافحة حشرة صانعات الأنفاق المهلكة لمحاصيل الخضراوات.