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FIELD AND SEME-FIELD EVALUATION OF BIOCIDES LOCALLY FORMULATED AGAINST THE FIELD COTTON LEAFWORM *Spodoptera littoralis* (BOISD.)

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ABSTRACT

Field and Semi-field experiments were carried out in Manshiat-Ganzor in Gharbya governorate during 2009 to study the effect of three biocides used for the control of the cotton leafworm. The treatments included Viroset 4% W.P., Protecto 9.4% W.P. and Prophect 7% W.P. at its half (150gr/F) , recommended (300gr/F) and double dose (600gr/F). The results indicate that the 2nd instar larvae of Gharbya cotton leafworm fed on the samples of treated leaves for each treatment, only 20, 30 and 20% larval mortality were observed after 3 days to the higher concentration (600gr/F) to viroset, Protecto and Prophect respectively, such larval mortality were increased to 65, 65 and 68% after 13 days to the samples collecting after (zero time). The data indicated that treatment of larvae with the tested biocides drastically reduced the rate of % pupation, % of emergency and increased the mortality and deformation in pupal and adult stages. The treatments also reduced egg production and % hatchability. With regard to hatchability , obtained results indicated that Protecto was the most effective (0% hatchability) followed by Viroset 12.11% and Prophect 15.3% when they were applied at their double dose at zero time of spray. The field experiments showed that Protecto was the most potent biocide in reducing the population of the cotton leafworm larvae after 8 days of application, particularly when this compound used at 600 gr/Feddan. The reduction in larval population after 8 days of treatments with Viroset , Protecto and Prophect at (600gr/F) were 40 , 80 and 77%.

The previous results suggest that biocides compounds may be recommended as an effective component of the future IPM programs against *Spodoptera littoralis* on cotton fields.

INTRODUCTION

Extensive use of chemical insecticides has been consumed for the control of the most economic insect *Spodoptera littoralis* on several crops . However, indiscriminate use of such chemicals have resulted in well known health and environmental problems. Alternative control strategies, such as using natural enemies of pest insects on biological control agents, are needed to solve these problems.

Baculoviruses have been acknowledged as potential biological control agents for agricultural pests, and are expected to have fewer negative impacts than chemical pesticides on animals, humans and the environment (Madoka and Nguyen , 2005).

Also, *Bacillus thuringiensis* crystal proteins are preferred and widely used as an alternative to chemical pesticides in pest management strategies against insect pests of agriculture crops (Roh *et al*, 2007).

The present studies were conducted to assess the efficiency of local bio-insecticide compounds Viroset 4%WP, Protecto 9.4%WP and Prophect

7%WP on the toxicity and biotic potential of 2nd instar larvae of *Spodoptera littoralis* (Gharbya strain) under laboratory conditions. Also, studies were conducted to determine the percentage of reduction of the population of the cotton leafworm under field conditions.

MATERIALS AND METHODS

1- Biocides used:

a- Viroset 4%WP granulosis virus (GV).

b- Protecto 9.4%WP based on *Bacillus thuringiensis* subsp kurstaki.

c- Prophect 7%WP contained 3.5% granulosis virus (GV) + 3.5% *B.t.* kurstaki.

These compound produced by the Plant Protection Research Institute, Agricultural Research Center, Dokki , Cairo , Egypt.

2- Insect strain:

The field strain of the cotton leafworm *Spodoptera.littoralis* was obtained from Gharbeya governorate. This strain was kept under normal laboratory conditions at 25º ± 2ºC and 70º ± 5º relative humidity. The egg-masses were kept separately until the eggs hatched and then provided with castor-oil leaves.

3- Field and semi-field experiments:

Field experiments were carried out in Manshiat – Ganzor in Gharbya governorate during successive season 2009. An area of two feddans was divided into 40 plots and cultivated with cotton plants variety (Giza 89) on 28/3/ 2009.

The experiment included three treatments. The treatments were carried out using the tested biocides Viroset , Protecto and Prophect at its half (150gr/F) , recommended (300gr/F) and double dose (600gr/F). All treatments including the control were replicated four times and the tested compounds were applied using a small motor sprayer. A completely randomized design was applied for each treatment.

For the semi-field experiment, random samples of treated leaves for each treatment were taken at 0, 3, 5, 7, 9, 11 and 13 days after spraying and transported to laboratory. Hundred 2nd instar larvae were fed on these treated leaves for 24h. and then fed on untreated leaves until pupation . Percentage of pupation and moth emergence were based on the number of normal pupae or moths obtained. For mating experiments the normal adults were grouped in pairs and each pair was placed in small cage provided with *Nearium oleanaer* leaves which served as an oviposition site. Insects resulted from such treatments were maintained under constant temperature 25º ± 2ºC and relative humidity 70º ± 5º to determine the different biological criteria.

For the field experiment, the average number of *S. littoralis* larvae present in 20 plants in each plot was recorded just before and after experiments 2,4,6 and 8 days of treatments. Percentage of larval reduction were calculated according to Hendrson and Telton (1955).

RESULTS AND DISCUSSION

The effect of the tested biocides on the percentage of larvae mortality , % pupation , % pupal mortality , % deformation in pupae , % emergence , % deformation in moths , Mean No. of egg/female and % hatchability of the Gharbeya cotton leafworm was studies. The results shown in Tables (1,2 and 3) represent the average % mortality of 2nd instar larvae of *S. littoralis* fed on treated cotton plant leaves for 24h. in laboratory and then fed on untreated leaves. As shown in Table (1) the tested virus (viroset) recorded very low mortality percentages in the larval stage reached only 35% , 50% and 65% to the half , recommended and double recommended dose after 13 days of treatment to the samples collecting after zero time. Viroset caused 8, 5 and 6% pupal deformation and 5, 7 and 10% adult deformation after zero time of spraying.

Alaa Eddeen (2008) found that the newly molted 2nd instar larvae of *S. littoralis* were susceptible to the applied concentrations of two tested isolates (Spli NPV and ACM NPV). Time to death showed some dependence on the initial concentration . Further more , it was observed that the highest peak of mortality was on 4-6 days post-infection . It is clear from the data shown in Table (2) that treatment of 2nd instar larvae with *Bacillus thuringiensis* Protecto drastically reduced the rate of pupation, emergency, egg production and hatchability . The percentage of pupation was 40 , 32 and 25% while it was 90% in control and the rate of emergence of moth ranged 25 , 20 and 15% while that of the control reached 85% for the three treatments used (150 , 300 and 600 gm/feddan) at zero time.

More over , the treated larvae at the double recommended dose (600 gr/f) at zero time did not lay eggs. An interesting observation in the case of larvae infected with higher concentrations of *Bacillus thuringiensis* was the sudden rise in mortality the penultimate day of pupation . This sudden surge in mortality may be attributed to metabolic changes that occur in larvae during the transition from larval to pupal stages . Similarly the deformities that occur in adults also increase at higher concentrations. There may be attributed to difficulties in moulting and shedding the exuviae as also suggested earlier (Srinivason *et al* 2001).

Shachindra *et al* (2009) investigated the degree of effectiveness of *Bacillus thuringiensis* aizawai for 2nd instar and *Bacillus thuringiensis* kurstaki for 1st instar larvae of *S litura*. All the development stages of *S. litura* were found to be both in *B.t.*a and *B.t.*k although to a varying degree . Similar trends regarding larval mortality , pupal deformity , pupal mortality , adult deformity and normal adult emergence were noted in case of all the stages on infection with various concentrations of both *Bacillus thuringiensis* used.

The data shown in Table (3) indicate that feeding the 2nd larvae of *S. littoralis* on half recommended, recommended and double recommended dose of Prophect for 24h.caused only 50, 62 and 68% larval mortality after 13 days of treatment to the samples collecting after zero-time of spraying.

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Abd-El-Hai (2008) revealed that the mixtures of viroset with *Bacillus thuringiensis* at their ¼ and ¾ recommended dose had potentiation effects . However, the mixtured of mentioned compounds at their ½ recommended dose produced additive effects by Co-toxicity factors ranging from 11.69 for Dipel 2x to 16.25 for Protecto. The highest potency was observed at the mixture of ¼ dose of *B. thuringiensis* with ¾ dose of Virocet .

The data of the field applications illustrated in Table (4) showed that Protecto was the most potent biocide in reducing the population of *S. littoralis* larvae after 8 days of application. The reduction percentage was 37 , 65 and 80% for the half and recommended and double recommended dose respectively .

Table (4): Percentage of reduction of *Spodoptera littoralis* larvae on cotton plant leaves treated with the tested biocides compounds.

Treatment	Rate / Feddan gr / Feddan	% reduction at indicated days			
		2	4	6	8
Viroset	150	1	9	19	8
	300	10	26	45	30
	600	25	39	65	40
Protecto	150	9	15	26	37
	300	18	41	55	65
	600	25	62	75	80
Prophect	150	0	3	24	36
	300	20	35	56	60
	600	36	50	62	77

Data revealed that prophect and Viroset come in the second and third order with reduction percentage of 36 , 60 and 77% and 8 , 30 and 40% after 8 days of application for this different treatments. Abd-El-Salam *et al* (2011) indicated that *Bacillus subtilis strain NRC313* was more potent than *Bacillus thuringiensis strain NRC335 against the third larval instar of the cotton leafworm.* Schroer *et al* (2005) indicated that use *Bacillus thuringiensis* and *Bacillus subtilis* in field experiment to controlling larvae of *Spodoptera littoralis* was efficient compared with Reldan as a phosphorus compound , whereas the mean percentage reductions were 55.7 , 67.4 and 89.4% for *Bacillus thuringiensis , Bacillus subtilis* and Reldan respectively in clover plants. Weekly applications of *Bacillus thuringiensis* achieved > 89% control of the diamond back moth, *Plutella xylostella* in cabbage plants under field conditions.

In conclusion, the biocides may provide a substitute for chemical insecticides to manage field infestations of cotton leafworm especially in light to moderate populations.

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التقييم الحقلى والنصف حقلى لبعض المركبات الحيوية المصنعة محليا ضد يرقات دودة ورق القطن

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تم تقييم تجربة حقلية فى محافظة الغربية (منشية جنزور) وتم اختيار كل من المركبات الحيوية فيروست 4٪ القابل للبلل والبروتكتو 9,4٪ القابل للبلل وبروفيكت 7٪ القابل للبلل ضد دودة ورق القطن0

تم تجربة هذه المركبات بنصف التركيز الموصى بة 150 جم/فدان والتركيز الموصى بة 300جم/فدان وضعف التركيز الموصى بة 600جم/فدان . تم رش هذه المركبات بتركيزاتها المختلفة فى محافظة الغربية (منشية جنزور) على نباتات القطن المزروعة بصنف (جيزة 89) فى 2009/3/28

للتقييم النصف حقلى ، تم اخذ عينات عشوائية من الأوراق المعاملة بعد الرش مباشرة وجفاف الأوراق (صفر وبعد 3 ، 5 ، 7 ، 9 ، 11 ، 13) يوم الى المعمل لتقييم هذه المركبات تحت الظروف المعملية0

وجد أن تغذية العمر الثانى ليرقات دودة ورق القطن لمدة 24 ساعة أعطت موت للطور اليرقى بلغ 20،30،20٪ بعد ثلاثة أيام من المعاملة زادت الى 68،65،65٪ بعد 13 يوم فى التركيز العالى 600جم/فدان لكل من الفيروست ، البروتكتو والبروفكت عند التغذية على الأوراق التى جمعت بعد الرش مباشرة وجفافها (صفر).

ووجد ان لهذه المركبات تأثير على خفض نسبة التعذير و نسبة خروج الفراشات ، كما انها تسبب زيادة فى موت وتشوة العذارى والفراشات بنسب مختلفة على حسب التركيز المستخدم 0 كما أنها تسبب نقص فى عدد البيض الموضوع ونسبة الفقس فوجد ان المعاملة بمركب البروتكتو بتركيز 600 جم/فدان قد نتج عنة فشل تام فى نسبة الفقس ووصل الى 12.11% عند المعاملة بمركب فيروست و15.3٪ عند المعاملة بمركب بروفيكت عند استعمال نفس التركيز عند التغذية على الأوراق التى جمعت بعد الرش مباشرة وجفافها (صفر)، ووجد ان مركب البروتكتو أكثر المركبات فعالية فى خفض مجتمع دودة ورق القطن بعد 8 ايام من المعاملة حيث وصلت نسبة الخفض الى 80٪ فى حالة استخدام ضعف التركيز الموصى بة يلية مركب البروفكت 77٪ ثم الفيروست 40٪.

من هذه النتائج المعملية والحقلية يمكن التوصية باستخدام هذه المركبات فى مكافحة دودة ورق القطن خاصة وعندما تكون الإصابة متوسطة أو ضعيفة.

قام بتحكيم البحث

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Table 1 : Effect of Viroset 4%WP against 2nd instar larvae of *Spodoptera littoralis* (Gharbya strain) under laboratory conditions.

Treatment Gr/feddan	Days of collecting samples after spray	% mortality in larvae days							% of pupation	% mortality in pupal	% deformation in pupal	% of emergency	% deformation in moth	Mean No of egg/ female	% of hatchability
		1	3	5	7	9	11	13							
150	0	0	5	10	10	20	30	35	52	5	8	40	5	118.2	55.5
	3	0	0	5	5	5	10	10	70	7	2	65	2	280.7	70.5
	5	0	0	0	0	5	5	10	80	5	5	70	5	405.1	80.3
	7	0	0	0	0	2	2	2	79	5	4	72	2	441.7	85.1
300	0	5	10	20	20	40	45	50	40	5	5	28	7	90.4	40.42
	3	0	0	5	5	7	8	15	67	8	5	57	5	300.9	65.4
	5	0	0	2	2	5	5	15	72	5	3	66	3	357.4	81.5
	7	0	0	0	0	4	5	5	80	5	5	75	1	424.3	81.8
600	0	10	20	35	40	40	45	65	27	2	6	17	10	92.33	12.11
	3	0	0	0	10	15	15	15	55	10	5	47	5	255.7	53.7
	5	0	0	0	10	15	15	20	63	7	5	57	5	390.2	76.7
	7	0	0	0	0	5	5	5	75	10	5	67	1	475.1	70.6
control		0	0	2	2	2	5	5	90	0	2	85	2	502.9	92.1

Table 2 : Effect of Protecto 9.4%WP against 2nd instar larvae of *Spodoptera littoralis* (Gharbya strain) under laboratory conditions.

Treatment Gr/feddan	Days of collecting samples after spray	% mortality in larvae days							% of pupation	% mortality in pupal	% deformation in pupal	% of emergency	% deformation in moth	Mean No of egg/ female	% of hatchability
		1	3	5	7	9	11	13							
150	0	0	5	20	30	45	50	50	40	5	5	25	1	79.1	10.3
	3	0	0	0	15	20	20	25	65	5	2	55	1	411.1	62.3
	5	0	0	0	5	5	10	15	75	2	5	70	1	410.2	79.5
	7	0	0	0	5	5	5	10	85	2	2	81	2	442.5	84.3
300	0	5	20	25	30	45	52	60	32	5	5	20	1	60.9	3.5
	3	0	0	20	20	25	30	30	56	7	2	45	1	392.4	53.7
	5	0	0	5	10	20	20	20	70	5	2	65	2	327.3	70.3
	7	0	0	0	0	10	15	15	80	2	1	76	2	415.3	82.1
600	0	10	30	35	40	55	60	65	25	5	2	15	2	0	0
	3	5	5	10	20	35	40	45	42	8	5	35	2	41.7	32.0
	5	0	0	0	1	5	5	5	68	2	1	61	1	445.1	67.3
	7	0	0	0	0	5	5	5	89	2	2	83	1	399.2	77.2
control		0	0	2	2	2	5	5	90	0	2	85	2	502.9	92.1

Table 3 : Effect of Prophect 7%WP against 2nd instar larvae of *Spodoptera littoralis* (Gharbya strain) under laboratory conditions.

Treatment Gr/feddan	Days of collecting samples after spray	% mortality in larvae days							% of pupation	% mortality in pupal	% deformation in pupal	% of emergency	% deformation in moth	Mean No of egg/ female	% of hatchability
		1	3	5	7	9	11	13							
150	0	0	5	10	20	35	45	50	45	2	1	42	5	192.3	90.1
	3	0	0	10	10	15	15	20	60	10	2	45	1	329.5	82.3
	5	0	0	0	10	10	10	15	76	5	2	70	1	390.1	86.2
	7	0	5	5	5	5	5	5	85	5	2	80	2	572.3	85.2
300	0	5	20	30	35	45	50	62	30	2	2	25	2	143.2	80.2
	3	0	0	0	15	20	25	40	51	5	5	35	2	415.6	75.6
	5	0	0	10	10	15	20	20	72	5	5	64	5	410.2	80.1
	7	0	0	0	0	5	5	10	80	5	5	73	2	613.2	83.4
600	0	10	20	40	45	50	60	68	19	5	5	10	2	80.1	15.3
	3	5	5	10	20	30	35	40	42	5	5	31	5	409.3	50.9
	5	0	0	0	20	20	25	30	60	5	7	48	5	325.7	77.7
	7	0	0	10	10	10	10	10	75	10	5	66	1	482.9	87.4
control		0	0	2	2	2	5	5	90	0	2	85	2	502.9	92.1