



EGYPTIAN ACADEMIC JOURNAL OF
BIOLOGICAL SCIENCES
TOXICOLOGY & PEST CONTROL

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ISSN
2090-0791

WWW.EAJBS.EG.NET

Vol. 13 No. 2 (2021)



Comparative Effect of Some Bio Insecticides with Chlorpyrifos on Cotton Leafworm

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ARTICLE INFO

Article History

Received: 21/10/2021

Accepted: 19/12/2021

Keywords:

Insecticidal activity
, Chlorpyrifos,
Dipel DF, cotton
leafworm.

ABSTRACT

The efficiency of Chlorpyrifos, Jojoba and Dipel DF against 2nd and 4th instars of the laboratory strain of *Spodoptera littoralis* was evaluated under laboratory conditions. Chlorpyrifos was the most toxic compound against the 2nd and 4th larval instars of *Spodoptera littoralis*. All the treated larvae were biologically affected by the three tested compounds. The effect was varied according to the larval instars and tested compounds. Therefore, the treated larvae have resulted in decreased pupation and adult emergence percentages, and the 2nd instar treated with Chlorpyrifos had the strongest effect in this respect. The treatment of 2nd instar with the three compounds induced the highest increase in larval, pupal duration and adult emergence percentages.

Hence, the larval treatment of 2nd and 4th instars with Chlorpyrifos and Dipel DF gave the shortest period of adult longevity, as compared to control. The larval treatment of 2nd and 4th instars with the three tested compounds increased the adult emergence, the treatment of 2nd instar with Chlorpyrifos and 4th instar with Dipel DF had the strongest effect in this respect.

INTRODUCTION

The cotton leafworm, *Spodoptera littoralis* (Boisd) is a key polyphagous pest in Egypt. Without a hibernation period, the cotton leafworm is active all year. It was the most destructive insect pest of the great variation of important vegetables and field crops, approximately 112 species belonging to 4411 families are attacked by this pest.

Among the wide range of hosts, cotton, soybean, maize, wheat and vegetable crops (e.g., tomato, potato and strawberry) are processes in insects such as chitin synthesis inhibitors, juvenile hormone mimics, and ecdysone agonists.

This group of insecticides consists of various compounds acting on insects of different orders by inhibiting chitin formation, thereby causing abnormal endocuticular deposition and abortive molting. The activity of biochemical sites (IPM). They have high pathogenicity for target pests. Safe for most non-target organisms, and have good integration with other pest control methods (Ibrahim *et al.*, 2010).

The principal aim of the present study was to evaluate the toxic effect of Dipel DF, Jojoba and Chlorpyrifos in addition to untreated check control against *S. littoralis*

MATERIALS AND METHODS

Insect Rearing:

The cotton leafworm, *Spodoptera littoralis* was reared in the laboratory for several generations at room temperature ranging between 25 - 26°C and 60 -65% R.H. Larvae were fed on castor bean leaves, *Ricinus communis* (L.) in a wide glass jar until pupation period and adults emergence. The newly emerged adults were mated inside glass jars supplied with a piece of cotton wetted 10% sugar solution as a feeding source for the emerged moths and Branches of Tafla (*Nerium oleander* L.) or castor bean leaves were placed as an oviposition site (El- Defrawi *et al.*, (1964). Egg masses were kept in plastic jars until hatching.

Insecticides Used:

These compounds were evaluated in laboratory tests against the second and fourth instar larvae of *S. littoralis*.

Test Procedures:

A weighted amount of Dipel DF was prepared in small doses according to the recommended rates 200 gm./feddan for Dipel DF.

Dipel, 6.4 % DF a selective bacterial insecticide containing 32×10^6 I.U. of *B. thuringiensis* var. *kurstaki* / gm. of product.

Jojoba, *Simmondsia chinensis* leaves were left to dry at room temperature (26 ± 2 °C) for one month. The dried leaves were grounded to a fine powder and extracted consecutively in a Soxhlet apparatus using ethanol solvent.

Crude extracts were dried and filtered over anhydrous sodium sulfate and were subjected to remove the solvents used in the extraction. All solvents used were previously redistilled using fraction column distillation. All the crude extracts obtained were kept in the freezer until bioassay.

Dursban insecticide (Chlorpyrifos) 48% E.C: The chemical name is *O,O*-diethyl *O*-3,5,6-trichloro-2-pyridyl phosphorothioate. The rate of application was 1000 cm³ / fed. Three concentrations (1, 1.5 and 2%) of tested materials.

Experimental Design:

The experiment was conducted according to the recommendations for agricultural pests, Ministry of Agriculture and Land Reclamation (1993) and Mohamed *et al.*, (2001).

The castor leaves dipped in only water solution and used as control. The exposure of 2nd and 4th instar larvae to the three compounds depended upon the larval feeding for 48h on treated leaves with these. Three replicates for each treatment each have 20 larvae for studying the latent effect, feeding introduced to the rest alive every 2 days until pupation.

Mortality count was recorded every 2 days then mortality percentage was calculated, developmental effect against both pupae and moth emergency were studied by recording total numbers of formed pupae and adult emergency for each treatment then calculating their percentages by the described method by El-Sisi and Farrag (1989) as the following:

Statistical Analysis:

The percentage of larval mortality were corrected according to Abbott's formula (Abbott, 1925).

$$\text{Percentage of Pupation \%} = \frac{\text{No. of formed pupae}}{\text{Initial No. of 2nd or 4th instar larvae}} \times 100$$

$$\text{Percentage of moth emergency \%} = \frac{\text{No. of formed moth}}{\text{Initial No. of 2nd or 4th instar larvae}} \times 100$$

RESULTS AND DISCUSSION

Toxicity and latent effect of commercial formulation of *Bt* Diple DF, jojoba oil and Chlorpyrifos were evaluated against the 2nd and 4th larval instars of *S. littoralis* at different concentrations, the obtained results are summarized and illustrated in Tables (1 and 2). The rates of corrected cumulative larval mortality ranged between 50 to 87 %, 30 to 45 % and 95 to 100 % for concentrations of 1-2 ml /100 ml water of the diple DF, jojoba oil and Chlorpyrifos, respectively against 2nd larval instar after 8 days post-exposure. While against the 4th larval instars of *S. littoralis* were 23 to 47 %, 11 to 21 % and 89 to 100 % for concentrations of 1-2 ml /100 ml water of the Diple DF, jojoba oil and Chlorpyrifos respectively. Careful examination of the above-mentioned data revealed that a variation in the effectiveness of the selected compounds against the different instar larvae of *S. littoralis* was increased with increasing the concentration and exposure time and decreased by increasing the insect instar, these findings were in accordance with those reported by (El-Sabagh *et al.*, 2017).

A significant reduction in the pupation and adult emergence percentages were achieved from the 2nd and 4th larval instars treated with different concentrations of diple DF, jojoba oil and chlorpyrifos compared with the control treatment. All the tested concentrations of chlorpyrifos yielded no pupation or adult emergence for both 2nd and 4th instars compared with untreated larvae 95, 93.4, 95 and 94.9 %, respectively table (1 and 2).

On the other hand, all the tested concentrations of Diple reduced the percentage of pupation to nearly half compared to the control, pupation percentages of 60, 50 and 30 % respectively were recorded for concentrations of 1-2 ml /100 ml water after treatment of the 2nd larval instar and 77, 66 and 53 % respectively after treatment of the 4th larval instar. Also, the percentage of adult emergence was notably decreased compared with the control, adult emergence of 46, 40 and 20 % respectively were recorded for concentrations of 1-2 ml /100 ml water after-treatment of the 2nd larval instar and 76, 65.5 and 50 % respectively after treatment of the 4th larval instar. These obtained results in the same trend recorded by El-Aw (2003) who reported that Dipel 2x significantly reduced pupae survival after treating *S. littoralis* larvae with *B.t.* products.

The percentage of pupation and adult emergence resulting from larvae treated with different concentrations of jojoba oil (1-2 ml /100 ml water) for 2nd instar were highly reduced (65, 55 and 55%) and slightly decreased for 4th instar (88, 85 and 79%) compared to untreated larvae. Also, treatment of both 2nd and 4th instar larvae with all jojoba oil (1-2 ml /100 ml water) different concentrations had reduced the percentage of adult emergence with (62, 54 and 54%) for 2nd and with (87.9, 84.3 and 73.6 %) for 4th instar larvae. The obtained results had been confirmed with Abdel-Razik and Mahmoud (2017) who stated that pupae mortality was high on the 2nd instar treatments compared with that of the 4th instars on the treatment with jojoba oil compared with control also, the percentages of emerged moths were decreased by increasing the extract concentrations. In addition, the highest mortality percentages of pupae were recorded with 1, 2 % of jojoba extract-treated on 2nd instars recording 55-65%, while it was ranged between, 88 and 79 % for 4th instar larvae at all tested concentrations.

Generally, the application of high concentrations caused a higher reduction percentage of pupation and adult emergence.

Table 1: Initial, latent and developmental effect of tested biocides against 2nd instar larvae of Cotton leafworm.

Treatments	Concentration	Mortality % after indicated days				Developmental effect	
		2	4	6	8	% Pupation	% Moth emergency
Diple DF	1.00	20.0	25.0	30.0	50.0	60.0	46.0
	1.50	20.0	36.0	56.0	65.0	50.0	40.0
	2.00	35.0	40.0	66.0	87.0	30.0	20.0
Jojoba	1.00	10.0	15.0	24.0	30.0	65.0	62.0
	1.50	20.0	30.0	39.0	42.0	55.0	54.0
	2.00	23.0	35.0	44.0	45.0	55.0	54.0
Chlorpyrifos	1.00	40.0	70.0	83.0	95.0	00.0	00.0
	1.50	50.0	78.0	90.0	100.0	00.0	00.0
	2.00	66.0	80.0	98.0	100.0	00.0	00.0
Control		0.0	0.0	1.0	3.0	95	93.4

Table 2: Initial, latent and developmental effect of tested biocides against 4th instar larvae of Cotton leafworm.

Treatments	Concentration	Mortality % after indicated days				Developmental effect	
		2	4	6	8	% Pupation	% Moth emergency
Diple DF	1.00	9.0	13.0	19.0	23.0	77.0	76.3
	1.50	14.0	20.0	23.0	33.0	66.0	65.5
	2.00	18.0	23.0	29.0	47.0	53.0	50.0
Jojoba	1.00	5.3	7.0	9.0	11.0	88.0	87.9
	1.50	6.0	9.0	12.0	15.0	85.0	84.3
	2.00	10.0	12.0	16.0	21.0	79.0	73.6
Chlorpyrifos	1.00	25.0	57.0	70.0	89.0	00.0	00.0
	1.50	37.0	62.0	83.0	92.0	00.0	00.0
	2.00	49.0	75.0	90.0	100.0	00.0	00.0
Control		0.0	3.0	4.0	4.6	95.0	94.9

The results in Table 1 indicate the toxicity and latent effect against 2nd instar larvae while table 2 indicates the toxicity and latent effect against 4th instar larvae of the cotton leafworms, there is a positive relationship between concentration & mortality, also the mortality increased with the increasing of the period. On the other hand, there is a negative relationship between concentrations and pupation percentage, there is an increasing pupation percentage with a low concentration. Finally, chlorpyrifos showed higher toxic & latent effects against both larval stages followed by Diple Df and Jojoba compared with control.

These results are in agreement with. Osman and Mahmoud (2009) mentioned that Dipel 2x, BioFly, Agrin, BioGaurd, Spinosad, Neemix, Mectin and Match provided higher mortality in the first instar larvae of *Spodoptera littoralis* compared to the third and fifth instar larvae, although Match, Mectin and Spinosad showed also excellent efficacy against the third larval stage at all tested concentrations.

The increased mortality percentages by increasing the concentrations of Dipel 2x agree with those previously reported by: Kareset *al.*, (1992) on larvae of the cabbage worm *Artogeia rapae* when testing Bactospeine; when he tested Dipel 2x, Ecotech bio and MVP₁₁ against *S. littoralis* and the potato tuber moth *Phthorimaea operculella*; where also Ecotech bio and MVP₁₁ were more effective than Dipel 2x against the second and fourth larval instars of *S. littoralis*, El-Khawas (2000) on the olive leaf moth, *Palpita unionalis* larvae by using the bioinsecticide.

Also, El-khayat *et al.* (2012), reported that the second instar larvae reflected a higher level of susceptibility towards all the tested insecticides that included: Insect growth regulators (Nomolt 15% Mimic 24% and Runner 24%); Bio-insecticides, Tracer, XDE and Dipel 2x; and Organophosphorus (Chlorpyrifos) than the fourth one. They found that LC50 and LC90 values, chlorpyrifos was the most effective insecticide recorded 0.1 and 0.809 ppm for 2nd instar larvae and 0.472 and 6.838 ppm for 4th instar larvae, respectively, while, tebufenozide appeared to be the least effective compound against both tested instars that gave 9.901 and 36.447 ppm against 2nd instar, and 65.736 and 1000.775 ppm against the 4th one, respectively. They reported that the rest compounds gave moderate effects in this respect.

Biological Parameters:

Data presented in tables (3 and 4) showed the efficacy of dipel DF, jojoba oil and Chlorpyrifos on the biological aspects (larval, pupal and adult emergency) of 2nd and 4th instar larvae of *S. littoralis*.

Treatment of the 2nd and 4th instar larvae with the tested compounds led to variable effects on the mean larval and pupal durations. As shown in table (3), treatment with a certain jojoba oil concentration (1ml /100 ml water) prolonged the mean larval duration (14 days) compared to the untreated check (13 days). On the other hand, treatment with the other tested compounds led to a decrease in the mean larval duration (12-8 days) compared to the tested control (13 days) of the 2nd instar. Regarding pupal duration, results in a table (3) indicated that all Chlorpyrifos concentrations (1-2 ml /100 ml water) were significantly shortened (0 days) compared to control (9 days) of the 2nd instar larvae. Also, all the rest treatment concentrations shortened pupal duration (6-8 days) compared to control (9 days) of the 2nd instar larvae except jojoba oil concentration (1 ml /100 ml water) on a par with the control.

Results are given in table (4) recorded that dipel DF prolonged the mean larval duration (7-9 days) of the 4th instar larvae in comparison with the control (7 days), while it was nearly (5-7 days) for the rest of the treatments concentrations. As for pupal duration results in a table (4) revealed that it was shortened treatments of the 4th instars compared with control (9 days) except jojoba oil concentration (1 ml /100 ml water) on a par with the control. It is clear that larval and pupal durations were decreased with the increase of concentration. At compound concentrations except for Jojoba (larval duration recorded 6 -7 at 1- 2 conc. as compared with control.

Results in Tables 3 & 4 indicate larval duration, % mortality of larvae, pupation percentage, pupation period & % of moth emergency in both 2 larval stages 2nd and 4th instar larvae. Results indicated that DipelDf is the most increased in larval and pupation duration followed by Jojoba, while Chlorpyrifos is the last one compared by the control

Also, El-khayat *et al.* (2012), reported that the second instar larvae reflected a higher level of susceptibility towards all the tested insecticides that included: Insect growth regulators (Nomolt 15% Mimic 24% a Runner 24%); Bio-insecticides, Tracer, XDE, and Dipel 2x; and Organophosphorus (Chlorpyrifos) than the fourth one. They found that LC50 and LC90 values, chlorpyrifos was the most effective insecticide recorded evidently favored by the cotton leafworm and severe damage is annually caused to most crop growers. However, the increasing consumption of synthetic pesticides in developing countries has led to a number of problems such as environmental pollution, adverse effects on non-target organisms and the development of insect resistance. Progress has been done during the past three decades to develop novel compounds affecting development such as respiration (difenthiuron) and activating the acetylcholine receptor (neonicotinoids) or the GABA receptor (avermectins) (Horowitz and Ishaaya 2004). Neonicotinoids interact with nicotinic acetylcholine receptors at the central and peripheral nervous system, resulting in

excitation and paralysis, followed by death. Neonicotinoids of potential use in agriculture are imidacloprid, acetamiprid, and thiamethoxam. Microbial insecticides such as *Bacillus thuringiensis* have been reported to provide inadequate control of *S. littoralis* and prime candidates for use in integrated Pest Management Program products. After 48h., the treated leaves were replaced by another untreated one and the larvae feed on it until the pupation. Three replicates consist of forty larvae for each concentration of tested series concentrations for any of the three tested compounds for each 2nd or 4th instar larvae were utilized in the treatment and control. Also, the observed malformations were recorded and photographed.

Table 3: Biological activity of Diplel DF, Jojoba and Chlorpyrifos against the 2nd instar larvae of *S. littoralis*

Treatments	Concentration	Developmental effect				
		Larval duration (days)	% Larval mortality	% pupation	Pupation duration (days)	% Moth emergency
Diple DF	1.00	12.0	55.0	40.0	08.0	36.0
	1.50	10.0	65.0	30.0	07.3	29.0
	2.00	08.0	87.0	10.0	06.0	7.0
Jojoba	1.00	14.0	30.0	65.0	9.0	62.0
	1.50	13.0	42.0	55.0	08.0	54.0
	2.00	10.0	45.0	55.0	08.0	54.0
Chlorpyrifos	1.00	08.0	95.0	00.0	00.0	00.0
	1.50	08.0	100.0	00.0	00.0	00.0
	2.00	08.0	100.0	00.0	00.0	00.0
Control		13.0	3.0	95.0	9.0	93.4

Table4: Biological activity of Diplel DF, Jojoba and Chlorpyrifos against the 4th instar larvae of *S. littoralis*..

Treatments	Concentration	Developmental effect				
		Larval duration (days)	% Larval mortality	% Pupation	Pupation duration (days)	% Moth emergency
Diple DF	1.00	9.0	23.0	77.0	08.0	76.3
	1.50	9.0	33.0	66.0	07.3	65.5
	2.00	7.0	47.0	53.0	06.0	50.0
Jojoba	1.00	6.0	11.0	88.0	9.0	87.9
	1.50	6.0	15.0	85.0	08.0	84.3
	2.00	7.0	21.0	79.0	08.0	73.6
Chlorpyrifos	1.00	7.0	89.0	00.0	00.0	00.0
	1.50	7.0	92.0	00.0	00.0	00.0
	2.00	5.0	100.0	00.0	00.0	00.0
Control		7.0	4.6	95.0	9.0	9.0

Acknowledgment

My sincerest appreciation and deep gratitude to the Soul of our Great Prof. Dr. Essam Mohamed Mohamed, Professor of Pesticide and Agricultural head of Sides Agric. Res. Station, at Bani Sweif for his guidance to work and his kind help.

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