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Comparative Studies, Under Laboratory Conditions, of Four Selected Insecticides on Pink Bollworm, *Pectinophora gossypiella* (Saund.)

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ABSTRACT

The insecticidal activities of four insecticides (i.e. synthetic pyrethroid "lambdathrin"; indoxacarb "steward"; mineral oil "masrona" and emamectin benzoate "proclaim") were evaluated on the 1^{st} instar larvae of the pink bollworm, *Pectinophora gossypiella*, as well as determining the biochemical changes and histological effects in 10-day treated insects after treatment with LC_{50s} of aforementioned insecticides. Lambdathrin was the most toxic insecticides followed by proclaim, then steward whereas masrona was the least toxic one. The corresponding LC_{50} values were 1.0097, 4.0423, 300.47, and 69917.34 ppm, respectively. Fluctuated biochemical changes were achieved. Total soluble protein content decreased insignificantly after treatment with lambdathrin, masrona, and proclaim. Otherwise, treatment with steward caused a significant increase in the protein content by 85.5% than control. For total lipids content, steward caused an insignificant decrease in the total lipids content by 44.62%. While lambdathrin and masrona caused a significant decrease by 50.78 and 72.58%, respectively. Also, it is noticeable that proclaim caused a significant increase in the total lipids content by 145.19-fold than control. For the determination of the changes in the AST and ALT activity, lambdathrin caused a significant increase in the AST activity by 99.93% and a significant decrease in the ALT activity by 57.12%. Steward activated both of the AST and ALT activities over control by 308.41 & 151.77%, respectively. Masrona didn't affect AST activity. while it caused a significant decrease in the ALT activity by 64.49%. On the other hand, treatment with proclaim didn't affect both AST and ALT activity.

According to histological effects, signs of intoxication have begun on the level of the mid-gut. Treated larvae showed morphologically, malformation, and destruction of the epithelial columnar cells, vacuolization, and sometimes detachment of the basement and peritrophic membranes of the epithelial cells after lambdathrin, steward, masrona and proclaim treatments. At the level of masrona and steward, treatments exhibited obvious damage to the boundaries of epithelial cells leading to disruption and fusion in the columnar epithelium cells and necrotic epithelium in some regions. On the other hand, larvae treated with both lambdathrin and proclaim produced less damage of the epithelial columnar cells. However, in both masrona and proclaim treatments, microvilli of brush border appeared to be normal.

INTRODUCTION

Pink bollworm (PBW), *Pectinophora gossypiella* (Saunders), is a major pest of cotton that damages squares, flowers, and bolls, and so it is being one of the most destructive pests in many of the major cotton-growing regions of the world including Egypt. Larvae burrow into bolls, through the lint, to feed on seeds, resulting in severe quantity and quality loss of cotton yield. To bypass losses and increase the yield, chemical insecticide application is extremely important (Aslam *et al*., 2004). Pyrethroids insecticides represented the major elements for controlling these pests for many years. To overcome problems of using chemical insecticides, currently, new groups of chemical compounds are being tested against lepidopteran pests such as indoxacarb (steward 15% SC), mineral oil (masrona 85% EC), and emamectin benzoate (proclaim 5% SG). Steward is a new insecticide that exhibits broad-spectrum activity against lepidopterist pests. When pest species are exposed to a toxic dose of steward, there is a rapid cessation of feeding within 1-4 hours and knockdown occurs within 1-2 days (Mitchell, 1999). Also, petroleum-based mineral oils like masrona have been used for insect pest control for over a century (Agnello, 2002). Proclaim a new bioinsecticide is modified isolation of the soil microorganism, *Streptomyces avermitilis*. It acts through stimulating the releasing of γ-aminobutyric acid (GABA), an inhibitory neurotransmitter, accordingly causing paralysis to insect, and therefore stop feeding within hours of ingestion (Anonymous, 2003).

The changes in the biochemical content especially total soluble protein and total lipids contents, as well as the transaminase enzymes activities such as alanine transaminase (ALT) and aspartate transaminase (AST) activities, have an important role in insects biological and physiological activities (Mead-Hala, 2000 and Khedr, 2002). Some biochemical effects were studied by many authors on mineral oils (Khatter & Abuldahab, 2010 and Abo El-Ghar *et al.*, 1995) and on emamectin benzoate (Abou-Taleb *et al.*, 2009). Also, the histological studies are very important to do on the midgut of the treated larvae because they show and explain the changes resulting from treating larvae with different tested compounds. Histopathological effects induced in the midgut for these pesticides were also examined by many authors on pyrethroids (Schouest *et al*., 1986; Younes *et al*., 2002 and Omar *et al*., 2006); by Youssef (2006) and Mouharib (2009) on Emamectin and by Hassan (2009) on steward.

The aim of this work is to give a spot of light on some toxicological, biochemical, and histopathological effects that may occur as a result of treating larvae of *P. gossypiella* with four insecticides differed in their mode of action which could use actively in sequence insecticides programs in order to help in delaying resistance development and controlling this serious pest.

MATERIALS AND METHODS

Insect:

The laboratory strain of newly hatched larvae of the pink bollworm (PBW), *P. gossypiella*, used in this study was obtained from a laboratory colony of Bollworms Research Department, Plant Protection Research Institute; Agricultural Research Center (ARC), reared for several generations away from any contamination with insecticides and maintained at 26 ± 1ºC temperature, 70 ± 5% RH. The pink bollworm larvae were reared on a modified artificial diet as described by Rashad & Ammar (1985).

Insecticides*:*

Four insecticides were selected against *P. gossypiella* for the research. The insecticides viz., lambada cyhalothrine (lambdathrin 5% EC), indoxacarb (steward 15% EC), mineral oil (masrona 85% EC), and emamectin benzoate (proclaim 5% SG) were obtained from their respective manufacturers and used in the present studies.

Toxicological Studies:

The study the toxicity of the four tested compounds against newly hatched larvae of *P. gossypiella*, a stock solution of the tested insecticides was prepared by diluting with distilled water to obtain serial aqueous concentrations as follows:

1-0.6250, 0.1563, 0.0782 and 0.0390 ppm of lambdathrin.
2-3750, 1875, 937.5, and 468.75 ppm of steward.
3-170000, 85000, 42500, and 21250 ppm of masrona.
4-10, 5, and 2.5 ppm of proclaim.

Five grams of diet folded into 3 Petri dishes (7 cm in diameter) as replicates, then 1 ml of each insecticide sprayed homogeneity on the surface of the diet. Another group sprayed with an equal volume of distilled water was used as control. Ten PBW newly hatched larvae were placed on the surface of the diet using a soft brush. one hour later the exposing all alive larvae were transferred individually to glass vials (2 X 7 cm) containing a small piece of the normal diet. Vials were plugged with absorbent cotton and incubated at 27 ± 1°C and 75 ± 5% RH. Larval mortalities were recorded after 24 hours of treatment. Percentages of mortalities were corrected according to Abbott's formula (1925) and the values of the LC_{50} were calculated according to Finny (1971). Based on LC_{50} values the toxicity indices are calculated according to Sun (1950).

Biochemical Studies:

1. Sample for Biochemical Assay*:* to access the latent biochemical effects of the tested insecticides, newly hatched larvae (≈100 larvae) of PBW were allowed to feed on an artificial diet containing LC_{50} of the tested insecticides for one hour, and then transferred to feed on the control diet. Ten days after treatment, batches of treated larvae were homogenized in distilled water using a Teflon homogenizer. The homogenates were centrifuged at 500 rpm for 10 minutes at 5º C. The supernatants were immediately assayed to determine the biochemical analysis under the study. Untreated larvae were used as control.

2. Technique of Biochemical Analysis: Colorimetric determination of total soluble protein in the total homogenate of *P. gossypiella* larvae was estimated by the method of Gornall *et al*. (1949). The total lipids were estimated according to the method of Schmit (1964). Aspartate transaminase (AST) and alanine transaminase (ALT) enzyme activities were determined colorimetrically according to the method of Reitman & Frankle (1957).

Histological Studies:

Fresh specimens of 10-days old *P. gossypiella* larvae surviving after-treatment of the 1^{st} larval instar with the LC_{50} values of the tested compounds were selected. Larvae were fixed in aqueous Bouine`s fluid, placed in xylene, embedded in paraffin wax, and stained with Ehrlich`s haematoxylin counter and Eosin and then mounting in Canda balsam. To compare the histological changes, specimens from the control were sampled and prepared as the previously described manner. All preparations of the histological examination were examined with a microscope and photomicrographs were taken.

Statistical Analysis:

Toxicological data were statistically calculated through a Proban program, software computer program (Jedrychowski, 1991). Analysis of variance (ANOVA) was conducted using Costat program software computer. Significant differences were determined according to LSD values of Duncan’s multiple range test (Duncan, 1955).

RESULTS AND DISCUSSION

Toxicological Studies:

The susceptibility of *P. gossypiella* larvae to lambdathrin, masrona, steward and proclaim was indicated in Table (1). The obtained resulted revealed that the tested insecticides varied in their efficiencies against PBW larvae. In addition, mortality percentages increased with the increasing of the used concentration. The corrected mortality percentages after 1 hour of treatment with lambdathrin ranged between 13.46-43.53%. The corresponding figures were 56.49-82.32; 24.33-69.81 and 33.75-78.54% for steward, masrona and proclaim, respectively. Graphically illustrated concentration-mortality regression lines (Figure 1) confirmed the same results as a positive relationship between the used concentrations of tested insecticides and the percentages of mortality. According to the LC_{50} values, lambdathrin was the most toxic insecticide against the 1st instar larvae of *P. gossypiella* (Ti = 100); followed by proclaim (Ti = 24.98) then steward (Ti = 0.0060), whereas masrona (Ti = 0.0014) was the least toxic one. The corresponding LC_{50} values were 1.0097, 4.0423, 300.4718, and 69917.35 ppm, respectively. Figure (1) also confirmed the same results where the toxicity line of lambdathrin was the nearest line to the vertical axial, meanwhile, the line of masrona was the farthest one. So, the present results verified that the lambdathrin, a conventional insecticide, is most effective than other insecticides in controlling PBW. These results are in agreement with those of Abdel-Hafez & Osman (2013); El-Zahi (2013); El-Dewy (2013) and Hanafy & El-Sayed (2013), and not in agreement with those of Gupta *et al*. (2004) who stated that emamectin benzoate had a highest relative toxicity (6.93) then indoxacarb (1.62) while lambda cyhalothrine had the least relative toxicity (0.19) on *S. litura*. Also, Ali *et al.* (2015) and Saleh *et al.* (2015) proved that emamectin benzoate was more toxic to *S. littoralis* than the conventional insecticide. Also, the recent results showed that proclaim followed lambdathrin in the toxicity, while steward and masrona were the least effective ones. This result is confirmatory to those of Ahmad *et al.* (2005) who reported that emamectin benzoate was more toxic than indoxacarb to *S. litura.* In addition, proclaim proved to be safer for beneficial insects as compared to conventional insecticides (Udikeri *et al*., 2004 and Abdullah *et al*., 2019), so it could have a potential role for the management of *P. gossypiella.*

On the other hand, data of the probit analysis shown in Table (1) revealed that proclaim treated insects had the highest slope value, which means that all populations are susceptible to this insecticide (Martínez-Carrillo, 1998). In addition, toxicity lines which graphically illustrated in Figure (1) differed in their slope, so that means these insecticides differed in their mode of action. This is helpful in resistance management whereas using effective pesticides with different modes of action in rotation or sequences could help in delay resistance development. Pyrethroids (lambdathrin) are considered axonic poisons that affect the nerve fiber through binding with the protein which controls the voltage-gated sodium channel. (Ali *et al*., 2015). Indoxacarb (steward) represents a new class of insecticides (the oxidiazines). It exhibits a novel mode of action where it blocks the movement of sodium ions into certain ion channels in the nerve cell, resulting in paralysis and death of the targeted pest species (Wise *et al*., 2006). So, its mode of action is the opposite of pyrethroids, whereas it delays the closing of sodium channels resulting in the prolonged transmission of nerve signals (Soderlund & Knipple 1995).

Mineral oils, like masrona, have different modes of action. It could block insect spiracles, causing die from asphyxiation; act as insect poisons by interfering with the fatty acids, and/or interacting with normal metabolism, and it may also disrupt how an insect feed. (IPM guidelines, 2017). Emamectin benzoate (proclaim) a new bioinsecticide was isolated from the fermentation of soil microorganism *S. avermitilis*. The mode of action involves stimulation of high-affinity γ-aminobutyric acid (GABA) receptors consequently increase the permeability of the membrane to chloride ion leading to paralysis (White *et al.*, 1997 and Yen & Lin, 2004).

Table 1: Toxicity of tested insecticides after 1 hour following treatment against the newly hatched larvae of *P. gossypiella*

Concentration (ppm)		Corrected mortality%	LC_{50} (95% confidence limits)	Slope ± SE	Toxicity index (Ti)
Lambdathrin	0.0390 0.0782 0.1563 0.6250	13.46 19.23 26.30 43.53	1.0097 (0.3315-1.4367E+07)	0.7823 ± 0.3589	100
Steward	468.75 937.5 1875 3750	56.49 66.21 74.95 82.32	300.4718 (0.0729-680.9962)	0.8462 ± 0.3686	0.3360
Masrona	21250 42500 85000 170000	24.33 38.56 54.54 69.81	69917.3469 (45791.501-1.1845E+05)	1.3452 ± 0.3613	0.0014
Proclaim	2.5 5 10	33.75 57.36 78.54	4.0423 (1.9308-6.1296)	2.0094 ± 0.7090	24.98

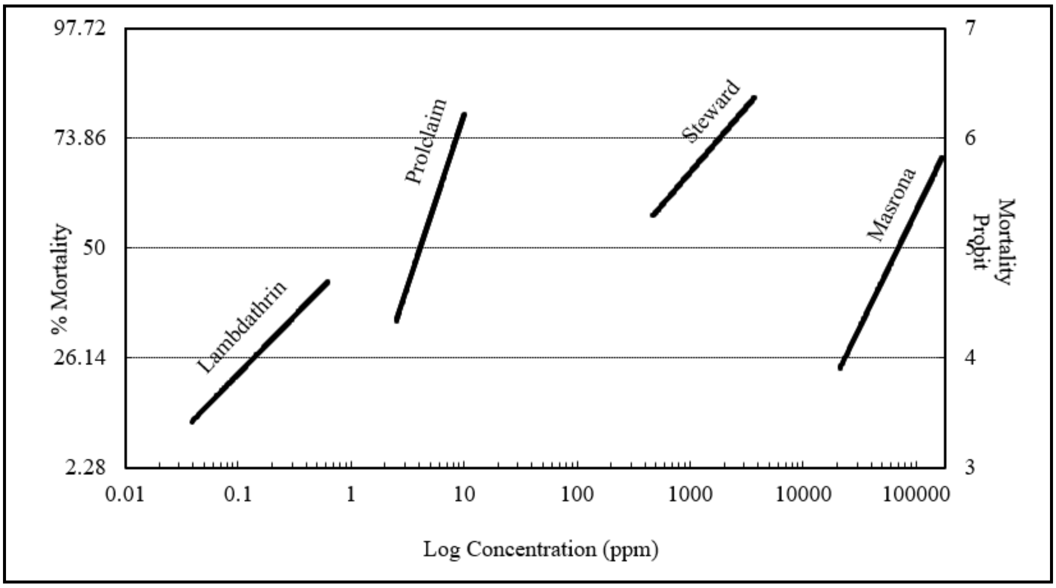

Fig. 1: Concentration log probit toxicity lines of tested compounds against newly hatched larvae of *P. gossypiella* after 1 hour following treatment.

Biochemical Studies:

Insecticides understudying have a good field activity on a number of lepidopterous insects (Wing *et al.*, 2000, Agnello, 2002, Argentine *et al.*, 2002 and Megahed *et al.*, 2013). Many toxicants have latent effects at high concentrations (Van Eck, 1979), so the LC_{50s} of the tested insecticides were used to measure the biochemical changes in the 10-days old larvae of *P. gossypiella* following treatment. Studying the biochemical changes in insects after exposure to insecticides may give some hints about their mode of action.

Total Soluble Protein Contents:

The main metabolites are total proteins, total carbohydrates, and total lipids. Any change in its balance induces confusion in the sequence of metamorphosis and metabolism (Cespedes *et al*., 2005 and Pavela, 2005). As shown in Table (2) and Figure (2), total soluble protein content in the control PBW was 9.01 mg/dl. As a result of treatments by lambdathrin, masrona and proclaim, the protein content decreased insignificantly. Means were 8.02, 7.08 and 7.007 mg/dl, respectively. Total proteins content decreased significantly in *S. littoralis* after treatment with mineral oil Kemesol 95% (Khatter & Abuldahab, 2010), and emamectin benzoate "Proclaim" Megahed *et al*. (2013); and in *P. gossypiella* after treatment with vertimec, a member of avermectin family, (Ahmed, 2015). It could be attributed to proteins bindings with foreign substances (Ahmed & Forgash, 1976) or the breakdown of the protein into AA then enter them to Krebs cycle as Keto acids (Shekari *et al*., 2008). Varied results were found by Dahi *et al* (2009) who found that treatment of 4th instar larvae of *S. littoralis* with avermictin increased protein content at dose 0.003 ppm. Also, El-Didamony (2012) indicated that the treatment of newly hatched larvae of *E. insulana* with proclaim increased total soluble protein than control.

Otherwise, treatment with steward caused a significant increase in the protein content by 85.5% than control. Broadway & Duffey (1986) mentioned that could be due to the post-ingestive compensatory mechanisms in insects, like the secretion of additional proteases. Recent results agree, to some extent, with the results of Gamil *et al*. (2011) who showed that avaunt (Indoxacarb 15% EC) caused a slight increase in the total protein of *S. littoralis* by 8.7% in 2nd instar and decreased by 24.9% in 4th instar.

Total Lipids Contents:

Treated newly hatched larvae of *P. gossypiella* with steward caused an insignificant decrease in the total lipids content by 44.62%. While lambdathrin and masrona caused a significant decrease than control by 50.78 and 72.58%, respectively; otherwise they have not differed significantly from steward. The representing means were 17.97, 20.35, and 10.01 mg/dl for lambdathrin, steward and masrona, respectively compared to 36.51 mg/dl for control. Also, it is noticeable that proclaim caused a significant increase in the total lipids content by 145.19-fold than control. Similar results were achieved by El-Didamony (2012) in *E. insulana* after treatment with proclaim.

In addition, results of Ahmed (2015) revealed that vertimec increased significantly in total lipids in the 4th instar larvae of *P. gossypiella* than control by 96.35%. In the study of Khatter & Abuldahab (2010), Kemesol 95% increased the haemolymph and fat body lipid contents of the treated larval stage of *S. littoralis.* They mentioned that might be due to increasing of the conversion rate of carbohydrate to lipid. Gamil *et al.* (2011) found that avaunt (Indoxacarb 15% EC) decreased lipids in *S. littoralis* by 82.7% in 2nd instar and by 67.5% in the 4th instar.

Table 2: Effect of LC_{50} of tested compounds on total soluble proteins and total lipids content (mg/dl) of *P. gossypiella* larvae

Treatment	Total Soluble Proteins Content		Total Lipids Content	
	Mean ± SD	%Change	Mean ± SD	%Change
Control	9.010^{b} ± 0.985	—	36.51^{b} ± 3.51	—
Lambdathrin	8.020^{b} ± 1.700	-10.99	17.97^{c} ± 1.74	-50.78
Steward	16.72^{a} ± 1.460	+85.57	20.35^{bc} ± 1.08	-44.26
Masrona	7.080^{b} ± 1.025	-21.42	10.01^{c} ± 1.02	-72.58
Proclaim	7.007^{b} ± 0.590	-22.23	89.52^{a} ± 19.91	+145.19
LSD (5%)	**2.209**		**16.56**	

Means followed by the same letter at the same column are not significantly different

$$\% \text{ Change} = 100 \times \frac{Treatment - control}{Control}$$

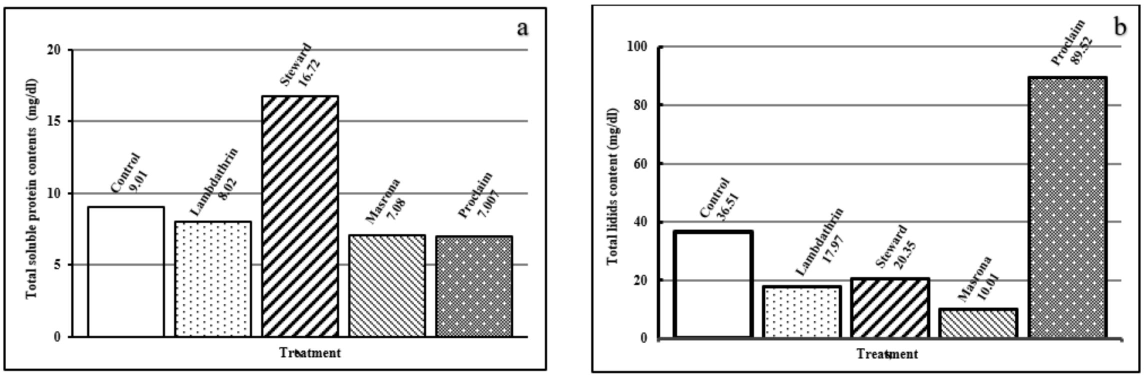

Fig. 2: Total soluble proteins (a) and total lipids (b) contents of 10-days old *P. gossypiella* larvae treated as newly hatched with LC_{50} of tested compounds.

Transaminase Enzymes (AST and ALT) Activity:

Amino acids transaminase enzymes responsible for amino acid pool balance in insects (Meister, 1957). Also, they are key enzymes in the formation of non-essential AA, in glucogenesis, and in most nitrogenous product metabolism (Tanani *et al*., 2009). Changes in its levels have been correlated with anabolism or catabolism of protein (Mordue & Goldworthy, 1973). Determination of the changes in the AST and ALT activities from control are shown in Table (3) and Figure (3). Recorded values for the transaminases activity in control insects were 14.62 & 85.30 µg pyruvate /ml, respectively. AST increased significantly by 99.93% and ALT decreased significantly by 57.12% after treatment with lambdathrin. Comparing the present results with other authors, Etebari *et al*. (2007) revealed that the biochemical effects of the chemical insecticides caused a decrease in the activity of ALT enzyme.

Indoxacarb "steward" activated both of the AST and ALT activities (59.71 & 214.76 µg pyruvate/ml, respectively). These activations were significantly over control by 308.41 & 151.77%, respectively. In accordance with our results, elevated levels in the transaminases activity achieved by Zibaee *et al*. (2011) and they attributed these elevations to the possible damages of the insecticide to hemocytes and fat bodies. On the contrary, Khaled & Farag (2015) revealed that indoxacarb caused a significant decrease in the transaminases activity in *S. littoralis* larvae. Etebari *et al.* (2005) mentioned that the reduction in transaminases activity induced by indoxacarb attributed to the hormonal control of protein synthesis and transaminases.

Table 3: Effect of LC_{50} of tested compounds on aspartate and alanine transaminase activities (µg pyruvate/ml) of *P. gossypiella* larvae

Treatment	Aspartate transaminase (AST)		Alanine transaminase (ALT)	
	Mean ± SD	%Change	Mean ± SD	%Change
Control	$14.62^{cd} \pm 1.31$	—	$85.30^{b} \pm 4.23$	—
Lambdathrin	$29.23^{b} \pm 1.98$	+99.93	$36.58^{c} \pm 1.08$	-57.12
Steward	$59.71^{a} \pm 11.54$	+308.41	$214.76^{a} \pm 17.25$	+151.77
Masrona	$20.30^{bc} \pm 2.12$	+38.85	$30.29^{c} \pm 1.71$	-64.49
Proclaim	$6.280^{d} \pm 0.88$	-57.05	$89.42^{b} \pm 8.60$	+4.83
LSD (5%)	**9.76**		**16.14**	

$$\% \text{ Change} = 100 \times \frac{Treatment - control}{Control}$$

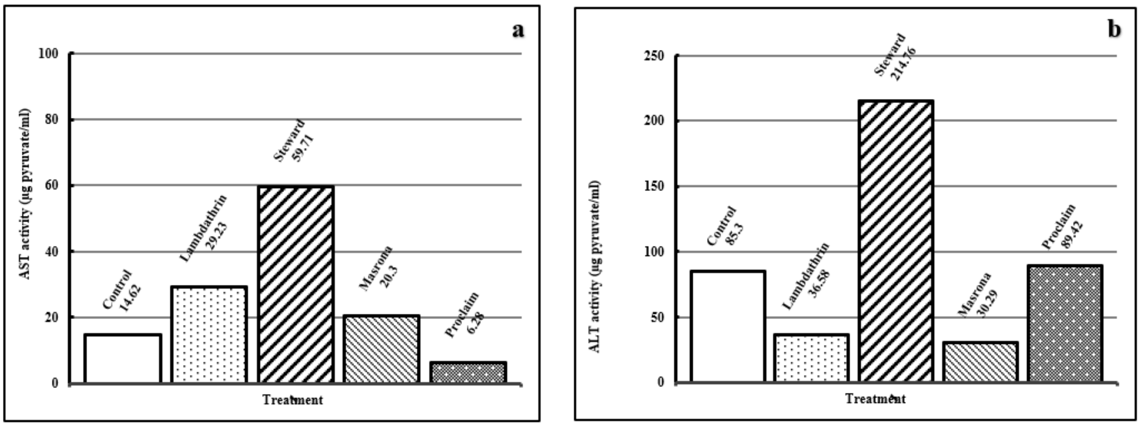

Fig. 3: Aspartate transaminase; AST (a) and Alanine transaminase; ALT (b) activities of 10-days old *P. gossypiella* larvae treated as newly hatched with LC_{50} of tested compounds.

Mineral oil "masrona" didn't affect AST activity, where it records 20.30 µg pyruvate/ml. while it caused a significant decrease in the ALT activity from control by 64.49%. on the other hand, treatment with emamectin benzoate "proclaim" induced insignificant inhibition in the activity of AST enzyme as they recorded 6.28 µg pyruvate/ml, compared to 14.62 µg pyruvate/ml for control. Otherwise, proclaim induced insignificant elevation in the ALT activity as it records 89.42 µg pyruvate/ml, meanwhile, control larvae had 85.30 µg pyruvate/ml. Rizk (1998) found that vertimec weakly affected the activities of the enzymes in total homogenates of PBW full-grown larvae (-0.26 and –3%). While proclaim increased the activity of AST and ALT enzymes in *S. littoralis* as mentioned by Abou-Taleb *et al.* (2009) and in newly hatched larvae of *E. insulana* as mentioned by El-Didamony (2012). On the contrary, El-Didamony (2012) mentioned that treating seven days old larvae of *E. insulana* with proclaim sublethal concentration decreased the activities of AST & ALT enzymes than control. So, the effect could be different according to the larval age. In this respect, Ahmed (2015) revealed that vertimec caused a significant increase in the activity of AST and ALT activity in 2nd instar larvae of PBW than control by 89.23 & 172.92%, respectively. As larvae reached full-grown, she noted that vertimec caused significant decrease in the level of AST than control by 22.5%, but didn't affect the ALT activity. Also, Rami reddy *et al.* (1992), Nath *et al. (*1997) and Singh *et al*. (1997) mentioned that there were fluctuations in the activities of transferases enzymes in silkworm larvae under different stress factors like parasitism, treatment with phosphorus pesticides and IGRs.

Histological Studies:

Histological studies are imperative to do on the larval midgut to show and explain the changes resulting from treating larvae with such insecticides. The histological structure of the midgut from untreated *P. gossypiella* larvae shows that it is composed of closely arranged columnar epithelium cells with a rather broad apex, bearing an apical regular microvilli border and every cell containing a large coarsely nucleus occupying a middle position within the cell. Also, small calyx-shaped goblet cells are seen in great numbers between the columnar cells, both types of cells rest on a basement membrane. The epithelium cells shield from the midgut lumen content by the pertrophic membrane (Fig. 4).

Indeed, the cellular damage as well as the degree of intoxication is not the same on all levels in the mid-gut and the intestinal regions. This fact seems linked to the difference between morphological and physiological cells in the intestine regions and also due to the difference in the mode of action of the tested treatments. (Lambert & Peferoen, 1992). For the treated larvae of *P. gossypiella* the signs of intoxication began on the level of the mid-gut. The histology of treated larvae on the level of the mid-gut region showed some morphological malformation and destruction of the epithelial columnar cells, vacuolization, and sometimes detachment of the basement and peritrophic membranes of the epithelial cells after lambdathrin, steward, masrona and proclaim treatments (Figs. 5 a, b, c & d, respectively). At the level of masrona and steward, treatments exhibited obvious damage to the boundaries of epithelial cells leading to disruption and fusion in the columnar epithelium cells and necrotic epithelium in some regions.

On the other hand, *P. gossypiella* larvae treated with both lambdathrin and proclaim produced less damage of the epithelial columnar cells. However, in both masrona and proclaim treatments, microvilli of brush border appeared to be normal. Insecticides with different modes of action can differ in their site of action. These compounds may affect the larvae as an acute toxicant. The histopathological effect was produced as a latent effect on the treated larvae. These results are in accordance with those of various different insect species. Tuan *et al.* (1994) showed the nuclei of the midgut, fat body, and epidermal cells were swollen after inoculation with NPV, followed by cell lysis and tissue disintegration of *S. exigua* larvae. Younes *et al*. (2002) investigated the histopathological effects induced in the midgut and integument of the 5^{th} instar larvae of cotton leafworm *S. exigua* treated as 4^{th} instar larvae with cypermethrin.

Hussein *et al.* (2002) conducted some histological and histochemical changes related to vertimec on the two economically important insect species, *P. gossypiella* and *E. insulana* in the laboratory. Data indicated that the two compounds caused destruction for the midgut epithelial cells. The histochemical studies revealed that both compounds affected the polysaccharides in the midgut epithelium. Gamil (2004) and Heba (2005) observed histological changes in *S. littroalis* mid-gut. Omar *et al.* (2006) recorded abnormalities in the mid-gut tissue of larvae of pink and spiny bollworms when treated with Chinmix and Spintor. Both treatments caused the destruction of epithelial cells in some points and separation of epithelial cells from basement membranes in the mid-gut treated larvae. Youssef (2006) observed disruption in the columnar epithelium cells, and stretching leading to tearing in the peritrophic membrane in midgut larvae of *S. littoralis* treated with abamectin and pyriproxyfen. Mouharib (2009) reported that Treating 4^{th} instar larvae of *P. gossypiella* with emamectin benzoate showed irregular cell membrane (shape), cell malformation, and disorder of the neurosecretory cells.

Generally, according to the results of this study, it could be concluded that the tested insecticides have acute toxicity, especially lambdathrin and proclaim, and latent side effects on the physiological and histological levels. This is in addition to the different modes of action. So, these insecticides can be a possible candidate to be applied in sequence insecticides programs in cotton fields.

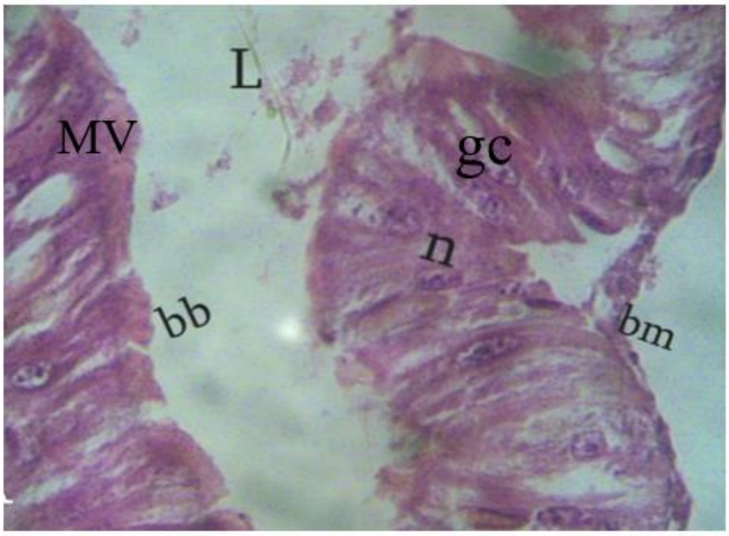

Fig. 4: Cross section in the midgut of untreated larvae of *P. gossypiella* showed a well layer of epithelial cells and lumen.

(n): Nuclei (L): Lumen of mid-gut (bm): Basal membrane
(bb): Microvilli of brush border (V): Vacuoles in mid-gut epithelial cells (gc): Goblet cell
(MV): Microvilli

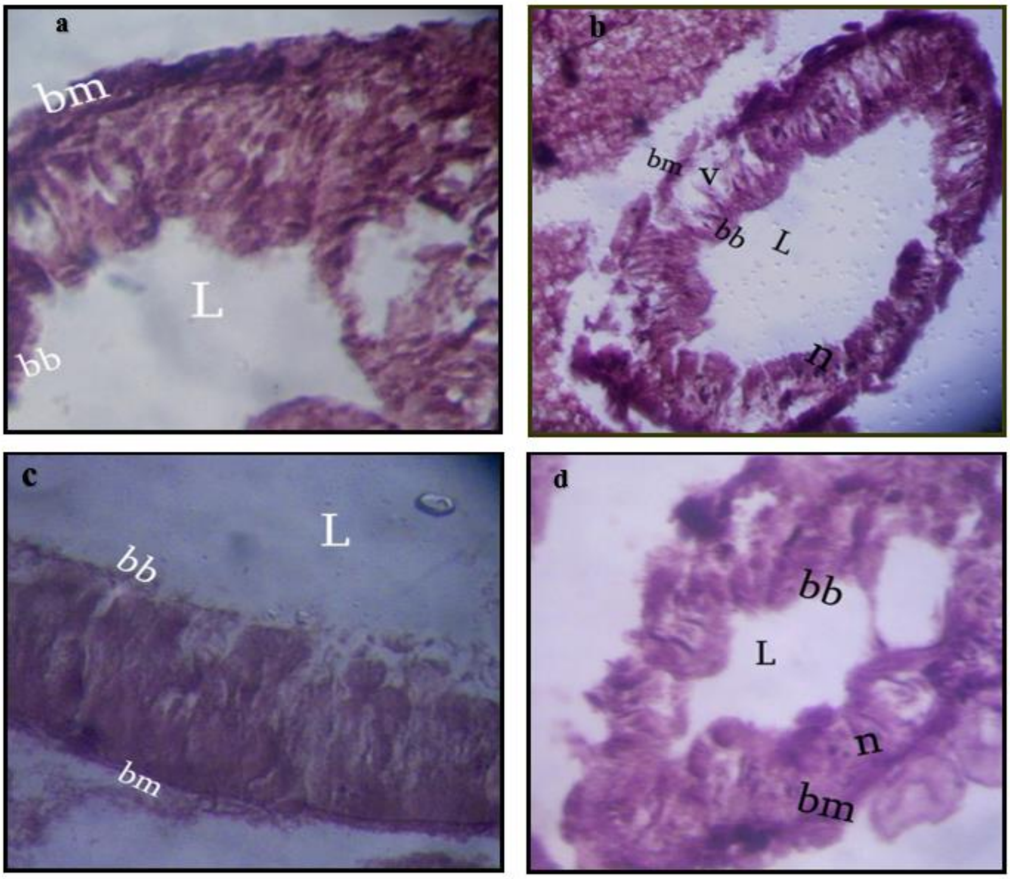

Fig. 5: Cross-section of mid-gut of *P. gossypiella* treated with LC_{50} of lambdathrin (a), steward (b), masrona (c), and proclaim (d).

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ARABIC SUMMARY

دراسات معملية لمقارنة تأثير أربع من المبيدات الموصى بها لدودة اللوز لقرنفلية

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قسم بحوث ديدان اللوز – معهد بحوث وقاية النباتات – مركز البحوث الزراعية

أجريت دراسات لتقدير سمية كلا من مركبات لامباداثرين، ستيوارد، مصرونا، وبروكلايم على العمر اليرقي الأول لدودة اللوز القرنفلية، بالإضافة للتأثيرات الفيسيولوجية والهستولوجية الناتجة عن معاملة اليرقات الحديثة بالتركيزات السامة النصفية، وقد تم تسجيل تركيزات نصف مميتة لهذه المركبات على التوالي كالتالي: 1.0097، 4.0423، 300.47 و69917.34 جزء من المليون. كذلك أوضحت الدراسات الكيميائية الحيوية انخفاضا غير معنويا في مستوى البروتين الكلي ليرقات دودة اللوز القرنفلية بعد تغذية الفقس الحديث لمدة ساعة على البيئة المعاملة بالتركيز النصف مميت لمركبات لامباداثرين، مصرونا وبروكلايم مقارنة بما هو عليه في اليرقات غير المعاملة. وعلى العكس من ذلك، فقد سبب مركب ستيوارد ارتفاع فى مستوي البروتين الكلي في اليرقات المعاملة بمقدار 85.5%. أما بالنسبة لمحتوى الدهون فقد تسبب مركب ستيوارد انخفاضا غير معنويا في مستوى الدهون الكلي بمقدار 44.62%، في حين سبب كل من لامباداثرين ومصرونا انخفاضا معنويا مقارنة بالكنترول بمقدار 50.78 و72.58%، على التوالي، وأيضا كان من الملاحظ أن البروكلايم سبب ارتفاعا واضحا في محتوى الدهون الكلي حيث بلغ 145.19 ضعف مثيله في الكنترول.

من ناحية أخرى سبب مركب لامبادثرين زيادة معنوية في نشاط انزيم AST بمقدار 99.3%، وانخفاضا معنوية في نشاط انزيم ALT بمقدار 57.12%، في حين كان تأثير مركب ستيوارد على كلا الانزيمين متشابه حيث سبب تنشيط كلا الإنزيمين بمقدار 308.41 و151.77% مقارنة بالكنترول. وعلى العكس من ذلك فلم يُسبب مركب مصرونا تأثيرا معنويا على نشاط AST، في حين سبب انخفاضا معنويا في نشاط انزيم ALT بمقدار 64.49%. هذا ولم يسجل مركب بروكلايم تأثيرا معنويا على نشاط كلا الإنزيمين.

على صعيد أخر، أدت معاملة اليرقات بالمركبات سابقة الذكر إلى ظهور تأثيرات هستولوجية في القناة الهضمية لدودة اللوز القرنفلية، فقد أظهرت العينات الهستولوجية في منطقة المعي الأوسط وجود ضعف ملحوظ في طبقة العضلات مع حدوث تهتك حاد وعدم انتظام في تركيب الخلايا الطلائية العمودية بالإضافة لفقدانها للترابط مع إحداث تلف في الغشاء المحيط بالغذاء، وكذلك لوحظ حدوث تكون لبعض الفجوات (Lesions) في طبقة الخلايا الطلائية المبطنة للمعي الأوسط لليرقات، هذا وقد اختلفت المركبات فيما بينها في مقدار هذا التأثير الحادث حيث وجد أن أكثر المركبات تأثيرا كان هو مركب لامبادثرين وستيوارد حيث سببا تكون العديد من الفجوات بالإضافة إلى حدوث تدمير حاد للخلايا الطلائية المبطنة للمعي.