

BIOCHEMICAL AND BIOLOGICAL EFFECTS OF INSECT GROWTH REGULATOR, TEFLUBENZURON ON *PECTINOPHORA GOSSYPIELLA* (SAUNDERS) (LEPIDOPTERA: GELECHIIDAE) AND *COCCINELLA UNDECIMPUNCTATA* (L.) (COLEOPTERA: COCCINELLIDAE)

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ABSTRACT: This work was conducted to study the effects of the insect growth regulator, Teflubenzuron on survival, reproduction parameters of *Pectinophora gossypiella*, (Saunders) (Lepidoptera: Gelechiidae) as well as on pupae and adults of *Coccinella undecimpunctata* (L.) (Coleoptera: Coccinellidae). As for *P. gossypiella* the LC_{25} and LC_{50} values were 19.36 and 78.59 ppm, respectively, when the newly hatched larvae treated by Teflubenzuron, while LC_{90} value was 1189.25 ppm. Teflubenzuron treatments significantly prolonged the developmental time of survived larvae and pupae, causing longer life span compared to control. Statistical analysis of the obtained data demonstrated highly significant differences between oviposition and reproductive adults resulted from treated larvae and the control. The treatment with LC_{50} (78.59 ppm) of Teflubenzuron reduced the total protein content, while the phenoloxidase content was high significantly decreased. Chitinase activity was significantly increased causing failure of the pupation process. The transaminase enzymes activity (Got and GPT) were highly increased. Regarding to *C. undecimpunctata*, there were different mortality percentages of pupal after one to 7 days of exposing to 300, 150, 75, 37.5, 18.75 and 9.375 ppm of Teflubenzuron, where mortality and malformed was increased by increasing Teflubenzuron doses. Furthermore, pupa duration of the predator was significantly prolonged by increasing IGR doses, while adult longevity and fecundity were significantly decreased. Total protein and phenoloxidase values were significantly reduced when the pupal stages were treated and adult fed on *P. gossypiella* eggs treated with LC_{50} of Teflubenzuron.

Key words: IGR, Control, Chitinase, protein, transaminase, phenoloxidase.

INTRODUCTION

The pink bollworm (PBW) *Pectinophora gossypiella* (Saunders) (Lepidoptera: Gelechiidae) is a major cotton pest in different countries and in the Arab republic of Egypt as well. Larvae attack the flower, buds and bolls of cotton causing loss in both quantity and quality of cotton yield (Anonymous, 1996).

The eleven-spot ladybird, *Coccinella undecimpunctata* (L.) is among the most abundant predators recorded in different crop in all Governorates. They are voracious predators, and combined with their high

abundance of pests may play an important role in the reduction of pest populations. Individual of *C. undecimpunctata* lack many of the characteristics suggested (assemblages) as necessary for a successful biological-control agent. They feed on a variety of prey (such as aphid, whitefly, jassid, pink bollworm and other pests). In Egypt agro ecosystems is largely unknown, and do not exhibit density-dependent tracking of prey populations. On the other hand, the use of some chemical during the presence of the predator in the field may be high affected the population,

biology, fecundity and biochemical content of the predator.

Insect growth regulators (IGR's) is considered as alternative way of synthetic insecticides for controlling *Spodoptera littoralis* (Raslan, 2002). They disrupt the physiology and development of the target insect. They tend to be selective and less toxic to non-target organisms than other pesticides (Gurr *et al.*, 1999). IGR's had been grouped in chitin synthesis inhibitors (CSIs) and interfere with the action of insect hormones such as juvenile hormone and ecdysteroids (Tunaz and Uygun, 2004). CSIs disrupt chitin biosynthesis (Gijswijt *et al.*, 1979) they prevent molting, and produce an imperfect cuticle (Hammock and Quistad, 1981). Also, they disrupt the hormonal balance (Soltani *et al.*, 1984). These compounds have no effects on parasitoids and other natural enemies (Ishaaya *et al.*, 2002). Also, it has low mammalian toxicity (Barazani, 2001).

A new approach in the insect control is to use substances that affect insect growth and development. These substances are insect growth regulators (IGR) which receiving more practical attention to provide for safe foods and clean environment. The chitin synthesis inhibitor (CSIs) introduce into the market as a novel insecticide named Teflubenzuron (TFB).

Most CSIs are mostly used as larvicides, as treated larvae develop until molting, but fail to molt due to inhibition of the synthesis of new cuticle, specifically, chitin biosynthesis.

For instance: some (IGRs), when directly applied to *Manduca* epidermal cells in vitro, inhibited endo-cuticular deposition (Miyamoto *et al.*, 1993). Moreover, chitin precursors of *Pieris* larvae (14C-glucose), *Manduca* larvae (14 C-glucosamine), *Mamestra* larvae (14 C-acetylglucosamine) and *Spodoptera* larvae (14-C-UDP-N-acetylglucosamine) were not incorporated into chitin in the presence of chitin synthesis

inhibitors. The role of insect hormones in reproduction has been extensively reviewed (Prabhaker and Toscano, 2007).

From the previous review, the aim of this work is to evaluate the effect of the insect growth regulator Teflubenzuron (TFB) on the pink bollworm (PBW) *Pectinophora gossypiella* and the eleven-spot ladybird *Coccinella undecimpunctata* (L.)

MATERIALS AND METHODS

Insect growth regulators (IGR) used:

Common name: Teflubenzuron (benzoylurea)
Trade name: Nomolt ® 15% Suspension Concentrate (SC).

Rate of application: 50 cm³ / 100 L.

Basic product: BASF Co.

Tested insects:

The susceptible laboratory strain of pink bollworm, *P. gossypiella* was reared for several generations under the laboratory conditions at 26±1°C and 75±5 R.H% at Bollworms Research Department, Plant Protection Research Institute, Agriculture Research Center, while the eleven-spot ladybird, *Coccinella undecimpunctata* (L.) predator stages were collected from the experimental farm of faculty of agriculture, Menoufia University at Shebin Elkom, Menoufia Governorate, from cotton field during 2015 season. Different eggs, larvae, pupae and adult stages of the predator were collected by hand and sweeping net and sent to the laboratory for the experiments as field strain.

Determination the toxicity on newly hatched larvae of *P. gossypiella*:

The toxicity of the tested compound against newly hatched larvae of *P. gossypiella* were studied. Different concentrations of the tested compound were sprayed on the upper surface of 15 g of artificial diet placed in Petri-dish (9cm in diameter). Six concentrations of tested compound IGR were prepared (300, 150,

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75, 37.5, 18.75 and 9.375 ppm).

Fourteen newly hatched larvae of the pink bollworm were placed on the surface of the treated diet using a soft brush. Another group of Petri-dishes was prepared containing the same diet and sprayed only with distilled water (used as control) and 60 larvae were placed on their surfaces. (three replicates for each treated and untreated were used) Larvae were allowed to feed on the tested diets for 24hr, and transfer to untreated diet. Afterwards larval mortalities were recorded after 24hr, 3 and 7 days of treatment.

LC₅₀ of newly hatched larvae were calculated, and larvae were allowed to feed on LC₅₀ treated diet (three replicates for each assay). After that, the survivors in each assay were counted and transferred to glass vials (2 x 7cm) containing 3 g of untreated diet and kept at 26±1°C and 75±5% R.H. Larvae of control were fed on untreated diet sprayed only with water instead of tested compound.

Larval mortality, abnormality and different biological effects on the larvae, pupae and adults (moths) were recorded. After moth emergence, three replicates each contained cage of (5-pairs/cage) of emerged moths that appeared morphologically not impaired were used to measure the reproductive potential of the insects in assay. Laid eggs were incubated under controlled conditions then counted after hatching to estimate the egg hatchability percentages.

Reduction in hatchability percentage was calculated according to Zidan and Abdel-Megeed (1987). Fecundity percentage was calculated according to Crystal and Lachance (1963) as follows:

$$\% \text{ Reduction hatchability} = \frac{\text{no. hatched eggs in check} - \text{no. hatched eggs in treatment}}{\text{no. of hatched eggs in check}} \times 100$$

$$\% \text{ Fecundity} = \frac{\text{no. of eggs / treated female}}{\text{no. of eggs / untreated female}} \times 100$$

The recorded data were statistically analyzed with one – way analysis of variance (ANOVA) (P < 0.05 %) (Snedecor, 1952) and Duncan's multiple range test of means.

Treating pupal stage of *Coccinella undecimpunctata* predator:

The larvae and pupae of *C. undecimpunctata* predator which were collected from the field were reared singly in tubes (7x 2.5cm²). Larvae were fed and daily observed until pupation for used in treatment.

Three replicates each contains 20 pupae (1-2 day old) of *C. undecimpunctata* were sprayed by different concentrations (300, 150, 75, 37.5, 18.75 and 9.375 ppm) of the tested compound in Petri-dish, after 1hr, pupae were translated singly in tubes (7 x 2.5 cm²). After that, pupal mortalities (from 10-14 days) and the time of adult emergency were recorded. Percentages of mortalities were corrected according to Abbott's formula (1925).

Feeding the adults of *C. undecimpunctata*:

Treatment of *P. gossypiella* eggs was done by dipping a piece of paper containing eggs treated with (LC50). Three replicates from eggs were used, (from 200 to 300 eggs on paper for each replicate). After that the papers were left until dried then introduced to *C. undecimpunctata* adults for feeding. Other of similar eggs were dipped in water used as a control. The treated and untreated eggs were offered to adults' predator daily until died.

Preparation of samples for biochemical assay:

Samples of *Pectinophora gossypiella* larvae were collected at 12-14 days after treatment of the newly hatched larvae (during the molting), while, *C. undecimpunctata* pupae and adults were collected at 7 -10 days after treatment with LC₅₀. Untreated larvae of *P. gossypiella* and *C. undecimpunctata* (pupae and adults) stages were used as control.

Biochemical analyses:

The total lipids were estimated by the method of Knight *et al.*, (1972), colorimetric determination of total soluble protein in total homogenate *P. gossypiella* larvae was carried out, as described by Bradford (1976). Transaminase enzymes. Aspartate amino transferase (AST) and alanine aminotransferase (ALT) enzyme activities were determined calorimetrically according to the method of Reitman and Frankle (1957). Determination of chitinase activity was prepared according to Bade and Stinson (1981) and the reaction mixture of enzyme assay according to Ishaaya and Swirski (1976). Determination of N-acetylglucosamine by the sensitive method of Waterhouse *et al.*, (1961). Phenoloxidase activity was determined according to modification of Ishaaya (1971) and Total

proteins were determined by the method of Bradford (1976).

RESULTS

Toxicity of Teflubenzuron on newly hatched larvae of *P. gossypiella*:

The LC₂₅, LC₅₀ and LC₉₀ values of the tested IGR, Teflubenzuron on the newly hatched larvae of *P. gossypiella* were 19.36, 78.59 and 1189.25 ppm, respectively.

Effects of LC₅₀ values of Teflubenzuron on the mortality, development period and fecundity of *P. gossypiella* Larval and pupal stages:

Data in Table (1) reported that the mortality in newly hatched larvae of PBW was gradually increased after 1 to 3 days reaching 59.6 % mortality, compared to 2% in control and raised to 71.9 % mortality after 1 to 18 days of application compared to 7% in control,.

Developmental period of larvae and pupal stages:

Significant differences occurred in the developmental periods of larval & pupal stages and total life span (from larva to adult stage) and adult fecundity (reproductive) of *P. gossypiella* in treated compared with untreated (Table 1).

Table (1): Effect of LC₅₀ value of Teflubenzuron on some biological parameters of *P. gossypiella* larval and pupal stages.

| Treatments | Mortality larvae % | | Time in days | | |
|------------------|--------------------------|--------------------------------------|---------------------|--------------------|-------------------|
| | 1-3-7 days from treating | Accumulative Mortality until 18 days | Larval (L) Duration | Pupa (P.) duration | Total L & P Stags |
| LC ₅₀ | 59.6±3.76 | 71.9±3.51b | 20.3±1.51b | 10.63±0.31b | 30.93±2.5 |
| Untreated | 2.0±0.58 | 7.67±0.88a | 14.60±0.35a | 7.36±0.3a | 21.96±1.3 |
| LSD 5% | | 0.254 | 1.665 | 0.306 | 2.331 |

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Data in Table (1) show that Teflubenzuron doses significantly prolonged the developmental period of survived larvae and pupae compared with the control. The treated larval duration averaged to 20.3 days/ larvae and 10.63 days/ pupae compared with 14.6 days/ larvae and 7.36 days/ pupae, respectively, in the untreated control. This period prolonged (approximately from 1.4 time / larvae and 1.6 times/ pupae) when newly hatched larvae treated with the growth inhibitor than control.

In the same table data illustrated significant increased in total immature stages of *P. gossypiella* resulted from the newly hatched larvae treated to 30.93 days compared with 21.96 days in control.

The obtained results supported by Kandel *et al.*, (2013) who recorded that the CSI, compound increased developmental period of *P. gossypiella* larvae and pupae, which led to longest total life span (from larval to adult stage), it estimated by 54.23 days/ life span in treated compared to 42.46 days/ life span in control, in addition treated larvae showed the presence of a double cuticle and could not shed the old cuticle.

Oviposition and reproductive potential:

Statistical analysis of the data in Table (2) demonstrated that highly significant differences between oviposition and reproductive adults resulted from treated

larvae and the control. The mean number of deposited eggs (fecundity) by females emerged from larvae treated with LC₅₀ value of Teflubenzuron were 160.0 eggs/female, compared to 226.67 eggs/female in untreated control.

These data indicated that the treated larvae by Teflubenzuron caused significantly high reduction in laid eggs by females to 29.4%. At the same trend, the treatment by Teflubenzuron caused high reduction in percentage of hatchability (fertility) reaching 42.7% compared with 5.4 in control. Similar trend was reported in the larval mortality with the first molting larvae.

Malformation of different stages of pink bollworm and the lady bird:

As shown in Figure (1, 2) the tested IGR, Teflubenzuron caused malformation in all stages of both pink bollworm, *P. gossypiella* (larvae, pupae and moths) and its predator, *C. undecimpunctata*, (pupae) which failed to complete their life cycles.

Biochemical studies:

The most important enzymes which play a key role in molting process in body of *P. gossypiella* were analyzed for their activities i.e. total protein, phenoloxidase, chitinase and N-acetyl glucoseamine, Free-amino acid, Transaminase enzymes, Aspartate aminotransferase (AST) and alanine aminotransferase (ALT).

Table (2): Effect of LC₅₀ value of Teflubenzuron on some biological parameters of *P. gossypiella* adult stage.

| Treatment | Adult emergency % | Total eggs | Hatchability % | Reduction % | Time in days | |
|------------------|-------------------|---------------|----------------|-------------|---------------------|---------------------------------|
| | | | | | Longevity of female | life span from larvae to adults |
| LC ₅₀ | 75.33 ±5.12 | 160.0 ±2.1 | 57.3 | 42.7 | 23.3 | 54.23±2.1 |
| Untreated | 95.33 ±0.91b | 226.67 ±4.91b | 94.6 | 5.4 | 18.55 | 42.46±2.1 |
| LSD 5% | 4.225 | 8.215 | 7.335 | 1.422 | 0.025 | 3.208 |



Larva



Larva



Pupa



Pupa



Moth



Moth

Figure (1) Malformation in cotton bollworm stages



Pupa



Pupa

Figure (2) Malformation in lady bird pupae

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Results in Table (3) show the effect of Teflubenzuron on some biochemical changes of PBW as the total contents of protein and phenoloxidase when the 1st instars larvae fed on diet treated with LC₅₀ (78.59 ppm) of Teflubenzuron.

The obtained results supported by Nehad M. El-Barkey *et al.*, (2009) who found that the treatment of *P. gossypiella* larvae and pupae with (IGRs) reduced the total protein content to 10.5 mg./gb.wt (approximately 50.1%) compared with 21.935 mg./gb.wt in control. Thompson *et al.*, (2005) reported that the reduction in total protein soluble has been found in analyzed larvae to be a key factor for elongated in development immature stages and reduction in number of eggs laid (adults fecundity). Furthermore, Simpson *et al.*, (2004), Raubenheimer and Simpson (2005) found that the ratio of proteins to soluble carbohydrates (P: C) has been necessary for development, or survivorship of some other insect species.

While the phenoloxidase which necessary in melanin production during cuticle content, the obtained results in Table (3) revealed that it decreased to 4.74 (O.D.

units/gb.wt) equal to 83.34% in the treatment, this decrease indicate that the treatment caused reduction in phenoloxidase approximately by 8 times than control.

The obtained results supported by Assar *et al.*, (2016) who found that after treating *S. littoralis* with the IGR, the activity of phenoloxidase was highly significantly decreased.

In addition, data in Table (3) reported that the chitins activity of the larvae treated with Teflubenzuron was 1101.67 compared to 1050 in control, while, the units (μg N-acetyl glucoseamine liberated $\times 10^3/\text{min/g}$. b.wt.) increased approximately to (2 times), it determined by 289.67 in larvae treated compared to 165.33 in larvae untreated. It could be concluded that Teflubenzuron caused incomplete larval molting and failed pupation.

The obtained results are confirmed with that obtained by Kandel *et al.*, (2013) who explained the effect of (IGRs) on chitin activity in the body of *P. gossypiella* larvae fed on diet treated with 78.59 ppm.

Table (3): Some biochemical changes in *P. gossypiella* larvae (during molting process) treated with Teflubenzuron (IGR) at 987.59 ppm

| Biochemical aspects | Treated | Control | decreased(-)or increased(+) |
|---|---------------|--------------|-----------------------------|
| Total protein(mg/g.b.wt) | 10.51±0.31 | 21.93± 0.7 | -52.1 |
| Free-amino acid(μg D,L-alanine/g.b.wt) | 831.17±20.4 | 622.3± 11.9 | +32.44 |
| Phenoloxidase(O.D. units/g.b.wt) | 4.74±0.23 | 28.47± 1.6 | 83.4 |
| N- acetyl-glucceamine(μg NAGA /g.b.wt) | 289.67±4.69 | 165.33±5.7 | +75.2 |
| Chitinase (μg NAGA $\times 10^3/\text{min/g}$.b.wt) | 1101.67±39.39 | 1050±16.12 | +4.92 |
| Aspartate aminotransferase (AST) (GOT) (U $\times 10^3/\text{g}$.b.wt) | 6670±380.8 | 663.66±217.6 | +905.03 |
| alanine aminotransferase (ALT)Gpt (U $\times 10^3/\text{g}$.b.wt) | 1533.3±34.3 | 1528.67±37.7 | +0.7 |

Transaminase enzymes (GOT and GPT or ALT and AST):

Data in Table (3) show the transaminase enzymes activity on larvae of *P. gossypiella* reared on diet treated by LC₅₀ (IGRs). The levels of GOT were highly increased to 6670 compared to 663.66 mg/ml in control, while, the transaminase enzymes GPT activity on treated PBW larvae was (1533.3 mg/ml) compared with (1528.67mg/ml) in untreated.

Toxicity of Teflubenzuron on pupae and adults of *C. undecimpunctata*:

The obtained data in Table (4) revealed that the LC₂₅ and LC₅₀ values were 37.2 and 109.9 ppm, respectively when the 1-2day old pupal stage treated by Teflubenzuron, and were 29.6 and 1189.25 ppm for adult stages of lady bird.

Adult mortality and malformed of *C. undecimpunctata*:

Data in Table (4) show the percentages of (pupal stages) mortality of *C. undecimpunctata* after using different levels of concentrations (300, 150, 75, 37.5, 18.75 and 9.375 ppm of Teflubenzuron) at 24h, 7 and 10 days. The differences among compound levels on pupae mortality rates were statistically significant. The percentages mortality of the predator were higher when the levels of Teflubenzuron

(TFB) increased (*Coccinella* adult stage; F= 2115 DF= 5, P < 0.05). A total of 66% pupa stages were died at recommended rate.

Results in table (4) showed the percentage of adults' emergence, the results indicated that percentage of adults emergence decreased by the increasing of used concentration. The vales were (6.6, 20, 51.66, 71.66, 38.33, 90 and 93.33 respectively), when 300, 150, 75, 37.5, 18.75 and 9.375 were used, respectively.

Malformation of lady bird pupae:

As shown in table, 4 and Figure (2) the tested IGR, Teflubenzuron caused malformation in predator, *C. undecimpunctata*, (pupae) which failed to complete their life cycles. The malformed percentages of the predator were decreased when the levels of (TFB) decreased from (32 to 1%.

Duration of pupal stages:

Statistical analysis of results in Table (4) revealed that there were highly significant differences between the time periods in day for pupae treatment and untreated. The duration in treatment averaged 14.3 days/pupa compared with 6.5 days, in the untreated pupae.

Table (4): Effect of different concentration of the tested IGR on toxicity and duration of *C. undecimpunctata* pupae

| Conc. (ppm) | number of pupae | Treated <i>C. undecimpunctata</i> pupae | | | %adults Emergence |
|-------------|-----------------|---|------------------------------|-----------------|-------------------|
| | | Mortality % (after 24h to 10 days) | Malformed % (from7-12 days) | Duration (days) | |
| 300 | 60 | 61.4 | 32 | - | 6.6 |
| 150 | 60 | 56 | 24 | 16-18 | 20 |
| 75 | 60 | 37.34 | 11 | 14-17 | 51.66 |
| 37.5 | 60 | 20.34 | 8 | 13-15 | 71.66 |
| 18.75 | 60 | 10.67 | 6 | 9-10 | 83.33 |
| 9.375 | 60 | 9 | 1 | 7-9 | 90 |
| Control | 30 | 6.67 | - | 6-7 | 93.33 |

Effect of IGR on Fecundity and longevity:

Results in Table (5) indicated that The highest number of eggs (189 eggs/ female) were deposited by *Coccinella* fed on PBW eggs untreated, while the predator females fed on eggs treated with IGR laid the lowest number of individual eggs (76.6 eggs/ female). The cumulative number of eggs laid per female of lady bird decreased to about 59.4% compared to control when adult females were treated and feed on eggs treated with recommended dose of the compound. The highest longevity was recorded by adult female fed with the eggs untreated. Teflubenzuron significantly decreased the oviposition period and longevity days for *C. undecimpunctata* females.

The oviposition period of females fed continuously on PBW eggs treated with the recommended LC₅₀ took 24.6 days compared with 37.9 days/ female in control; during this period the number of eggs were sharply decreased to the half compared to control.

Data in Table (6) show the changes in some biochemical analyzes of *C. undecimpunctata* in total protein and

phenoloxidase when the pupal stage treated with LC₅₀ of Teflubenzuron.

Treatment with LC₅₀ IGR reduced the total protein content to 6.29 in pupae compared with 8.77 in control and 7.9 in treated adults compared to 13.28 in control, while the phenoloxidase which necessary in melanin production during cuticle composition increased approximately 2 times, it estimated by 9.9° (O.D. units/g b. wt) in treated pupae compared with 5.96 in control.

DISCUSSION

Biological aspect:

Nehad M. El-Barkey *et al.*, (2009) found that Hexaflumuron (IGR) caused a prolongation in larval and pupal developments of *S. littoralis*, where durations were 22.3, 20.6 and 20.4 days, respectively for larvae after egg treated and 10.8, 10.0 and 11.3 days, respectively for pupae. Also, they found high reduction in total laid eggs, percentage of hatchability and longevity in adult stage and low concentration of IGR reduced the fecundity and egg hatching and increased the sterility of adults resulted from treated larval and pupal stages.

Table (5):Effect of Teflubenzuron on soma biological aspects for *C. undecimpunctata*

| Conc. (LC ₅₀) Ppm | Initial number of pupa | Treated <i>C. undecimpunctata</i> pupae | | | % adults emergence |
|----------------------------------|------------------------|---|-------------------------|----------------|--------------------|
| | | Mortality% | Malformed% | Duration | |
| 106.9ppm | 60 | 48 | 32.3 | 14.3±0.6 | 67.7 |
| Control | 60 | 3 | 1 | 6.5±0.2 | 96 |
| LSD 5% | - | 0.255 | 0.745 | 1.684 | 3.558 |
| Adult stage | | | | | |
| | Oviposition | Total egg/ female | Reduction in laid eggs% | % hatchability | Longevity |
| 106.9ppm | 24.6±0.3 | 76.6±0.3 | 64.2 | 66 | 37.3 |
| Control | 37.9±0.3 | 189.±0.3 | - | 89 | 46.5 |
| LSD 5% | 2.635 | 5.693 | - | 2.112 | 2.441 |

Table (6): Biochemical aspects of 987.59 ppm Teflubenzuron dose on two stages of *C. undecimpunctata*

| Biochemical aspects | Pupal stage | | Adults stage | |
|---|--------------|------------|--------------|---------------|
| | Treated | Control | Treated | Control |
| Total protein(mg/g.b.wt) | 6.29±0.2 | 8.77±0.3 | 7.6±0.6 | 13.28±0.63 |
| Free-amino acid(µg D,L-alanine/g.b.wt) | 721±15.61 | 990±20.81 | 955±25.31 | 1013.28±33.63 |
| Phenoloxidase(O.D. units/g.b.wt) | 9.95±0.26 | 5.96±0.2 | 3.9±0.3 | 4.33±0.4 |
| N- acetyl-glucosamin(µg NAGA /g.b.wt) | 344.67±8.44 | 237.0±20.9 | 221±11.6 | 226.0±20.9 |
| Chitinase (µg NAGA x10 ³ /min/g.b.wt) | 1953.67±20.4 | 2141±6.4 | - | - |
| Aspartate aminotransferase (AST)(GOT) (U x 10 ³ /g.b.wt) | 246.0±4.6 | 267.0±2.7 | 215±20.8 | 364.6±12.6 |
| alanine aminotransferase (ALT)GPT (U x 10 ³ /g.b.wt) | 1616±3.4 | 168.66±5.6 | 698±31.6 | 1237.36±13.5 |

The obtained data are in agreement with many authors tested different IGRs against Lepidopterous insects, e.g., *P. gossypiella*, Flint *et al.* (1978), Moawad and Khidr, (1982). Also, *Spodoptera littoralis*, Ismail, (1980); El-Deeb *et al.*, (1991); Sokar, (1995); Shaurub *et al.*, (1999) and Abdel-Aal, (2003).

Hewady *et al.*, (2002) found that Neemazal disrupt larval and pupal development of pink bollworm, resulted malformations, reduction in longevity fecundity and fertility moth. Sammour *et al.*, (2008) studied the effect of Chlorfluazuron and Leufenuron on *S. littoralis*, and indicated that this compound decrease adult emergence, longevity, fecundity and egg hatchability. Moursy and Salem (1995), Macro and Vinuela (1994) and Lyra *et al.*, (1999) reported that IGRs caused morphological alternations of ovipositor and inhibition of the ovarian growth. Kellouche and Soltani (2006) found that Hexaflumuron decrease the longevity and fecundity of *Callosobruchus maculatus*. Also, Al-Shannaf

and Kandil (2005) determined the LC₅₀ of spinosad for one and two days old eggs of *Helicoverpa armigera* as 2.56 and 1.31 ppm, respectively.

Physiological aspects:

The obtained results are in agreement with Assar *et al.*, (2016) who studied the effect of Hexaflumuron and Teflubenzuron (IGR's) against the 4th instar larvae of *Spodoptera littoralis* on total carbohydrates, proteins, lipids, acetylcholinesterase, chitinase, phenoloxidase, carbohydrates hydrolyzing enzymes, non-specific esterases, phosphatases and transaminase enzymes, and reported that total proteins and lipids content were significantly decreased also, Hexaflumuron and Teflubenzuron caused an increase in total carbohydrates, moreover, the tested IGRs significantly increased the invertase activity but decreased the activity of trehalase and amylase in case of Teflubenzuron. The tested IGRs significantly decreased the activity of alkaline (AIP) phosphates. Teflubenzuron induced a significant increase

on aspartate aminotransferase (AST) and alanine aminotransferase (ALT). Furthermore, Acetylcholinesterase (AChE) activity was significantly increased with Teflubenzuron while decreased with Hexaflumuron, as well as Phenoloxidase and Chitinase activity significantly increased.

Similar results were found by Mostafa, (1993) and Sokar, (1995) on *S. littoralis*. The protein pool of the haemolymph consider a source of protein synthesis for growth and development of the adult stage (Florkin and Jeanuiaux, 1964).

El-Sheikh *et al.*, (2013) and Florkin and Jeanuiaux, (1964) reported that Teflubenzuron caused changes in trehalase on *S. littoralis*, the rapid decrease of glucose concentration of last larval instar of the cotton leafworm, was probably caused by high metabolic activity.

These results were also agree with the results which conducted by Abd El-Mageed and Shalaby, (2011) who reported that Teflubenzuron appeared reduction in acetylcholinesterase. El-Sheikh *et al.*, (2013) stated that increase in chitinase activity and fluctuated changes were recorded when *S. littoralis* was exposed to Teflubenzuron.

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التأثيرات الكيميائية و البيولوجية لمنظم النمو الحشري تيفلوبينزيرون على دودة اللوز
القرنفلية (LEPIDOPTERA: GELECHIIDAE) وابو العيد احدى عشر نقطة
(COLEOPTERA: COCCINELLIDAE)

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الملخص العربى

أجريت هذه الدراسة لمعرفة تأثير منظم النمو الحشري تيفلوبينزيرون على كلا من نسبة حياة وتكاثر حشرة دودة اللوز القرنفليه وعلى العذارى والاطوار الكاملة للمفترس ابو العيد ١١ نقطه. بالنسبه لدودة اللوز القرنفليه كانت قيم الجرعات المميته لربع المجموع والجرعات المميته لنصف المجموع هي ١٩,٣٦ ، ٧٨,٥٩ جزء فى المليون على التوالي عند معاملة اليرقات حديثه الفقس بال تيفلوبينزيرون فى حين كانت قيمة الجرعه المميته لـ ٩٠٪ من المجموع هي ١١٨٩,٢٥ جزء فى المليون ، وقد أدت المعامله بال تيفلوبينزيرون الى زياده معنويه فى فترة نمو وتطور اليرقات والعذارى مقارنة بمعاملة الكنترول. وأوضح التحليل الإحصائى للنتائج الى وجود إختلافات معنويه بين فترة وضع البيض وتكاثر الحشره الكامله الناتجه من معاملة اليرقات مقارنة باليرقات الغير معامله. وأظهرت النتائج ان المعامله بالجرعه المميته لنصف المجموع بال تيفلوبينزيرون عند ٧٨,٥٩ جزء فى المليون أدى الى إنخفاض محتوى البروتين الكلى للحشرات المعامله فى حين ان محتوى الـ phenoloxidase انخفض بشكل معنوى. وأظهرت النتائج ان نشاط إنزيم الـ Chitinase زاد بشكل معنوى مسببا فشل فى عملية التعذر، وزاد نشاط إنزيم الـ transaminase بشكل معنوى.

بالنسبه لحشرة أبو العيد ١١ نقطه كانت نسب الموت مختلفه لكلا من العذارى والحشرات الكامله بعد يوم الى سبعة أيام من التعرض لـ ٣٠٠ ، ١٥٠ ، ٧٥ ، ٣٧,٥ ، ١٨,٧٥ ، ٩,٣٧٥ جزء فى المليون من الـ تيفلوبينزيرون حيث زادت نسب الموت بزيادة تركيز الـ تيفلوبينزيرون، علاوه على ذلك زادت مدة التعذر للمفترس بشكل معنوى مع زيادة جرعة منظم النمو فى حين انخفضت طول فترة العمر والخصويه للحشرات الكامله بشكل معنوى. انخفضت أيضا بشكل معنوى قيم المحتوى الكلى للبروتين والـ phenoloxidase عندما عوملت العذارى وتغذت الحشرات الكامله بالجرعه القاتله لنصف المجموع من منظم النمو تيفلوبينزيرون.

Biochemical and biological effects of insect growth regulator, teflubenzuron ...