

Toxicity Evaluation and Field Performance of Certain Insect Growth Regulators for *Spodoptera littoralis* Management in Cotton

Mervat H.A. Metayi¹, and Manal A. M. Attia^{2*}

ABSTRACT

Spodoptera littoralis (Boisd) (Lepidoptera: Noctuidae), management requires the implementation of effective and sustainable control strategies. In this study, we aimed to compare the insecticidal activity and field performance of four commonly used insect growth regulators (IGRs) against *S. littoralis* larvae for helping in decision making to get the most effective and sustainable pest management for *S. littoralis* under local condition. The tested IGRs included Benzoylphenyl ureas BPU (Lufenuron, Chlorfluazuron, Diflubenzuron), and an ecdysone receptor agonist (Methoxyfenozide). Laboratory bioassays were conducted to determine the toxicity of these IGRs against 2nd and 4th instar of *S. littoralis* larvae. Lufenuron exhibited the highest toxicity, with the lowest LC₅₀ values (0106 & 0.069 and 0.889 & 0.453 mg/l) at 72 and 96h of exposure, respectively. Chlorfluazuron, Diflubenzuron, and Methoxyfenozide displayed relatively lower toxicity compared to lufenuron, as their toxicity indexes were (2.59, 1.99 and 4.50) refereeing to the lowest LC₅₀ value of Lufenuron against the 2nd instar after 72h of exposure. Moreover, positive significant correlations were observed between the log LC₅₀ values of each IGR and other tested ones (pairwise comparison) which alert the possibility of cross resistance between these IGRs. Field experiments were performed over two cotton seasons to assess the efficacy of the IGRs in reducing *S. littoralis* larval populations. All treatments resulted in a significant reduction in larvae compared to the control group. Lufenuron consistently demonstrated the highest initial efficacy (73.5 and 75.2%) and mean reduction (83.9 and 81.5%) after 15 days of treatment in both 2020 and 2021 seasons, respectively. In contrary, the lowest efficacy was recorded for Diflubenzuton with mean reduction (68.2 and 71.2) after 15 days of treatment in 2020 and 2021 cotton seasons, respectively. This finding highlights the importance of selecting IGRs with higher toxicity to achieve effective larval control. The work underscores the potential of lufenuron as a highly effective IGR for managing *S. littoralis*. Furthermore, our research emphasis the crucial of the continuous evaluation of IGRs efficacy

and persistence as well as resistance and cross resistance to IGRs under local field conditions.

Keywords: Lufenuron; Methoxyfenozide; Benzoylphenyl ureas; Ecdysone receptor agonist; Cotton leaf worm control; IGRs toxicity

INTRODUCTION

Spodoptera littoralis (Boisd) (Lepidoptera: Noctuidae) is a damaging polyphagous agricultural pest that present and widespread in Africa, Europe and Asia (CABI, 2022). *S. littoralis* attacks several field and vegetable crops (Salama *et al.*, 1971) including cotton. In Egypt, cotton is one of national income component. The Egyptian production was 59000-ton of lint and 100750-tone of seeds in 2020 (FAOSTAT, 2023). Moreover; the high demand for cotton and its price elevation has led to increase the cultivated area by 30% in 2021, according to a press report from the Ministry of agriculture released on Thu, 7 Oct, 2021, World Cotton Day. *S. littoralis* causes extensive direct injury on cotton and vegetables in addition to the environmental impact resulting from its control. Chemical control is one of the critically important tools for managing *S. littoralis* which cannot be given up. However, the misuse of conventional insecticides since 1950's has led to the development of resistance (El-Guindy *et al.*, 1982 and Moustafa *et al.*, 2023) and environmental problems (Damalas & Eleftherohorinos, 2011 and Singh *et al.*, 2018). These conditions along with the environmental concern force researchers to find alternatives or, at very least, specific chemical to replace the traditional insecticides. Insect growth regulators (IGRs), the third generation of insecticides, is a promising alternative to traditional insecticides due to their selectivity (Kodandaram *et al.*, 2010) and low toxicity to non-target organisms (Sánchez-Bayo, 2012). According to the official recommendation of the Ministry of Agriculture and Land Reclamation (2022), the application of conventional insecticides, while the egg-masses of *S.littoralis* are dominated, is not desirable,

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¹ Plant Protection Research Institute, Sabahia, Alexandria, Egypt;

E-mail: aboelhamedmervat@gmail.com

²Bioassay Research Department, Central Agricultural Pesticide Laboratory,

Agricultural Research Center, Dokki, Giza,Egypt

E-mail: manal.attia77@gmail.com

*Corresponding author:

Manal A. Attia

Bioassay Research Department, Central Agricultural Pesticide Laboratory,

Agricultural Research Center, Dokki, Giza,Egypt

E-mail: manal.attia77@gmail.com/ Orcid no. <https://orcid.org/0000-0002-9884-947X>

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while IGRs are recommended to control the recently hatched larvae. IGRs disrupt the molting process as they interfere with the natural function of the endocrine system in insects, in the end affecting their development, reproduction or metamorphosis (Kai *et al.*, 2009). The main categories of IGRs are Juvenile hormone (JH) mimics and chitin synthesis inhibitors (CSIs) (Tunaz and Uygun, 2004). Several studies have investigated the efficacy of IGRs against *S. littoralis* (El-Sheikh & Aamir, 2011; Elsayed *et al.*, 2017 and Abdel Aziz & El-Gabaly, 2021) evaluated the efficacy of lufenuron against *S. littoralis*. Similarly, Mageed *et al.* (2018) investigated the effectiveness of chlorfluazuron against *S. littoralis* and reported that the mortality and sublethal effects of chlorfluazuron in insects, can provide a truly selective insecticide. Benzoylphenyl ureas BPU, have been used as pesticides since the 1970s (Retnakaran & Wright, 1987 and Spomer & Sheets, 2019) this group includes Lufenuron, chlorfluazuron and diflubenzuron which belong to CSIs, according to Insecticide Resistance Action Committee (IRAC, <https://www.irac-online.org/modes-of-action>) they are inhibitors of chitin biosynthesis affecting enzyme chitin synthase 1; thus inhibiting chitin polymerization (Douris *et al.*, 2016) while, methoxyfenozide has a different mode of action, since it fits to Ecdysone receptor agonists. Despite the widespread use of lufenuron, chlorfluazuron, diflubenzuron and methoxyfenozide to control *S.littoralis* in cotton fields there is a shortage of research comparing their efficacy and persistence under local Egyptian conditions in particularly given the obvious climate changes. In Egypt, these four IGRs insecticides are recommended and utilized for controlling *S. littoralis* in cotton fields. However, the efficacy of these insecticides may fluctuate making selection of the most suitable insecticide impractical and costly. The comparative of both laboratory toxicity and field efficacy of such insecticides are in demand.

Therefore, the present study aims to compare the efficacy of lufenuron, chlorfluazuron, diflubenzuron, and methoxyfenozide against *S. littoralis* larvae with respect to their toxicity and reduction rates for helping in decision making to get the most effective and sustainable pest management under local condition.

MATERIALS AND METHODS

Insecticides:

Lufenuron (Match[®] 5%EC) and Chlorfluazuron (Atabron[®]5%EC) were provided by Syngenta. Diflubenzuron (Deflux[®] 48%SC) was provided by Kanza group and Methoxyfenozide (Runner[®] 24%SC) was provided by Dow Agrosiences.

Laboratory Experiments:

Insects: A susceptible strain of *S. littoralis*, have been raising for many generations according to the methodology established by Eldefrawi *et al.* (1964), in a laboratory at the Plant Protection Research Station, Alexandria, Egypt under a controlled environment (27 ± 2 °C, rh of $65\% \pm 5$ and 12: 12 L:D). The *S. littoralis* larvae were fed castor oil leaves while the adults fed 10 % sucrose solution.

Bioassay procedure: A standard leaf dip bioassay technique (Eldefrawi *et al.*, 1964) was conducted, where castor oil leaves were dipped for 30 seconds in water dilutions and allowed to dry for an hour at room temperature. A series of 6-8 dilutions of each insecticide separately were testes along with the control group where larvae were fed leaves treated with tap water. Treated leaves were introduced to ten larvae of second or fourth instar larvae of *S. littoralis* in cups covered with muslin lids and placed in an incubator with the above controlled conditions. Each concentration was replicated three times and mortality was observed after 72 and 96 h after treatment.

Field experiments:

The field experiments were carried out for two consecutive cotton seasons (2020-2021) in Abu El Matamir, El-Beheira Governorate. The arrangement of the experimental plots followed completely randomized design (CRD) and each treatment was represented by three randomly assigned plots. Each of these plots was sized at 84 square meters and was separated from each other by leaving unplanted inter-row spaces. The utilized farming techniques were in accordance with the standards of "good agricultural practice". Cotton cultivar Giza 86 was sowing May 3rd, 2021 and May 6th, 2022. Recommended rates of lufenuron;125 ml/feddan, chlorfluazuron; 200 ml/feddan, diflubenzuron; 125 ml/feddan and methoxyfenozide;125 ml/feddan were applied separately by spraying 200 liters/ feddan using knapsack sprayer apparatus. Control plots were treated exclusively with water. The treatments were conducted during the early morning on July 27, 2021 and July 30, 2022. The data collection was also performed during the same time of the day, before treatment and on the third, fifth, seventh, tenth, and fifteenth-days post-treatment. The cotton leafworm larval instars were counted on ten labeled plants per plot relying on the insecticide evaluation protocol outlined by the Egyptian Ministry of Agriculture (Mohmed *et al.*, 2019 and Amein *et al.*, 2023). The reduction percentages were calculated according to Henderson and Tilton (1955) referring to the untreated (control), and subsequently the data were subjected to statistical analysis.

Statistical analysis

The mean lethal concentrations LC_{50} values their confidence limits and slopes were estimated using Ldp Line ® software according to Finney (1971). The reduction percentages of larvae were subjected to one-way ANOVA analysis (SAS, 1985 and Steel & Torrie, 1980), followed by Student-Newman Keul post hoc test. The bivariate relationships between Log LC_{50} values of each IGRs and the other tested ones were analyzed using Pearson's correlation analysis using IPM SPSS statistic 20.

RESULTS

The bioassay data revealed that, all four tested IGRs exhibited significantly different insecticidal activity levels against the 2nd and 4th instar larvae of *S. littoralis*. The LC_{50} values of Lufenuron, Chlorflauzuron, Diflubenzuron, and Methoxyfenozide against the 2nd instar larvae (Table1) after 72 h. of exposure were 0.106, 0.275, 0.477, and 0.211 mg/l, respectively. These values significantly decreased to 0.069, 0.199, 0.195, 0.179 mg/l, respectively at 96 h post treatment. Lufenuron was found to be the most toxic among the tested IGRs, with the lowest LC_{50} values (0.106 and 0.069 mg/l) against the 2nd instar larvae after 72 and 96 h of exposure, respectively. This pattern of toxicity was also observed against the 4th instar larvae of *S.littoralis* (Table 2), where Lufenuron was the most toxic insecticide with the lowest LC_{50} values (0.889 and 0.453 mg/l) after exposure periods of 72 and 96 h, respectively. Chlorflauzuron, Methoxyfenozide and Diflubenzuron followed Lufenuron in terms of toxicity with LC_{50} values of (1.352 and 0.944 mg/l), (1.642 and 0.953 mg/l) and (3.18 and 1.84 mg/l) after 72 and 96 h of treatment. The toxicity index of Chlorflauzuron, Diflubenzuron and Methoxyfenozide against the 2nd instar were (2.59 & 2.88), (4.50 & 2.826) and (1.99 & 2.59) after 72 and 96 h of treatment, respectively. Similarly, for the 4th larval instar of *S.littoralis* the toxicity index of Chlorflauzuron, Diflubenzuron and Methoxyfenozide, were (1.52, 1.85 and 3.58) and (2.08, 4.06 and 2.10) after exposure periods of 72 and 96 h, respectively. Furthermore, as it is obvious the 2nd instar larvae were more susceptible to all tested insecticide which was confirmed by pairwise correlation analysis between the log LC_{50} values against 2nd and 4th instar of Lufenuron, Chlorflauzuron, Diflubenzuron and Methoxyfenozide in one side and instar in the other side since there was a strong positive correlation between instar and toxicity (correlation is significant at the 0.01 level, 2-tailed, with coefficient of 0.942, 0.936, 0.899 and 0.966), respectively. In addition, the correlation between log LC_{50} values of the tested IGRs revealed a strong relation

between each insecticide and the other tested ones (Table 3).

Field experiments results presented in (Tables 4 and 5) showed that, all insecticides treatments significantly reduced the number of cotton leafworm larvae at all time intervals. Considering results during fifteen days after treatments in 2020 season, it is obvious that the maximum larvae reduction observed after seven days following treatments. Lufenuron showed the highest efficacy, with a mean reduction of 88.40 % at day 7 after treatment, followed by Methoxyfenozide (80.40%), Chlorflauzuron, (77.00%) and Diflubenzuron (74.30%). Yet, there was no statistically significant difference between Chlorflauzuron and Methoxyfenozide. The larval reduction showed a gradual decrease over the following days, however, the statistical analysis revealed that this decline was non-significant. Lufenuron still exhibited the highest efficacy with a mean reduction of 83.90%, while Diflubenzuron showed the lowest efficacy with a mean reduction of (68.20%). The results obtained from the 2021 season (Table 4) were consistent with those of the 2020 season, where all tested IGRs displayed significant reduction in the number of cotton leafworm larvae across all time intervals. Lufenuron continued to exhibit the highest level of efficacy, demonstrating an average reduction of (87.20%) at the 7th day post-treatment, followed by Methoxyfenozide (75.70%), Chlorflauzuron (75.00%), and Diflubenzuron (74.80%). However, there were no significant differences between Methoxyfenozide, Chlorflauzuron and Diflubenzuron at the 7th day after treatment. Although the larval reduction exhibited a gradual decline over subsequent days, also the statistical analysis indicated that this decline was non-significant. Likewise, at 15 days post-treatment, Lufenuron still maintained the highest efficacy level, resulting in a mean reduction of 81.50%. Conversely, Diflubenzuron displayed the lowest level of efficacy, with a mean reduction of (71.20%).

Considering the average reduction of *S. littoralis* larvae over the experiment a period of fifteen days post-IGRs spraying in cotton fields, the results (Fig.1) demonstrate that all IGRs tested exhibited differences in reduction in the number of *S.littoralis* larvae. Lufenuron displayed the highest efficacy in both 2020 and 2021 seasons, resulting in average reduction of 83.40 % and 82.72%, respectively within the 15 days post-treatment. Conversely, Diflubenzuron showed the lowest level of efficacy in both 2020 and 2021 seasons, with average larvae reduction of 69.76% and 72.43% during fifteen days post-treatment, respectively. However, in 2021 season there was no significant difference in larvae reduction between Methoxyfenozide and Chlorflauzuron, while, only Lufenuron showed

significant larvae reduction compared to other IGRs (Fig 1).

Table 1. Toxicity of lufenuron, chlorflauzuron, diflubenzuron and Methoxyfenozide against 2nd instar larvae of *S. littoralis* after different exposure periods

Insecticide	Exposure period (h)	LC ₅₀ mg/l	Confidence limits mg/l	Slope ± SD	X ²	Toxicity index
Lufenuron	72	0.106	0.083 - 0.135	1.14 ± 0.13	0.035	1.00
	96	0.069	0.053 - 0.087	1.18 ± 0.13	2.870	1.00
Chlorflauzuron	72	0.275	0.207 - 0.402	1.10 ± 0.13	2.340	2.59
	96	0.199	0.152 - 0.278	1.05 ± 0.13	0.266	2.88
Diflubenzuron	72	0.477	0.320 - 0.898	0.94 ± 0.14	0.285	4.50
	96	0.195	0.150 - 0.269	1.08 ± 0.13	1.370	2.83
Methoxyfenozide	72	0.211	0.183 - 0.245	1.04 ± 0.14	0.235	1.99
	96	0.179	0.153 - 0.223	1.28 ± 0.15	1.870	2.59

Table 2. Toxicity of lufenuron, chlorflauzuron, diflubenzuron and Methoxyfenozide against 4th instar larvae of *S. littoralis* after different exposure periods

Insecticide	Exposure period (h)	LC ₅₀ mg/l	Confidence limits mg/l	Slope ± SD	X ²	Toxicity index
Lufenuron	72	0.889	0.693 - 1.131	1.12 ± 0.14	0.565	1.00
	96	0.453	0.332 - 0.581	1.17 ± 0.13	1.159	1.00
Chlorflauzuron	72	1.352	1.053 - 1.787	1.06 ± 0.14	2.652	1.52
	96	0.944	0.751 - 1.180	1.22 ± 0.14	0.330	2.08
Diflubenzuron	72	3.180	2.326 - 4.957	1.03 ± 0.15	0.263	3.58
	96	1.840	1.435 - 2.492	1.11 ± 0.13	0.640	4.06
Methoxyfenozide	72	1.642	1.493 - 1.961	1.19 ± 0.16	1.765	1.85
	96	0.953	0.772 - 1.345	1.07 ± 0.13	1.134	2.10

Table 3. Pairwise correlation coefficient assessment between log LC₅₀ values of the tested IGRs

Insecticide	Correlation coefficient (r)			
	Lufenuron	Chlorflauzuron	Diflubenzuron	Methoxyfenozide
Lufenuron		0.993**	0.981**	0.993**
Chlorflauzuron			0.986**	0.984**
Diflubenzuron				0.957**
Instar	0.942**	0.936**	0.899**	0.966**

** Correlation is significant at the 0.01 level (2-tailed).

Table 4. Reduction in number of cotton leafworm larvae at different periods of IGRs treatments at Abou Elmatameer 2020 season

Treatments	Period after treatment				
	3-day	5-days	7-days	10-days	15-days
Lufenuron	73.5 a ± 2.8	85.4 a ± 3.3	88.4 a ± 4.1	85.8 a ± 3.1	83.9 a ± 5.7
Chlorflauzuron	64.2 b ± 3.5	74.5 c ± 0.5	77.0 cd ± 3.7	75.0 b ± 2.9	74.5 b ± 2.1
Diflubenzuron	63.0 b ± 2.6	72.5 c ± 4.1	74.3 d ± 2.9	70.8 c ± 3.5	68.2 c ± 3.2
Methoxyfenozide	74.5 a ± 3.1	78.2 b ± 2.3	80.4 c ± 3.1	75.8 b ± 3.7	73.1 b ± 3.0

Numbers within the same column with a letter in common are not significantly different according to analysis of variance (ANOVA)

Table 5. Reduction in number of cotton leafworm larvae at different periods of IGRs treatments at Abou Elmatameer 2021 season

Treatments	Period after treatment				
	3-day	5-days	7-days	10-days	15-days
Lufenuron	75.2 a ± 2.5	84.7 a ± 4.5	87.2 a ± 3.5	85.0 a ± 4.1	81.5 a ± 3.5
Chlorflauzuron	70.5 b ± 4.2	77.3 cd ± 2.2	74.8 b ± 1.9	71.9 b ± 1.8	69.5 b ± 3.2
Diflubenzuron	68.3 b ± 2.1	74.9 d ± 3.7	75.0 b ± 5.1	72.3 b ± 4.2	71.2 b ± 4.3
Methoxyfenozide	74.8 a ± 3.1	79.1 bc ± 2.3	75.7 b ± 3.8	71.4 b ± 3.2	70.5 b ± 3.4

Numbers within the same column with a letter in common are not significantly different according to analysis of variance (ANOVA)

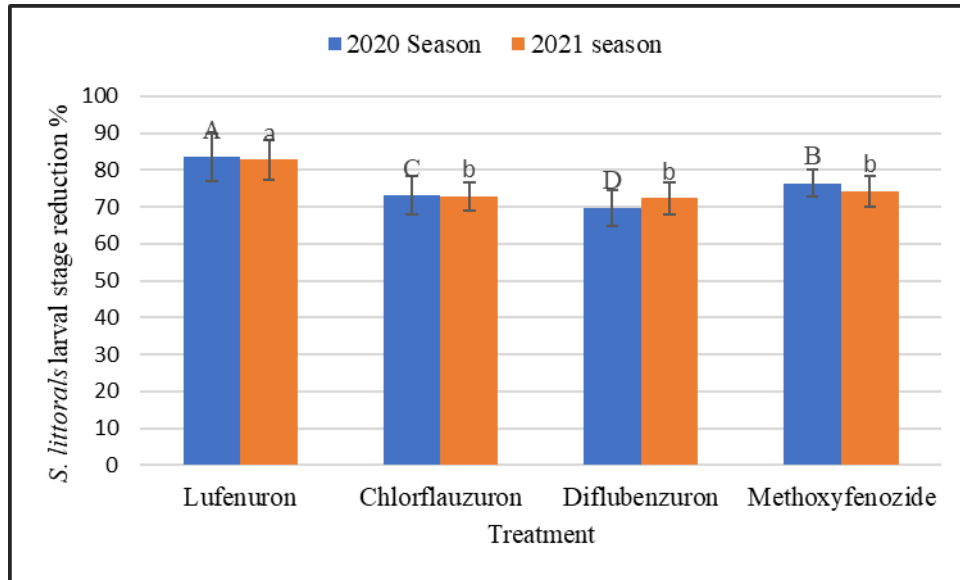


Fig. 1. Average reduction in *S. littoralis* larvae number after fifteen days of IGRs spraying in cotton field across two consecutive seasons

DISCUSSION

The results of this study showed that all four tested IGRs exhibited different levels of insecticidal activity against the second and fourth instar larvae of *S. littoralis*. Lufenuron, in particular, was found to be the most toxic among the tested IGRs, with the lowest LC₅₀ values against both the 2nd and 4th instar larvae after 72 and 96 h of exposure. The other IGRs, Chlorflauzuron, Methoxyfenozide and Diflubenzuron followed Lufenuron in terms of toxicity. However, these differences in toxicity were not significant between Chlorflauzuron and Methoxyfenozide. Moreover, the 2nd instar larvae were more susceptible to all tested insecticides as well a strong relationship between instar and toxicity was calculated in this work, which is in agreement with an earlier published study detailed that the smaller 1st and 3rd instars are more sensitive to IGRs (El-banna *et al.*, 2020). These results are in the line with previous study reported that, Lufenuron was more toxic to *S. littoralis* compared with Hexaflumuron and more toxic to 2nd instar larvae compared with the fourth one

(Seham, 2015). Also El-Sheikh and Aamir (2011) stated that, among three tested IGRs Lufenuron displayed the highest toxicity to both 2nd and 4th instar larvae of *S. littoralis*. However, Tabozada *et al.* (2014) found that, Lufenuron was less toxic than Flufenoxuron to both 2nd and 4th instar larvae of *S. littoralis* using both feeding or dipping bioassay techniques. Furthermore, Methoxyfenozide reported to be less toxic to the 4th Instar larvae of *S. littoralis* than Flufenoxuron while it was more toxic than Flufenoxuron and Teflubenzuron (Sabry and Khedr, 2014). Although among these tested insecticides Methoxyfenozide has different mode of action, a strong positive correlation between it and the tested insecticide was recorded which throw the light of the possibility of cross resistance between them all. This finding supported by what found in a study assessed the resistance and cross resistance between certain IGRs since they reported resistance ratios of (1.23– 5.46) to Lufenuron, (1.69 –6.78) to Chlorflauzuron in field strains collected from life coventrates in Egypt and monitored for three successive years. They also found a significant correlation between Lufenuron and

Chlorfluazuron but non-significant correlation between Lufenuron and hexaflumuron (Mokbel *et al.*, 2019). In contrast, Elhadek (2021) evaluated the resistance status for IGRs in Menofia and Gharbia governorate in Egypt and found no resistance developed in *S. littoralis* field strains comes from both sides. Also, Elhadek *et al.* (2020) reordered very low resistance ratios values to Lufenuron, Chlorfluazuron, and Diflubenzuron in Menofia and Gharbia field strains. Therefore, continuous evaluation of IGRs efficacy and susceptibility are in demand to avoid resistance and cross resistance development.

The field experiment results revealed that, the reduction percentages of larvae after application of tested insecticides were relatively high, indicating that these compounds were effective in reducing the population of *S. littoralis* larvae. However, the reduction percentage of Lufenuron was the highest among the tested compounds, representing that it is the most effective insecticide against the population of *S. littoralis* larvae in Abou Elmatamer. Previous research mention the same finding in which Lufenuron showed the highest mean reduction % of *S. Littoralis* (Mohamed *et al.*, 2019). The effectiveness of Lufenuron continued up to 15 days with silty non-significant decrease. This is in agreement with what recorded by Mohamed *et al.* (2019) as they stated that Lufenuron was effective in *S. littoralis* larvae reduction in field up to 10 days of treatment recording the highest reduction (97%). The same trend of high effectiveness of Lufenuron was found also by El-Kawas and Khedr (2019) they reported that, among all tested pesticides Lufenuron represents the highest initial and mean reduction after 10 days flowed by Methoxyfenozide. Furthermore, Lufenuron reported to cause mean reduction of *S.littoralis* ranging between 78.57 and 80.15% during two cotton seasons, which is close to 82.82% mortality as the of the conventional insecticide chlorpyrifos (Ismail *et al.*, 2023). Other researchers stated that, Lufenuron, Diflubenzuron and Chlorfluazuron resulted in mean reduction percentages ranging between 82.1 and 85.7 after 15 days of treatment (Awad *et al.*, 2014). These results are consistent with the laboratory experiments results, indicating that lufenuron and chlorfluazuron are the most effective IGRs against *S. littoralis*. The field experiments also suggest that multiple applications may be necessary to achieve effective control, and that the effectiveness of the IGRs increases with longer exposure times. The mortality caused by IGRs reported to be due to molting-disruption (Abdel Rahman *et al.*, 2007). The primary cause of this effect is due to the inhibition of chitin formation (Ishaaya & Casida, 1974 and Post *et al.*, 1974), leads to abnormal deposition of the endocuticle and molting frailer (Mulder and Gijswijt, 1973). IGRs compounds are reported to

interfere with the chitin-protein structure of the peritrophic membrane, thereby impeding its function of forming new cuticle and protecting the secreting cells from any harm (Clarke *et al.*, 1977 and Khedr, 2002). Moreover, lufenuron and chlorfluazuron reported to negatively affect fecundity and ovicidal effectiveness of *S. littoralis* (Zidan *et al.*, 2013). The differences in efficacy between the IGRs observed in this study could be due to differences in the mode of action. There is limited information available on the mode of action of these insecticides against *S. littoralis*. However, studies suggest that chitin synthesis inhibitors such as benzoylphenyl urea including lufenuron and diflubenzuron disrupt the formation of the insect cuticle by inhibiting the synthesis of chitin since significant decrease in chitin amount was receded (Merzendorfer, 2013). While, Insect growth regulators, such as chlorfluazuron and Methoxyfenozide, affect the insect's hormonal system by disrupting the synthesis and/or regulation of ecdysone hormone. These substances act by binding to the ecdysone receptor and competing with 20HE, which is a natural hormone involved in the molting process (Berghiche *et al.*, 2008). The results here suggest that Lufenuron is the best choice in this local area. Moreover, as Methoxyfenozide mode of action varied from Lufenuron they can be used in rotation. However, there is possibility for cross resistance which may need further studies at the local condition. Generally, this study light the shad on the importance of continuous evaluation of the susceptibility and field efficacy of the in-use IGRs as well as the resistance and cross resistance status under local condition in particular with the current climate changes situations and the misuse of insecticides.

CONCLUSION

According our results all tested IGRs are effective against *S. littorales* in Abou Elmatamer. Moreover, the difference in susceptibly suggest preference towards Lufenuron. Meanwhile Methoxyfenozide has different mode of action and still effective; thus, under this local condition our study elects it to use in rotation with lufenuron. Furthermore, the results put in light the concern about resistance and cross resistance since the follow up is crucial for making the most suitable decision for selection of the right IGRs that fits the specific target in local area.

REFERENCES

- Abdel Aziz, M. F., and A. R. El-Gabaly. 2021. Toxicological and Biological Studies on Using Lufenuron, Chlorpyrifos, Spinosad and Emamectin Benzoate Insecticides for Controlling Cotton Leafworm, *Spodoptera littoralis* (Boisd.). Egyptian Academic Journal of Biological Sciences, F. Toxicology & Pest Control, 13(2):225-232.

- Abdel Rahman, S. M., E. M. Hegazy and A. E. Elwey. 2007. Direct and latent effects of two chitin synthesis inhibitors to *Spodoptera littoralis* larvae (Boisd). Am. Eurasian J. Agric. Environ. Sci, 2(4): 457-467.
- Amein, N., A. Abdelal and E. Said. 2023. Effectiveness of Teflubenzuron, Emamectin benzoate, and Alfacypermethrin on Fall Armyworm, *Spodoptera frugiperda* (JE Smith) (Noctuidae: Lepidoptera), under Laboratory and Field Conditions. Egyptian Academic Journal of Biological Sciences. A, Entomology, 16(1):133-139.
- Awad, H. A., A. Z. El-Naggar, H. M. EL-Bassouiny and H. M. Tadros. 2014. Efficiency of certain evaluated IGRs and conventional insecticides on the incidence of common lepidopterous insect-pests of cotton plant. Alex. Sci. Exch. J.35: 87-97.
- Berghiche, H., M. Houamria, S. Van de Velde, N. Soltani and G. Smagghe. 2008. Effect of two insect growth regulators on the ecdysteroid contents in eggs of the mealworm. Belgian J. of Zoology. 138(2): 140-145.
- CABI.2022. 'Spodoptera littoralis (cotton leafworm)', CABI Compendium. CABI International. doi: 10.1079/cabicompendium.51070.
- Clarke, L., G. H. R. Temple and J. F. V. Vincent.1977. The effects of a chitin inhibitor Dimilin on the production of peritrophic membrane in the locust, *Locusta migratoria*. Journal of Insect Physiology.23(2): 241-246.
- CoStat Statistical Software. 1990. Microcomputer program analysis version 4.20, CoHort Software, Berkeley, CA. <https://www.cohort.com>
- Damalas, C. A. and I. G. Eleftherohorinos. 2011. Pesticide exposure, safety issues, and risk assessment indicators. International journal of environmental research and public health.8(5):1402-1419.
- Douris, V., D. Steinbach, R. Panteleri, I. Livadaras, J. A. Pickett, T. Van Leeuwen and J. Vontas. 2016. Resistance mutation conserved between insects and mites unravels the benzoylurea insecticide mode of action on chitin biosynthesis. Proceedings of the National Academy of Sciences. 113(51): 14692-14697.
- El-Banna, H. M., M. M. M. A. El-Sabagh, S. M. Abd El-Kareem and S. A. Ibrahim. 2020. Susceptibility of different stages of a field strain of the cotton leaf worm *Spodoptera littoralis* (Boisd.) to two bioinsecticides and two insect growth regulator compounds under laboratory conditions. Uttar Pradesh Journal of Zoology. 41(12):20-27.
- Eldefrawi M. E., A. Topozada, N. Mansour and M. Zeid. 1964. Toxicity studies on the Egyptian cotton leafworm, *Prodenia litura*. I. Susceptibility of different larval instars of *Prodenia* to insecticides. J. Econ. Entomo. 57: 591 – 593.
- El-Guindy, M. A., S. M. Madi, M. E. Keddis, Y. H. Issan and M. M. Abdel Sattar.1982. Development of resistance to pyrethroids in field populations of the Egyptian cotton leafworm, *Spodoptera littoralis* Boisd. Int. Pest Control.24: 6-11.
- Elhadek, M. 2021. Monitoring of Pesticides Resistance in *Spodoptera littoralis* from Some Governorates of Egypt: A Pilot Study Dependent on Biochemical Responses. Egyptian Academic Journal of Biological Sciences, F. Toxicology & Pest Control, 13(1), 25-31.
- Elhadek, M., S. Hafez and R. Ali. 2020. Insecticide resistance and efficacy evolved in field populations of *Spodoptera littoralis* in Egypt cotton season. Egyptian Academic Journal of Biological Sciences, F. Toxicology & Pest Control. 12(1):31-35.
- El-Kawas, H. and M. Khedr. 2019. Performance of Certain Insect Growth Regulators on Cotton Leaf Worm, Sucking Pests and Their Impacts on Common Predators in Egyptian Cotton Fields. Pakistan Entomologist. 41(2):101-110.
- El-sayed, E. K., M. A. Z. Massoud and M. A. Attia. 2017. Biochemical and biological influences of sub-lethal concentrations of emamectin benzoate and certain IGR insecticides against *Spodoptera littoralis* (Lepidoptera: Noctuidae). Alex. Sci. Exch. J. 38. (2):212-219.
- El-Sheikh, E. S. A. and M. M. Aamir. 2011. Comparative effectiveness and field persistence of insect growth regulators on a field strain of the cotton leafworm, *Spodoptera littoralis*, Boisd (Lepidoptera: Noctuidae). Crop Protection. 30(6):645-650.
- FAOSTAT.2023. <https://www.fao.org/faostat/en/#data/QCL> [accessed 4/27/2023]
- Finney D. J. 1971. Probit analysis, 3rd. edition, Cambridge Univ. Press, pp.333.
- Henderson, C. F. and E. W. Tilton. 1955. Tests with acaricides against the brown wheat mite. Journal of economic entomology. 48.(2):157-161.
- Ishaaya, I. and J. E. Casida . 1974. Dietary TH 6040 alters composition and enzyme activity of housefly larval cuticle. Pesticide Biochemistry and Physiology. 4(4):484-490.
- Ismail, S. M., A. S. H. Abo-Shanab and M. A. El-Malla. 2023. Field Evaluation of Certain Compounds Against *Spodoptera littoralis* (Lepidoptera: Noctuidae): Their Impact on its Predator, *Chrysoperla carnea* (Neuroptera: Chrysopidae). Proceedings of the National Academy of Sciences, India Section B: Biological Sciences: 1-6.
- Kai, Z. P., J. Huang, S. S. Tobe and X. L. Yang. 2009. A potential insect growth regulator: synthesis and bioactivity of an allatostatin mimic. Peptides. 30(7):1249-1253.
- Khedr, M. M. 2002. Effect of some plant extracts and insect growth regulators applied to control cotton leafworm on honeybees, *Apis mellifera* (L) (Doctoral dissertation, M. Sc. Thesis, Fac. Agric. Zagazig Univ., Egypt).
- Kodandaram, M. H., A. B. Rai and J. Halder. 2010. Novel insecticides for management of insect pests in vegetable crops: A review. Vegetable Science.37. (2):109-123.
- Ageed, A. A., M. El-bokl, A. A. Khidr and R. Said. 2018. Disruptive effects of selected chitin synthesis inhibitors on cotton leaf worm *Spodoptera littoralis* (Boisd.). Australian Journal of Basic and Applied Sciences, 12(1).
- Merzendorfer, H. 2013. Chitin synthesis inhibitors: old molecules and new developments. Insect science, 20(2): 121-138.
- Mohmed, W., A. E. H. H. Mohana, H. El-Sharkawy and H. AL-Shannaf. 2019. FIELD EVALUATION OF SOME INSECTICIDES AGAINST *Spodoptera littoralis* AND INSECT PREDATOR *Coccinella spp.* IN COTTON FIELDS. Journal of Productivity and Development, 24(4):919-927.

- Mokbel, E. M. S., E. A. Fouad and S. A. El-Sherif. 2019. Resistance monitoring of cotton leaf worm, *Spodoptera littoralis* (Boisduval) (Lepidoptera: Noctuidae) against certain alternative insecticides of four different field populations in Egypt. *J Biol Chem Environ Sci.* 14(1): 319-333.
- Moustafa, M. A., R. I. Moteleb, Y. F. Ghoneim, S. S. Hafez, R. E. Ali, E. E. Eweis and N. N. Hassan. 2023. Monitoring Resistance and Biochemical Studies of Three Egyptian Field Strains of *Spodoptera littoralis* (Lepidoptera: Noctuidae) to Six Insecticides. *Toxics*, 11(3): 211.
- Mulder, R. and M. J. Gijswijt. 1973. The laboratory evaluation of two promising new insecticides which interfere with cuticle deposition. *Pesticide Science.* 4(5): 737-745.
- Official Recommendation book for Agriculture pests control Ministry of Agriculture and land reclamation. 2022. "<http://www.apc.gov.eg/Files/Releases/Recomm22.pdf>"
- Post, L. C., B. J. De Jong and W. R. Vincent. 1974. 1-(2, 6-disubstituted benzoyl)-3-phenylurea insecticides: inhibitors of chitin synthesis. *Pesticide Biochemistry and Physiology*, 4(4): 473-483.
- Retnakaran, A. and J. E. Wright. 1987. Control of insect pests with benzoylphenyl ureas. *Chitin and benzoylphenyl ureas*: 205-282.
- Sabry, H. M. and M. A. Khedr. 2014. Biochemical and histological variations induced by IGRs in *Spodoptera littoralis* (Boisd.). *Glob. J. Environ. Sci. Toxicol.* 1(2): 163-178.
- Salama, H. S., N. Z. Dimetry and S. A. Salem. 1971. On the host preference and biology of the cotton leaf worm *Spodoptera littoralis* (Boisd.). *Zeitschrift für Angewandte Entomologie*, 67(1-4): 261-266.
- Sánchez-Bayo, F. 2012. Insecticides mode of action in relation to their toxicity to non-target organisms. *J. Environ. Anal. Toxicol.* S, 4, S4-002.
- SAS Institute Inc. 1985. *SAS User's Guide: Statistics*, Version 5 Edition. Cary, NC. 956 p.
- Seham, M. I. 2015. Influence of Two Insect Growth Regulators on Chitinase Activity. *Alex. J. Agric. Sci.* 60(3):303-307.
- Singh, N. S., R. Sharma, T. Parween and P. K. Patanjali. 2018. Pesticide contamination and human health risk factor. *Modern age environmental problems and their remediation*: 49-68.
- Spomer, N. A. and J. J. Sheets. 2019. Chitin Biosynthesis and Inhibitors volume III: (1067-1101). In: Jeschke P., Witschel, g Krämer, M.W and Schirmer, U. *Modern Crop Protection Compounds*, 3rd Ed. John Wiley & Sons pp.1784.
- Steel, G. D and J. H. Torrie. 1980. *Principles and Procedures of Statistics: A Biometrical Approach*. McGraw-Hill, New York.
- Tabozada, E. K., S. A. El-Arnaouty and S. M. Sayed. 2014. Effectiveness of two chitin synthesis inhibitors; flufenoxuron and lufenuron on *Spodoptera littoralis* (Lepidoptera: Noctuidae) and side effects of sublethal concentrations of them on two hymenopteran parasitoids. *Life Sci. J.* 11(10): 239-245.
- Tunaz, H. and N. Uygun. 2004. Insect growth regulators for insect pest control. *Turkish Journal of Agriculture and Forestr*
- Zidan_Lobna, T. M., M. H. Rashwan and M. A. A. Abd-El-Razik. 2013. Comparative curative and preventive ovicidal effectiveness of certain selected IGRs and insecticides against the cotton leaf worm and sweet potato whitefly. *New York Sci. J.* 6(2): 83-91.

الملخص العربي

تقييم السمية والكفاءة الحقلية لبعض منظمات نمو الحشرات لمكافحة دودة ورق القطن على نباتات القطن

ميرفت حسنين أبو الحمد مطاوع و منال أحمد محمد عطية

بالقطع التجريبية الغير معاملة. على الدوام أظهر الوفينيرون أعلى خفض للتعداد بعد ٣ أيام من المعاملة (٧٣,٥% و ٧٥,٢%) وكذلك متوسط الخفض فى نهاية التجربة بعد ١٥ يوم من المعاملة (٨٣,٩% و ٨١,٥%) فى كل من الموسم ٢٠٢٠ و ٢٠٢١ (على التوالي). على النقيض من ذلك، تم تسجيل أدنى فعالية لمبيد ديفلوبنزورون بأقل متوسط نسبة خفض للتعداد (٦٨,٢% و ٧١,٢%) بعد ١٥ يوماً من المعاملة في موسمي القطن لعامي ٢٠٢٠ و ٢٠٢١ على التوالي. علاوة على ذلك أظهرت النتائج وجود علاقة طردية بين لوغاريتم الجرعات المسببة لموت ٥٠% من الأفراد المعاملة لكل مبيد المبيدات الأخرى المختبرة مما يشير إلى احتمال أن دودة ورق القطن يمكنها أن تتطور فيها مقاومة مشتركة بين هذه المبيدات. تلقى نتائج هذه الدراسة الضوء على أهمية اختيار المبيد المناسب الأعلى سمية من مجموعة منظمات نمو الحشرات لمكافحة فعالة ومستدامة ليرقات دودة ورق القطن. هذا ويؤكد البحث على خطورة المتابعة فى تقييم كفاءة وثبات وايضا تطور المقاومة لهذه المبيدات تبعا للظروف المحلية.

الكلمات المفتاحية: لوفينورون، ميثوكسيفينوزايد، بنزوفينايل يوريا، محفزات مستقبل الإكديزون، مكافحة دودة ورق القطن على نباتات القطن، سمية منظمات نمو الحشرات.

تتطلب إدارة مكافحة دودة ورق القطن تنفيذ استراتيجيات التحكم الفعالة والمستدامة. هذه الدراسة تهدف إلى مقارنة سمية والكفاءة الإبادية فى الحقل لأربعة من منظمات نمو الحشرات الشائعة الاستخدام فى مكافحة دودة ورق القطن للمساعدة فى إتخاذ القرار الأمثل فى الحصول على مكافحة فعالة طبقا للظروف المحلية. المبيدات المختبرة شملت (البنزوفينايل يوريا لوفينورون، كلورفلوآزورون، ديفلوبنزورون) ومبيد من محفزات مستقبل الاكديزون (ميثوكسيفينوزايد). اجريت اختبارات التقييم الحيوى لتقدير سمية هذه المبيدات على الأعمار(الثانى والرابع) لدودة ورق القطن. أظهر مبيد لوفينورون أعلى سمية حيث كانت قيمة الجرعة المتسببة فى قتل ٥٠% من الأفراد هى الأقل مقارنة بباقي المبيدات المختبرة (٠,٠٦٩ & ٠,١٠٦ مجم/لتر) عند ٧٢ و ٩٦ ساعة بعد التعرض للمبيد على التوالي. اما كل من مبيدات كلورفلوآزورون و ديفلوبنزورون و ميثوكسيفينوزايد كانت أقل سمية حيث كان مؤشر السمية اعتمادا على سمية الوفينيرون على العمر الثانى و المنخفضة نسبياً كمرجع (١,٥٢ و ٣,٥٨ و ١,٨٥) على التوالي. كما تم إجراء التجارب الحقلية خلال موسمين قطن متتاليين لتقييم كفاءة تلك المبيدات -سالفة الذكر- فى خفض تعداد يرقات دودة ورق القطن على نباتات القطن فى الحقل. نتج عن جميع المعاملات خفض معنوى لتعداد الحشرة مقارنة