The Journal of Basic and Applied Zoology

RESEARCH Open Access

Assessment of the perturbation induced by chitin synthesis inhibitors lufenuron, flufenoxuron and hexaflumuron in the house fly, *Musca domestica* vicina (Diptera: Muscidae)

Mohammad A. Tanani[1], Ahmed I. Hasaballah[1] and Rehab M. Hussein[2*]

Abstract

Background: The larvicidal and biochemical effects of chitin synthesis inhibitors (CSIs), namely lufenuron, flufenoxuron and hexaflumuron against the newly molted penultimate instar larvae of the house fly *Musca domestica*, were investigated.

Methods: Different concentrations from each tested compound were applied on forty individuals of *M. domestica* 2nd instar larvae. Four replicates were used for each concentration.

Results: The recorded LC_{25} and LC_{75} values were (166.11, 68.33 and 56.43 ppm) and (732.33, 283.02 and 248.45 ppm) for lufenuron, flufenoxuron and hexaflumuron, respectively. The results showed significant ($P < 0.05$) increase of mortality in larvae treated with different tested CSIs compounds. Mortality was greater in larvae treated by hexaflumuron than lufenuron and flufenoxuron. The main metabolites were tested in the larval whole-body tissue homogenate and findings could be summarized as follows: tested concentrations of CSIs (a) predominantly reduced the total carbohydrate, protein, lipid and cholesterol content at certain ages tested. (b) Disturbed the total carbohydrate content particularly for larvae treated with LC_{75} concentration of hexaflumuron. (c) Exerted the protein and lipid profiles and this effect was much more pronounced in larvae treated with hexaflumuron. (d) Reduced the quantitative cholesterol content and this reduction was found to be increased with development.

Conclusions: Tested CSIs in particular hexaflumuron showed remarkable larval toxicity and reduced the main metabolites content in the larval whole-body tissue homogenate of the house fly, *M. domestica*.

Keywords: Larvicides, Lufenuron, Flufenoxuron, Hexaflumuron, *M. domestica* vicina

Background

The house fly, *M. domestica* (Diptera: Muscidae), is a well-known livestock pest of human health importance. It constitutes a worldwide problem wherever poor sanitation and bad hygienic conditions exist (Chintalchere et al., 2020; Hasaballah, 2021). Moreover, the biology and ecology of dipterous flies make it ideal organisms to carry and disseminate human and animal pathogens such as helminth parasites, protozoan cysts, viruses and bacteria (Fotedar et al., 1992; Greenberg, 1973; Hasaballah, 2015, 2018).

Insect growth regulators (IGRs) are class of compounds that have the possibility to interfere with insect developmental processes and are safe for the environment. Moreover, this class provides an opportunity to be used as alternatives to conventional insecticides in the management of insect pests of both agricultural and public health importance (Mondal & Parween, 2000). IGRs have

*Correspondence: ratef@sci.cu.edu.eg

[2] Zoology Department, Faculty of Science, Cairo University, Giza 12316, Egypt
Full list of author information is available at the end of the article

been used in different applications and were described as agents that elicit their primary action on insect metabolism, ultimately interfering and disrupting the process of growth, development when applied during the sensitive period of insect development (Ishaaya & Horowitz, 1997).

Biochemical effects of IGRs on carbohydrate, protein and lipid profiles of insects were extensively tested previously (El-Kordy et al., 1995; Tanani et al., 2012; Shaurub et al., 2020). The quantity of lipids available may be a result of the balance between food catch and maintenance, growth and reproduction need. The balance is disturbed by toxic chemicals. Lipids are important energy source for insects that are regulated by neuroendocrine-controlled feed-back loops (Downer, 1985). IGRs have specific target site actions which adversely interfere with the growth, and development of insects such as reproduction and metamorphosis (Khan, 2021). In addition to mortality induced by IGRs, their sublethal effects on biochemical and biological parameters must be considered. Sub-lethal concentrations of IGRs may possess adverse effects to different stages of insect pests through interfering with metabolism.

Flufenoxuron has shown potential for the control of urban pests such as German cockroach, *Blattella germanica* (Reid et al., 1992); *Coptotermes sp.* (Ahmed and French 2013) and *Reticuliternes* spp. (Getty et al., 2000). Lufenuron has been used in animal health markets for the control of fleas, heartworms and fungal infections on domestic dogs, cats and other animals (Dean et al., 1999; Ben-Ziony & Arzi, 2000). Additionally, hexaflumuron is a powerful insecticide that controls different insect pests such as lepidopteran, coleopteran, homopteran and dipteran pests (Rojas & Morales-Ramos, 2004).

The aim of this study was to evaluate the larvicidal activity of certain chitin synthesis inhibitors as well as to study their effects on some metabolic parameters such as total carbohydrate, protein, lipid and cholesterol content in the larval whole-body tissue homogenate of the house fly, *M. domestica* tested at various time intervals.

Methods

Rearing technique

The *Musca domestica* colony was obtained from the Medical Insect Research Centre, Dokki, Giza. Egypt. Adults were allowed free access to sugar, and cotton pads soaked in milk powder dissolved in water (10% w/v). Larvae were reared according to the method described by Krafsur et al. (1985) on a mixture of sterilized bran (38 g), milk powder (2 g) and water (60 ml) and maintained under these laboratory conditions 27 ± 2 °C and 70 ± 5% relative humidity. The culture was maintained for several generations at the Laboratory of Entomology, Animal house, Faculty of Science, Al-Azhar University.

Chemicals and application technique

Chemicals were kindly supplied by the Laboratory of Insecticides, Plant Protection Research Institute, Dokki, Giza. Lufenuron (Match 10% EC-CAS No. CG A-184699), *N*- [2,5-dichlor- o-4-(1,1,2, 3, 3hexa-fluoropropoxyl) phenyl) amino 2,6 diflubenzamide (CA)]. Flufenoxuron (Cascade 10% ECC- AS No.1014-63-69-8), *N*- [4-2-chloro-4-(trifluoromethyl) phenoxy]-2-fluorophenyl] amino] carbonyl]- 2, 6-difluorobenzamide. Hexaflumuron (Consult 10%, ECCAS No. 86479), *N*-(3,5-dichlo-ro-4-(1,1,2,2 tetrafluoroethoxy) phenyl]3-(2,6 difluorobenzoyl) urea. Each compound was dissolved in distilled water to prepare different concentrations of (200, 400, 600 and 800 ppm) for lufenuron and (50, 100, 200 and 400 ppm) for flufenoxuron and hexaflumuron. The larval artificial diet was mixed with different tested concentrations of each compound. Forty individuals of newly 2nd instar larvae of *M. domestica* were immediately transferred to glass jars containing in the treated media. Four replicates were used for each concentration, while the control larvae were fed on an artificial diet mixed with distilled water.

Larval mortality and biochemical effects

Larval mortality percentage was estimated using the equation of Briggs (1960): larval mortality $(\%) = A - B/A \times 100$, where (A) is the number of tested larvae, and (B) is the number of tested pupae. At early 2nd instar larvae, 30 individuals were grouped and weighed for estimating the total carbohydrate, protein, lipid and cholesterol content. Measurements were determined at different time periods (24 h, 48 h and 72 h). These criteria were estimated in the whole-body larval homogenate. Three pools were used as replicates. The collected samples were homogenized by using a fine electric homogenizer tissue grinder for 3 min in centrifuge tube containing 0.5 ml of saline solution. Additional 1.5 ml saline was added to make a total volume of 2 ml of saline in each measurement. After centrifugation at 8000 rpm (in cooling centrifuge), the supernatant was transformed for further metabolite measurements using Pye Unicam SP6-450 Uv/Vis 50 spectrophotometer. Determination of the total carbohydrates was processed according to Singh and Sinha (1977). Determination of the total protein was processed according to Bradford (1976). Determination of total lipid was processed according to the method of Knight et al. (1972). Determination of the total cholesterol was made by the enzymatic colorimetric method of Richmond (1973). Results were expressed in mg per gm of fresh body weight.

Statistical analysis

One-way analysis of variance (ANOVA), confidence limits and Chi-square values were estimated using statistical package for social sciences (SPSS ver., 25). Pairwise comparisons between samples were made using Holm Sidak post hoc method. *P* value was considered significant at < 0.05. Larval mortality data were subjected to *probit* analysis to calculate the LC_{25} and LC_{75} values. Results were represented as mean ± SD.

Results

The larvicidal activity of tested CSIs, lufenuron, flufenoxuron and hexaflumuron using different concentrations, was tested against 2nd instar larvae of the house fly, *M. domestica* (Table 1). Larval mortality increased significantly ($P < 0.05$) with tested concentrations. Hexaflumuron exhibited the most pronounced larval control pattern with complete larval mortality achieved at 400 ppm, while the same mortality percentages were achieved at (1000 ppm for lufenuron) and (> 400 ppm for flufenoxuron). From the *probit* analysis results, LC_{25} and LC_{75} values recorded (166.14 and 732.33 ppm), (68.33 and 283.02 ppm) and (56.43 and 248.45 ppm) for lufenuron, flufenoxuron and hexaflumuron, respectively.

According to the data assorted in Table 2, the total carbohydrate content in the untreated larvae whole-body tissue homogenate increased with developmental time and it recorded (25.6 ± 1.7; 27.3 ± 1.8; and 29.4 ± 1.6 mg/g fresh body weight) at 24 h, 48 h and 72 h, respectively, while total carbohydrate content remarkably decreased after treatment with lufenuron, flufenoxuron and hexaflumuron. The most notable reduction was for hexaflumuron (- 43.1) for larvae tested 72-h post-treatment. In general, considerable reduction in carbohydrate content was noticed in almost all treatments (Table 2).

As regard to total protein content, no specific trend was detected for the control congeners tested at different time intervals, while CSIs severely induced protein profile particularly for larvae tested 48-h and 72-h post-treatment. Hexaflumuron was comparatively the most potent tested CSI with notable increased protein content. From another point of view, treatment with LC_{75} concentration slightly promoted the total protein content of treated larvae with different tested CSIs. In Table 3, the obtained data revealed increased stimulation of the total protein content for larvae tested 48-h and 72-h post-treatment, but the most notable effect was recorded for larvae tested after 48 h of hexaflumuron treatment (+ 30.8).

Table 1 Larvicidal activity of lufenuron, flufenoxuron and hexaflumuron against the penultimate instar larvae of the house fly, *Musca domestica*

Treatments	Concentrations (ppm)	Larval mortality (%)	Regression equation	LC_{25} (LCL–UCL) (ppm)	LC_{75} (LCL–UCL) (ppm)	χ^2
Lufenuron	Control	0.0 ± 0.0a	$Y = 0.0883X + 10.333$	166.114 (131.847–204.847)	732.335 (645.223–815.608)	9.338 n.s
	200	26.67 ± 2.27b				
	400	46.67 ± 1.77bc				
	600	66.33 ± 1.73 cd				
	800	76.67 ± 3.52de				
	1000	100.0 ± 0.0e				
Flufenoxuron	Control	0.0 ± 0.0a	$Y = 0.2329X + 9.0854$	68.332 (63.492–72.109)	283.017 (238.019–326.082)	5.319 n.s
	50	13.33 ± 0.78b				
	100	36.67 ± 1.54c				
	200	60.0 ± 2.0d				
	300	83.67 ± 1.37e				
	400	96.67 ± 3.12e				
Hexaflumuron	Control	0.0 ± 0.0a	$Y = 0.2252X + 10.041$	56.425 (50.545–63.379)	248.450 (221.993–281.604)	4.164 n.s
	50	20.0 ± 1.0b				
	100	33.633 ± 1.58b				
	200	56.67 ± 1.33c				
	300	76.67 ± 1.78d				
	400	100.0 ± 0.0e				

Larval mortality presented as Mean ± SD of three replicates, means with different letters are significantly different $P < 0.05$. (LC_{25}) concentration that kills 25% of population, (LC_{75}) concentration that kills 75% of population, (LCL) lower confidence limit, (UCL) upper confidence limit, χ^2 Chi-square, (n. s.) not significant at Alpha = 0.05

The total lipid content of larvae treated with LC_{25} compared with the control is summarized in Table 4. Data showed a marked decrease in the total lipid content induced by lufenuron, flufenoxuron and hexaflumuron compounds for larvae tested 24-h, 48-h and 72-h post-treatment (Table 4). Hexaflumuron showed the highest

Table 2 Effect of lufenuron, flufenoxuron and hexaflumuron on the total carbohydrate content of the house fly *M. domestica* penultimate larval instar treated with the LC_{25} and LC_{75} concentrations at different time intervals

Concentrations	Tested (CSIs)	Total carbohydrate (mg/g fresh body weight)					
		24 h	Change (%)	48 h	Change (%)	72 h	Change (%)
LC_{25}	Lufenuron	23.6 ± 1.3	− 7.81	19.2 ± 1.5	− 29.6	24.5 ± 1.7	− 16.6
	Flufenoxuron	21.4 ± 1.7	− 16.4	18.3 ± 1.6	− 32.9	22.8 ± 0.9	− 22.4
	Hexaflumuron	22.7 ± 1.8	− 11.3	20.5 ± 1.8	− 24.9	24.7 ± 1.4	− 15.9
	Control	25.6 ± 1.7	–	27.3 ± 1.8	–	29.4 ± 1.6	–
LC_{75}	Lufenuron	20.8 ± 1.2	− 18.7	22.6 ± 1.3	− 17.2	18.9 ± 1.7	− 35.7
	Flufenoxuron	21.4 ± 1.4	− 16.4	21.8 ± 1.7	− 27.4	19.8 ± 1.4	− 32.6
	Hexaflumuron	17.7 ± 1.5	− 30.8	19,8 ± 1.5	− 28.9	16.7 ± 1.7	− 43.1
	Control	25.6 ± 1.7	–	27.3 ± 1.8	–	29.4 ± 1.6	–

Table 3 Effect of lufenuron, flufenoxuron and hexaflumuron on the total protein content of the house fly *M. domestica* penultimate larval instar treated with the LC_{25} and LC_{75} concentrations at different time intervals

Concentrations	Tested (CSIs)	Total protein (mg/g fresh body weight)					
		24 h	Change (%)	48 h	Change (%)	72 h	Change (%)
LC_{25}	Lufenuron	22.8 ± 2.5	+ 21.3	37.6 ± 2.5	+ 18.2	54.8 ± 3.2	+ 4.9
	Flufenoxuron	24.7 ± 2.2	+ 10.2	39.7 ± 3.6	+ 17.3	53.8 ± 2.5	+ 3.4
	Hexaflumuron	26.3 ± 1.3	+ 20.4	40.5 ± 2.3	+ 19.5	59.7 ± 3.4	+ 14.3
	Control	22.4 ± 2.2	–	35.7 ± 3.8	–	52.2 ± 4.9	–
LC_{75}	Lufenuron	22.6 ± 2.3	+ 0.8	44.2 ± 3.6	+ 23.8	56.7 ± 2.2	+ 8.7
	Flufenoxuron	23.7 ± 2.5	+ 5.8	39.6 ± 3.4	+ 10.9	55.8 ± 2.5	+ 6.8
	Hexaflumuron	26.9 ± 2.4	+ 23.3	46.7 ± 3.2	+ 30.8	61.5 ± 4.7	+ 17.8
	Control	22.4 ± 2.2	–	35.7 ± 3.8	–	52.2 ± 4.9	–

Table 4 Effect of lufenuron, flufenoxuron and hexaflumuron on the total lipid content of the house fly *M. domestica* penultimate larval instar treated with the LC_{25} and LC_{75} concentrations at different time intervals

Tested concentrations	Tested (CSIs)	Total lipid (mg/g fresh body weight)					
		24 h	Change (%)	48 h	Change (%)	72 h	Change (%)
LC_{25}	Lufenuron	39.2 ± 2.1	− 7.5	29.6 ± 2.2	− 39.2	32.5 ± 4.2	− 45.2
	Flufenoxuron	36.4 ± 1.7	− 14.1	31.6 ± 3.8	− 35.1	29.8 ± 2.7	− 59.8
	Hexaflumuron	34.5 ± 26	− 18.6	25.6 ± 2.4	− 47.4	26.7 ± 4.4	− 55.1
	Control	42.4 ± 1.7	–	48.7 ± 3.4	–	59.4 ± 3.8	–
LC_{75}	Lufenuron	32.2 ± 2.7	− 24.1	34.8 ± 3.9	− 28.5	32.4 ± 2.3	− 45.4
	Flufenoxuron	33.9 ± 2.3	− 20.0	34.7 ± 3.2	− 28.7	34.6 ± 2.7	− 41.7
	Hexaflumuron	29.4 ± 2.1	− 30.6	36.7 ± 2.8	− 24.6	29.8 ± 3.7	− 49.8
	Control	42.4 ± 1.7	–	48.7 ± 3.4	–	59.4 ± 3.8	–

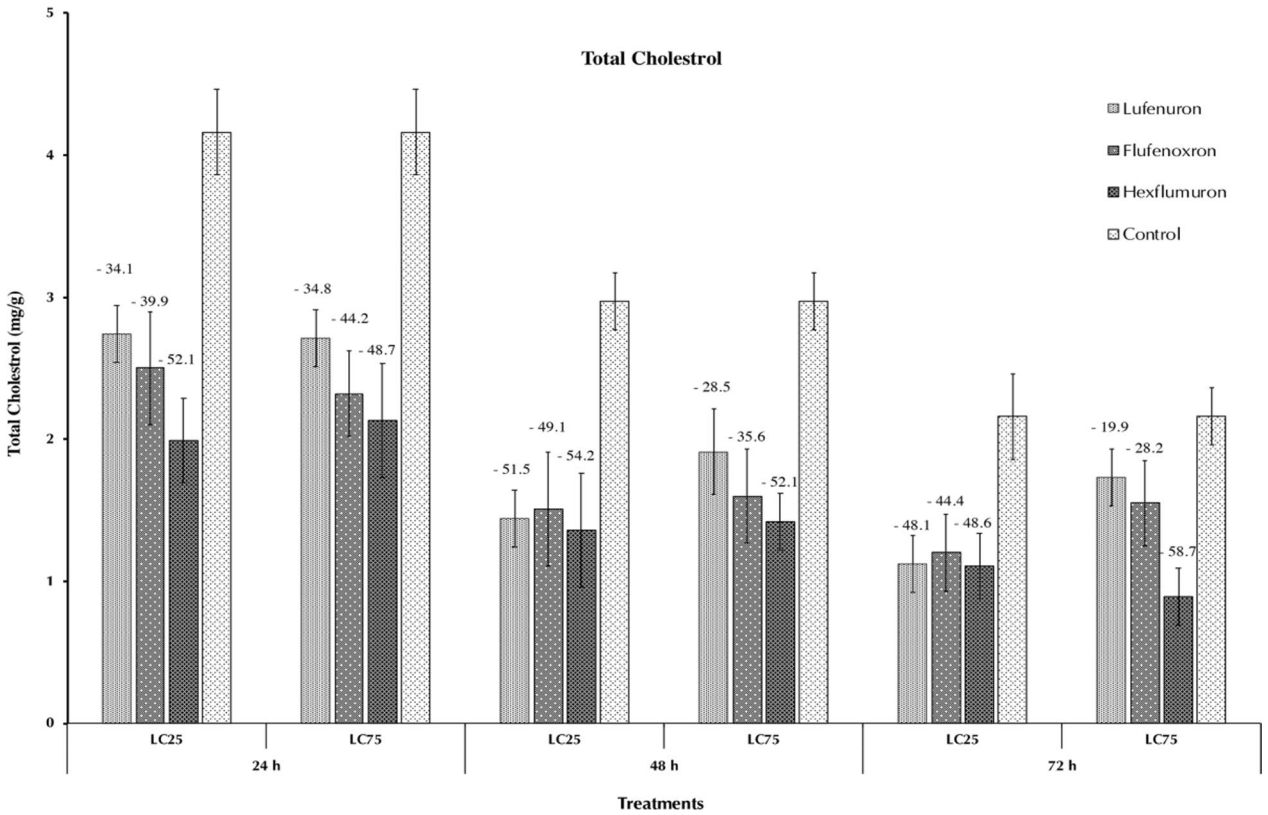

Fig. 1 Effects of LC_{25} and LC_{75} concentrations of lufenuron, flufenoxuron and hexaflumuron on the total cholesterol content of the house fly, *Musca domestica* 2nd larval instar tissue homogenate tested at various time intervals. The change percentages are included upon each column. Data presented as mean with SD of three replicates

reduction percentage (−47.4) for larvae tested 48-h post-treatment, while flufenoxuron recorded the highest reduction percentage (−59.8) for larvae tested 48-h post-treatment. In addition, the perturbation of the total lipid content was also recorded for larvae treated with LC_{75} concentrations. The total lipid content was obviously declined regardless of the tested compounds. The prevalent reducing potency of each compound was gradually increased with the progression of time intervals. The most potent reduction of lipid profile was recorded for hexaflumuron compound in larvae tested 48-h and 72-h post-treatment with reduction percentages of (-24.6 and—49.8), respectively, compared to the control congeners.

As clearly shown in Fig. 1, tested CSIs unexceptionally exhibited inhibitory effect on the total cholesterol content in fresh body weight 2nd instar larvae. Also, tested CSIs reduced the total cholesterol content, and this decrease was found to be increased with development of treated 2nd instar larvae. Hexaflumuron deteriorated the total cholesterol content, and the maximum reductions (that reached up till -54.2) were exhibited for larvae treated with LC_{25} concentrations. Additionally, LC_{75} concentration exerted the most potent reducing action of the total cholesterol content in the fresh body weight for larvae tested at different time intervals. Total cholesterol content reduced to (48.7, 52.1 and 58.7%) for larvae tested 24-h, 48-h and 72-h post-treatment, respectively, compared to the control congeners (Fig. 1).

Discussion

Insect growth regulators (IGRs) are characterized by instant biodegradation, low toxicity and safety to surrounding environment. Chitin synthesis inhibitors (CSIs) are IGRs inhibit molting or produce a deficient cuticle and are considered as effective development suppressors for the entire life cycle of insect pests (Hammock & Quistad, 1981). Although their effects on the same species differ excessively, tested compounds, namely lufenuron, flufenoxuron and hexaflumuron, belong to the same group of IGRs and have the same mode of action (Nasr et al., 2020).

The present study showed that larval mortality increased significantly ($P<0.05$) with the increase in

tested concentrations. Hexaflumuron exhibited potential control agent for *M. domestica* larvae with complete larval mortality achieved at concentration of 400 ppm. The recorded LC_{25} were (166.11, 68.33 and 56.43 ppm) and LC_{75} values were (732.33, 283.02 and 248.45 ppm) for lufenuron, flufenoxuron and hexaflumuron, respectively. Similar results were obtained by (Vazirianzadeh et al., 2007) who found that cyromazine and triflumuron have to be applied in house fly larvae control programs; Donahue et al. (2017) found similar results when applied cyromazine against *Musca domestica, Stomoxys calcitrans* and *Fannia canicularis;* Nasr et al. (2020) tested the same CSIs against the house fly, *M. domestica* and found that the least toxicity values of tested sub-lethal concentrations were displayed for lufenuron, while hexaflumuron scored the highest toxicity followed by flufenoxuron.

Insects share with other invertebrates the common pathways of carbohydrate and lipid metabolism. Although much has been presumed based on overt similarities to more extensive studies on mammals and higher taxa, many aspects of intermediary metabolism have been examined in several insects and different insect tissues including synthesis and storage of carbohydrates and fats. Carbohydrates provide energy for muscles, fuel for nervous system, help in fat metabolism and prevent protein from being used as energy. The production or utilization of carbohydrates as affected by tested IGRs was tested previously (Gäde, 2004). The carbohydrate reserves differ with different developmental stages of insect, and synthesis of carbohydrates is usually affected by tested CSIs (Shen et al., 2018; Wang et al., 2018). In this study, three vital CSIs (a class of IGRs) were experimented to determine their effects on the carbohydrate in the fresh body weight of the 2nd larval instar of the house fly, *M. domestica*. Obtained data clearly displayed considerable reduction in the total carbohydrate content as compared with the control. Some previous studies reported elevated carbohydrate content in tested insect species as a response to the action of tested IGRs, such as El-Kordy (1985) using triflumuron and diflubenzuron against *M. domestica;* Abuo El-Ela et al. (1990) who stated that treatment of *M. domestica* larvae with altosid (ZR-515) led to decrease in carbohydrates in 1-day-old pupae and increased in 3- and 5-day-old pupae. Others reported opposite results (El-Kordy et al., 1995; Tanani et al., 2012). Contradictory findings may be due to differences in species sensitivity, potency of the IGRs or the selected developmental stage.

Proteins are biological macromolecules consisting of one or more long chains of amino acid. Protein synthesis is necessary for growth and reproduction. Proteins are essential for cell division and to control chemical reactions in the process of cell metabolism. In the current study, no certain trend recorded for the total protein content of larvae tested 24-h post-treatment. Tested CSIs severely induced protein profile particularly for those larvae tested at 48-h and 72-h post-treatment. Hexaflumuron was comparatively the most potent tested CSI with notable increase in protein content. These results are in agreement with Amer 1990 who found an increase in protein content of *S. littoralis* pupae after larval treatment with the anti-Juvenile hormone mevalonic acid; Basiouny (2000) who estimated considerable increments of proteins throughout different developmental stages of *Muscina stabulans* after treatment of larvae with IKI-7899 and XRD-473; Ul Haq et al., 2010 who recorded increase in protein profile of the fly, *Bactrocera cucurbitae* as a response to the juvenile hormone analogue, methoprene. The explanation of these results obviously indicates the disturbing effect of IGRs or even CSIs on protein profile that may interrupted the hormonal balance or enzymatic hierarchy in insects that reflect their effects on growth, development, morphogenesis reproduction as reported previously (Bakr et al., 2008; Cloyd, 2003). However, these increasing trends in the total protein may be attributed to failure in the insect tissues to uptake the produced and released proteins from fat body and hemolymph during the pupal or adult apolysis.

The total lipid content was found to be decreased as induced by lufenuron, flufenoxuron and hexaflumuron compounds in larvae tested 24-h, 48-h and 72-h post-treatment. The most pronounced effect was recorded for larvae treated with hexaflumuron. This result is in harmony with (Ahmad et al., 1989) who recorded decrease in the total lipid content in cotton leaf worm *Spodoptera littoralis* (boisduval) as a response to Bay Sir-8514 application; Amer et al., 2005 who found that Diofenolan treatments remarkably reduced the lipid content along the pupal stage of *M. domestica*; additionally, Perveen (2011) applied the sublethal doses of Chlorfluazuron at LC_{10} and LC_{30} to determine the reduction in the total lipid content in *Spodoptera litura*; Djemaoun et al., (2015) reported the same results when treated *Blattella germanica* with indoxacarb. In general, tested IGRs pronouncedly interfere with not only the synthesis of lipids but also their mobilization as promoted to be converted into other metabolites or fatty acids that were considered as important indicators for chemical stress (Moreau et al., 2002).

Most insects have cholesterol as their dominant body sterol, which serve as vital cellular membrane constituent that allow rigidity and permeability; additionally, they are essential precursors to steroid hormones that required for ecdysone and other metabolic processes such as molting and metamorphosis. Herein, tested CSIs reduced the total cholesterol content, and this decrease

was found to be development dependent. Higher concentrations exerted reduction of the total cholesterol content in the fresh body weight for larvae tested at different time intervals. Similarly, flufenoxuron treatments caused increased activity of β-esterase, while Alpha-esterase was increased after treatment with hexaflumuron in *S. littoralis* (Mohamed & Azab, 2002); Shakeet and Bakshi (2010) found that monocrotophos and cypermethrin deduce the reduction cholesterol in *Chrotogonus trachypterus*; additionally, the total esterase activity was elevated with acetamiprid, bifenthrin, chlorpyriphos, deltamethrin, emamectin benzoate, fipronil, imidacloprid, profenophos and lufenuron when tested against *M. domestica* (Farooq & Freed, 2018). Overall, lipid turnover is regulated by neuroendocrine-controlled feedback loops that may explain the decrements in the total cholesterol which with other metabolites are essential for molting and further development.

Conclusions

Relatively low lethal concentrations were obtained in particular those of hexaflumuron against the public heath important house fly, *M. domestica*. Based on obtained results, tested chitin synthesis inhibitors CSIs, namely lufenuron; flufenoxuron and hexaflumuron, generally obstructed the constituents of the total carbohydrate, protein, lipid and cholesterol in the penultimate larval instar. Thus, it could be concluded that tested CSIs—which are considered as environmentally safe alternatives to synthetic chemical insecticides—may have a promising role in integrated management programs for the house fly.

Abbreviations

CSIs: Chitin synthesis inhibitors; IGRs: Insect growth regulators; w/v: Weight/volume; ANOVA: Analysis of variance; SD: Standard deviation; ppm: Part per million; LC: Lethal concentration.

Acknowledgements

The authors declare that there is no source of funding to be acknowledged.

Author contributions

AIH and MAT designed the research idea, overall work and wrote the manuscript. RMH helped with the experimental protocols. AIH and MAT worked on calculations and statistical analysis. RMH revised the drafted manuscript and made necessary corrections. All authors read and approved the final manuscript.

Funding

This research did not receive any funding.

Availability of data and materials

The dataset used during the current study is available on reasonable request.

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no conflict of interests.

Author details

[1]Zoology and Entomology Department, Faculty of Science, Al-Azhar University, Nasr City, Cairo 11884, Egypt. [2]Zoology Department, Faculty of Science, Cairo University, Giza 12316, Egypt.

Received: 5 February 2022 Accepted: 5 June 2022
Published online: 17 June 2022

References

Abou El-Ela, R., Guneidy, A. M., El-Shafei, A. M., & Ghali, O. I. (1990). Effects of Altosid (ZR515) on oxygen consumption, carbon dioxide out\put and carbohydrate content of organophosphorous resistant strain of *Musca domestica*. *Journal of the Egyptian Society of Parasitology., 20*(1), 307–318.

Ahmad, Y. M., Ali, F. A., & Mostafa, A. M. A. (1989). Effects of two benzoylphenyl urea derivatives on haemolymph constituents of *Spodoptera littoralis* (Boisd.) larvae. *Alexandria Science Exchange., 10*, 209–220.

Ahmed, B. M., & French, J. R. (2013). The efficacy of flufenoxuron as potential bait toxicant against termites. *African Journal of Agricultural Research., 8*(28), 3688–3694.

Amer, M. S. (1990). Effects of the anti-JH synthesis (Mevalonic acid) on the main metabolites of Spodoptera littoralis. *Egyptian Journal of Applied Science., 5*(1), 82–91.

Amer, M. S., Ghoneim, K. S., Al-Dali, A. G., Bream, A. S., & Hamadah, KhSh. (2005). Effectiveness of certain IGRs and plant extracts on the lipid metabolism of *Musca domestica* (Diptera: Muscidae). *Journal of the Egyptian Academy of Environmental Development., 6*(1), 53–67.

Bakr, R. F., Hussein, M. A., Hamouda, L. S., Hassan, H. A., & Elsokary, Z. F. (2008). Effect of some insecticidal agents on some biological aspects and protein patterns of desert locust, *Schistocerca gregaria* (Forskal). *Egyptian Academic Society for Environmental Development., 9*(2), 29–42.

Basiouny, A. L. (2000). Some physiological effects of certain insect growth regulators (IGRs) on the false stable fly, *Muscina stabulans* (Fallen.) (Diptera: Muscidae) Ph.D. Thesis, Faculty of Science, Al-Azhar University -Cairo, Egypt.

Ben-Ziony, Y., & Arzi, B. (2000). Use of lufenuron for treating fungal infections of dogs and cats: 297 cases (1997–1999). *Journal of the American Veterinary Medical Association., 217*(10), 1510–1513. https://doi.org/10.2460/javma.2000.217.1510

Bradford, M. (1976). Rapid and sensitive method for the quantification of microgram quantities of proteins utilizing the principle of protein dye binding. *Annual Biochemistry., 72*, 248–254.

Briggs, J. N.)1960). Reduction of adult house fly emergence by the effective *Bacillus* sp. on the development of immature forms. *Journal of Insect Pathology, 2*, 418–432.

Chintalchere, J. M., Dar, M. A., & Pandit, R. S. (2020). Biocontrol efficacy of bay essential oil against housefly, *Musca domestica* (Diptera: Muscidae). *The Journal of Basic and Applied Zoology*. https://doi.org/10.1186/s41936-020-0138-7

Cloyd, R. A. (2003). Effect of insect growth regulators on citrus mealybug *Planococcus citri* (Homoptera: Pseudococcidae)-Egg production. *Horticiemce., 38*(7), 1397–1399.

Dean, S. R., Meola, R. W., & Meola, S. M. (1999). Mode of action of lufenuron in adult *Ctenocephalides* felis (Siphonaptera: Pulicidae). *Journal of Medical Entomology., 36*, 486–492. https://doi.org/10.1093/jmedent/36.4.486

Djemaoun, A., Habes, D., & Soltani, N. (2015). Effects of ingested indoxacarb (Oxadiazine) on biochemical composition of ovaries in *Blattella germanica* (Dictyoptera: Blattellidae). *Journal of Entomology and Zoology Studies., 3*(3), 362–366.

Donahue, W. A., Showler, A. T., Donahue, M. W., Vinson, B. E., & Osbrink, W. L. (2017). Lethal effects of the insect growth regulator Cyromazine against three species of filth flies, *Musca domestica, Stomoxys calcitrans*, and *Fannia canicularis* (Diptera: Muscidae) in cattle, swine, and chicken manure. *Journal of Economic Entomology., 110*(2), 776–782.

Downer, R. G. H. (1985). Lipid metabolism. In "Comprehensive insect physiology, biochemistry and pharmacology" Ceds: Kerkut, G.A and Gilbert, L.I., Pbl. *Pergamon Press, Oxford, 10*, 75–114.
El-Kordy, M. W. (1985). Effect of triflumuron and diflubenzuron on larval development and emergence of *Musca domestica*. Ph.D. Thesis, Al-Azhar University, Egypt.
El-Kordy, M. W., Gadallah, A. I., Abas, M. G., & Mostafa, S. A. (1995). Effect of pyriproxyfen, flufenoxuron and teflubenzuron on some biochemical aspects of *S. littoralis*. *Al-Azhar Journal of Agricultural Research., 21*, 223–238.
Farooq, M., & Freed, S. (2018). Mortality, biological, and biochemical response of *Musca domestica* (Diptera: Muscidae) to selected insecticides. *Journal of Entomological Science., 53*(1), 27–45.
Fotedar, R., Banerjee, U., Singh, S., Shriniwas, A., & Verma, A. K. (1992). The house fly *Musca domestica* as a carrier of pathogenic microorganisms in a Hospital Environment. *Journal of Hospital Infection., 20*(3), 209–215. https://doi.org/10.1016/0195-6701(92)90089-5
Gäde, G. (2004). Regulation of intermediary metabolism and water balance of insects by neuropeptides. *Annual Review of Entomology., 49*, 93–113.
Getty, G. M., Haverty, M. I., Copren, K. A., & Lewis, V. R. (2000). Response of *Reticulitermes spp.* (Isoptera: Rhinotermitidae) in northern California to baiting with hexaflumuron with Sentricon termite colony elimination system. *Journal of Economic Entomology., 93*, 1498–1507. https://doi.org/10.1603/0022-0493-93.5.1498
Greenberg, B. (1973). *Flies and disease* (Vol. II, p. 447). Princeton University Press.
Hammock, C. D., & Quistad, G. B. (1981). Metabolism and mode of action of juvenile hormone, juvenoids and other insect growth regulators. In D. H. Hutson & T. R. Roberts (Eds.), *Progress in pesticide biochemistry*. Wiley.
Hasaballah, A. I. (2015). Toxicity of some plant extracts against vector of lymphatic filariasis, *Culex pipiens*. *Journal of the Egyptian Society of Parasitology., 45*(1), 183–192.
Hasaballah, A. I. (2018). Impact of gamma irradiation on the development and reproduction of *Culex pipiens* (Diptera; Culicidae). *International Journal of Radiation Biology., 94*(9), 844–849.
Hasaballah, A. I. (2021). Impact of paternal transmission of gamma radiation on reproduction, oogenesis and spermatogenesis of the housefly, *Musca domestica* L. (Diptera: Muscidae). *International Journal of Radiation Biology*. 1–27.
Ishaaya, I., & Horowitz, A. R. (1997). Insecticides with novel mode of actions: Overview. In I. Ishaaya & D. Degheele (Eds.), *Insecticides with novel mode of actions, mechanisms and application* (pp. 1–39). Springer.
Khan, H. A. A. (2021). Posttreatment temperature influences toxicity of insect growth regulators in *Musca domestica*. *Parasitology Research., 120*, 435–441. https://doi.org/10.1007/s00436-020-06998-5
Knight, J. A., Anderson, S., & Rawle, J. M. (1972). Chemical basis of the sulfo-phospho-vanillin reaction for estimating total serum lipids. *Clinical Chemistry., 18*, 199–202.
Krafsur, E. S., Black, I. V., Church, C. J., & Barnes, D. A. (1985). Age structure and reproductive biology of a natural house fly (Diptera: Muscidae) population. *Environmental Entomology., 14*, 159–164. https://doi.org/10.1093/ee/14.2.159
Mohamed, H.A. and Azab, A.M. (2002). Effect of Insect Growth Regulators and Binary Mixtures on Enzymes Activity of Egyptian Cotton Leaf Worm, *Spodoptera littoralis*, (Boisd) larvae. Proceedings of 2nd International Conference, Plant Protection Research Institute, Cairo Egypt (1): 617–622.
Mondal, K., & Parween, S. (2000). Insect growth regulators and their potential in the management of stored-product insect pests. *Integrated Pest Management Reviews., 5*, 255–295. https://doi.org/10.1023/A:1012901832162
Moreau, R. A., Whitaker, B. D., & Hicks, K. B. (2002). Phytosterols, phytostanols, and their conjugates in foods: Structural diversity, quantitative analysis, and health promoting uses. *Progress in Lipid Research., 41*, 457–500.
Nasr, E. E., Selim, Sh., & Tanani, M. A. (2020). Toxicological and biological indices of the house fly, *Musca domestica* after CSIs treatments lufenuron, flufenoxron and hexaflumuron. *Journal of the Egyptian Society of Parasitology., 50*(2), 322–332.
Perveen, F. (2011). Effects of sublethal doses of chlorfluazuron on the ovarian biochemical constituents of *Spodoptera litura*. *African Journal of Biotechnology., 10*(62), 13598–13604.
Reid, B. L., Appel, A. G., Demark, J. J., & Bennett, G. W. (1992). Oral toxicity, formulation effects, and field performance of flufenoxuron against the German cockroach (Dictyoptera: Blattellidae). *Journal of Economic Entomology., 85*(4), 1194–1200. https://doi.org/10.1093/jee/85.4.1194
Richmond, W. (1973). Preparation and properties of cholesterol oxidase from *Nocardia* sp. and its application to the enzymatic assay of total cholesterol in serum. *Clinical Chemistry., 19*, 1350–1356.
Rojas, M. G., & Morales-Ramos, J. A. (2004). Disruption of reproductive activity of *Coptotermes formosanus* (Isoptera: Rhinotermitidae) primary reproductive by three chitin synthesis inhibitors. *Journal of Economic Entomology., 97*, 2015–2020. https://doi.org/10.1603/0022-0493-97.6.2015
Shakeet, P., & Bakshi, S. (2010). Effect of cypermethrin on the biochemical parameters on gonads of *Chrotogonus trachypterus* (Blanchard) (Orthoptera: Acrididae). *Indian Journal of Agricultural Research., 44*(4), 242–250.
Shaurub, E. S. H., Abdel-Aal, A. E., & Emara, S. A. (2020). Suppressive effects of insect growth regulators on development, reproduction and nutritional indices of the Egyptian cotton leafworm, *Spodoptera littoralis* (Lepidoptera: Noctuidae). *Invertebrate Reproduction & Development., 64*(3), 178–187. https://doi.org/10.1080/07924259.2020.1741454
Shen, D., Wang, L., Ji, J., Liu, Q., & An, C. (2018). Identification and characterization of C-type lectins in *Ostrinia furnacalis* (Lepidoptera: Pyralidae). *Journal of Insect Science., 18*(2), 24. https://doi.org/10.1093/jisesa/iey011
Singh, N. B., & Sinha, R. N. (1977). Carbohydrate, lipid and protein in the development stages of *Sitopheles oryzae* and *Sitopheles granaries*. *Annals of Entomological Science of America., 70*, 107–111.
Tanani, M. A., Ghoneim, K. S., & Hamadah, K. S. (2012). Comparative effects of certain IGRS on the Carbohydrates of hemolymph and fat body of the desert locust, *Schistocerca gregaria* (Orthoptera: Acrididae). *Florida Entomologist., 95*(4), 928–935. https://doi.org/10.1653/024.095.0416
Ul Haq, I., Caceres, C., Hendrichs, J., Teal, P., Wornoayporn, V., Stauffer, C., & Robinson, A. S. (2010). Effects of the juvenile hormone analogue methoprene and dietary protein on male melon fly *Bactrocera cucurbitae* (Diptera: Tephritidae) mating success. *Journal of Insect Physiology., 56*(11), 1503–1509. https://doi.org/10.1016/j.jinsphys.2010.04.018
Vazirianzadeh, B., Jervis, M., & Kidd, N. (2007). The effects of oral application of cyromazine and triflumuron on house-fly larvae. *Iranian Journal of Arthropod Borne Diseases., 1*(2), 7–13.
Wang, Z. L., Wang, X. P., Li, C. R., Xia, Z. Z., & Li, S. X. (2018). Effect of dietary protein and carbohydrates on survival and growth in larvae of the *Henosepilachna vigintioctopunctata* (F.) (Coleoptera: Coccinellidae). *Journal of Insect Science., 18*(4), 3. https://doi.org/10.1093/jisesa/iey067

Publisher's Note