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Evaluation of chlorpyrifos and *Beauveria bassiana* as a strategy in the Egyptian sugar beet fields: impact on *Spodoptera littoralis* (Boisduval) and its associated predators populations and the sugar beetroot yield



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Abstract

Background The susceptibility of pests and natural enemies to conventional insecticides is a critical element in judging the success of integrated pest management programs implementing biological control as a tactic. In this study, the susceptibility of the cotton leafworm *Spodoptera littoralis* (Boisd.) (Lepidoptera: Noctuidae) to an organophosphorus insecticide (chlorpyrifos) and the entomopathogenic fungus *Beauveria bassiana* was evaluated through field applications during two successive sugar beet seasons. The populations of the coccinellid, chrysopid, and formicid predators were estimated to indicate pesticide risk assessment strategy. The root and sugar yields were compared during the whole experiment to emphasize the impact of both control strategies on the pre-harvest loss in this strategic crop.

Results Results indicated that over the course of the two seasons of the investigation, chlorpyrifos significantly reduced the population density of *S. littoralis* and its associated predators. It showed a detrimental effect on *S. littoralis* larval population than untreated areas, causing an overall reduction of 97 and 92% during the 1st and 2nd seasons, respectively. The biopesticide based on entomopathogenic fungus *Beauveria bassiana* caused an overall reduction of *S. littoralis*, reaching 96 and 65% during the 1st and 2nd seasons, respectively. Meanwhile, *B. bassiana* slightly affects the population of predators compared to insecticide-free areas, so it was recommended for application in sugar beet fields without providing considerable risk to the associated insect predators. The sugar beetroot production and its sugar content increased significantly in both treatments during the two growing seasons compared to non-treated plots.

Conclusion Chlorpyrifos is not recommended for pest control; instead, *B. bassiana* is considered an effective biopesticide in the Egyptian sugar beet fields infested with *S. littoralis* as part of an integrated pest management program. Their effect was direct through reducing pest population and indirectly increasing the sugar beetroot production and increasing its total sugar content. Hence, both the sugar beetroot quality and quantity were improved favoring the microbial agent over the chemical insecticide in application for mitigating the chemical hazards toward the associated predators.

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Keywords Sugar beet, Spodoptera littoralis, Chlorpyrifos, Beauveria bassiana, Predators, Control

Background

Sugarcane and sugar beet production was accompanied by cultural and economic importance to Egyptians, especially in Upper Egypt and Delta, respectively. Concerning sugar concentration, beets have 13–18% compared to 11% in sugarcane. The sugar beet (*Beta vulgaris* L., Chenopodiaceae) pests are considered one of the most destructive hazards to the sugar beet industry in Egypt. To compensate for the sugar needs gap, Egypt directed to boosting an alternative crop, the sugar beet, as its water consumption is less than that of sugarcane (Mehareb et al. 2022). Sugar beet crop quality is a much more important attribute than its quantity as an indicator of yield cost (El-Zayat 2022). In this respect, even minor infestation will affect yield cost through an indirect effect on crop quality.

Insect pests of the sugar beet include wireworms, sugar beetroot maggots that cause damage to germinating seeds, seedlings, and roots. Leaf injury is also caused by webworms, grasshoppers, cutworms, armyworms, and leaf miners. All these pests can also transmit plant microbial diseases, such as fungal diseases, leading to crop destruction (Peter 2022). The cotton leafworm Spodoptera littoralis (Boisd.) (Lepidoptera: Noctuidae) is one of the fiercest pests infesting all growing stages starting from seedling up to crop harvest of the sugar beet causing severe surplus losses (El-Fergani 2019). In Egypt, sugar beet is cultivated in August, September, and October. Infestation of the sugar beet with S. littoralis occurs during early plantation (Khalifa 2018), while S. ocellatella (the sugar beet moth) infests late plantations (Shalaby and El-Samahy 2010). Both of them are considered an economically important sugar beet pests. Chemical control strategy relies on both conventional and nonconventional chemicals such as chlorpyrifos, emamectin benzoate, and variable insect growth regulators (Agricultural Pesticide Committee (APC) recommendation 2020). Synchronizing the predatory insect population together with the pest population is the core of the predators' population stability in field crops to guarantee selfpropagation and predator sustainability. To achieve this synchronization, insecticide side effects and pernicious impact on nontarget species of natural enemies should be lowered. Hence, optimizing the application dose in a way to lower the pest population below the economic injury level and maintaining the natural enemy population is necessary.

Predatory species associated with sugar beet pests include members of the Coleoptera (Coccinellidae, Staphylinidae), Hymenoptera (Formicidae), Neuroptera (Chrysopidae), Diptera (Stelzl and Devetak 1999). The redflanked ladybird Scymnus interruptus Goeze (Coleoptera, Coccinellidae), the green lacewings Chrysoperla carnea (Stephens) (Neuroptera: Chrysopidae), and some predatory ant species (Hymenoptera: Formicidae) are important generalist predators for many insect pests including S. littoralis due to their polyphagous feeding behavior (Hegazy 2018). The reason for selecting those species in the present study is that they were recorded to be abundant during the early plantation of the sugar beet (Khalifa 2018). The susceptibility of these predators to conventional and organic pest management programs is a key element in integrated pest management. Fluctuations in both the pest and its associated predators' numbers in sugar beet field are changing as a result of insecticide treatment regimes (El-Dessouki et al. 2014). In recent years, there has been an increasing demand for eco-friendly biological pesticides such as those based on entomopathogenic organisms such as fungi. These biopesticides are commercially available, inexpensive, and environmentally safe. Several studies have targeted the suppressive effect of both conventional insecticides and biological pesticides on both the insect pest and the natural enemy populations to conserve the associated arthropod predators in the natural fauna (Bažok et al. 2016).

In the present study, the cotton leafworm S. littoralis field's population was suppressed using chlorpyrifos, one of the most widely used organophosphorus insecticides and a potential biopesticide product based on entomopathogenic fungus, Beauveria bassiana, through field application during two successive growing seasons (2020-2021). This study also aimed to detect the effect of these recommended pesticides on the population density of the most common associated arthropod predators, C. carnea (eggs and larvae), formicide species and Scymnus interruptus (adults) in sugar beet fields infested with S. littoralis during the early growing season. In addition, the sugar beetroot yield and the sugar content were estimated during application in the two consecutive growing seasons. As a net result, the bestrecommended practice was the one that manages the pest population in a way of supporting the predators' existence as well as preventing direct and indirect sugar beet yield loss in pest management programs in Egypt.

Methods Pesticides tested

- 1. Chlorpyrifos: Organophosphorus compound: For *O*,*O*-Diethyl *O*-(3, 5, 6-trichloropyridin-2-yl) phosphorothioate, cholinesterase inhibitor, 48% E.C., China, the recommended application dose is 11/fed-dan.
- 2. Beauveria bassiana: For entomopathogenic fungus, 1×10^8 CFU/gm, the recommended dose is 250 g/100 l. The bioinsecticide formulated product was obtained from the Bio-insecticide Production Unit, Plant Protection Research Institute, Agricultural Research Centre, Giza, Egypt.

Field experiments

The location of the field treatments was at Kafr El-Sheikh Governorate (Dafarea district, latitude 31.1089° north; longitude 30.9427° east), Egypt. At the end of September, the local sugar beet variety (PLENO) was cultivated in an experimental area of about 168 m^2 using a randomized complete block design during two seasons (2020 and 2021). Equal plots were divided throughout the area (42 m^2 each). For both the treated and untreated areas, four plots were assigned for each treatment. To evaluate the effect of the tested insecticides on the associated predators, two plant rows were left without spraying between plots. The spraying process was carried out once every season at sunset on October 13.

Aqueous suspensions of the recommended dose of both the insecticide and the fungal product were prepared. The applied dose is 2.5 gm/l for *B. bassiana*. The recommended insecticide dose of 1 l/feddan was prepared by dissolving 42 ml of chlorpyrifos (48% E.C.) in 8 l water. A total volume of 8 L was prepared from both pesticides, and the fungal suspension was kept in a fridge (4 °C till use). Water is used in control. Treatments were

Determination of pest and predators' populations

For the determination of the pest and the predators' numbers, the insects were collected manually by visual record inspection and using pitfall trap for ant collection according to Higgins and Lindgren (2012). Numbers of S. interruptus (adults) were estimated using sampling with the sweeping net. Numbers of S. littoralis (larvae) and C. carnea (eggs and larvae) were estimated by visual examination according to El-Dessouki et al. (2014) with some modifications. For S. littoralis sampling, 10 plants/plot/ treatments were selected randomly few hours prior to the first application and after 3, 7, and 10 days of chlorpyrifos and the fungal bioinsecticide application. A total of 40 plants/replicate were examined for larval S. littoralis infestation and predator's number determination. The collected samples were transferred to plastic jars to identification and counting process in the laboratory. Reduction percentage of the S. littoralis larvae was calculated for each treatment throughout the two seasons.

The associated predators *S. interruptus, C. carnea*, and formicide population densities were also estimated before treatment and after 3, 7, and 10 days of treatments.

Root and sugar yield estimation

The Egyptian Ministry of Agriculture's guidelines for standard agricultural practices were adhered to. For both treated and control plots, the roots were weighed post-harvest, the mean root yield estimate was determined as Kg/42m², and the total production was calculated as ton/feddan. A sucrometer apparatus was used to determine the sugar yield (ton/feddan) and the sugar content (%) according to the Association of Official Analysis Chemists (1990) at the laboratory of Sugar Crops Research Department, Sakha Agricultural Research Station, ARC, Egypt.

Statistical analysis

The reduction percentage of infestation was calculated by Henderson and Tilton (1955) formula, as follows:

Reduction % =
$$\left\{1 - \frac{n \text{ in } C \text{ before treatment } \times n \text{ in } T \text{ after treatment}}{n \text{ in } C \text{ after treatment } \times n \text{ in } T \text{ before treatment}}\right\} \times 100.$$

carried out using a motorized knapsack sprayer (20 l total capacity). Normal agricultural practices were applied using the recommended field rate for both pesticides (http://www.apc.gov.eg/ar/APCReleases.aspx). A control group was set using water only. The application dose was one for every replicate/season. A completely randomized design was followed.

where *n* is the insect population, *C* control, and *T* treated.

Data were statistically compared using Students t-test for paired samples and one-way analysis of variance (ANOVA, $P \le 0.05$). A multiple comparison test was applied to test the significance using Minitab statistical package (v.16) (Minitab Ltd., Coventry, UK).

Results

The efficacy of the organophosphorus compound chlorpyrifos (48% E.C.) and the entomopathogenic fungus *B. bassiana* in suppressing early infestation of *S. littoralis* in sugar beet field was studied over the two successive seasons (2020–2021). Simultaneously, the ability of the most associated insect predators, *S. interruptus*, *C. carnea*, and formicide ants, to withstand the effect of both treatments throughout the experiment was indirectly determined by following up the predators' population density in response to insecticide treatment. The population density of *S. littoralis* larvae and the predators was determined before and after treatments, and percentage of the population reduction was determined (Figs. 1, 2, 3, 4 and 5).

Reduction of S. littoralis infestation

The daily reduction percentage in *S. littoralis* larval population density was determined in sugar beet fields, following the application of chlorpyrifos and *B. bassiana* throughout the growing season of 2020 (Fig. 1). Before any treatment, the larval population increased by



3 days 7 days 10 days

Fig. 1 Daily reduction percentage of *Spodoptera littoralis* larval population in sugar beet field after treatments with *Beauveria bassiana* and chlorpyrifos pesticides during the growing seasons (2020–2021)



Fig. 2 Cumulative reduction percentage of *Spodoptera littoralis* larval population in sugar beet field after treatments with *Beauveria bassiana* and chlorpyrifos pesticides during the growing season (2020–2021)

Associated Predators' Population during the First Season



Fig. 3 Population density of the pest- associated predators in sugar beet field after treatments with *Beauveria bassiana* and chlorpyrifos pesticides at 3, 7, and 10 days post-treatment during the first growing season (2020)



Fig. 4 Population density of the pest-associated predators in sugar beet field after treatments with *Beauveria bassiana* and chlorpyrifos pesticides at 3, 7, and 10 days post-treatment during the second growing season (2021)







1.4-fold in a natural infestation pattern. Just one day after treatment, the recommended dose of chlorpyrifos was observed to be highly efficient against S. littoralis larvae compared to the control group, causing an 88% reduction in pest infestation. Moreover, chlorpyrifos treatment recorded more than 90% pest population reduction 3 days post-treatment indicating a potent effect. After three and seven days of treatments, a significant increase in pest reduction percentage was recorded ($P \leq 0.05$), in comparison with the untreated area, ranging from 92.25 to 97.38%, respectively. A high residual effect was inferred as chlorpyrifos caused 100% reduction in pest infestation up to 10 days post-treatment. On the other hand, a gradual decline in population size was recorded after B. bassiana application reaching 40.96, 63.38, and 75.43% after three, seven, and ten days, respectively, compared to the untreated region (Fig. 1).

During the second growing season (2021), the normal infestation pattern in the non-treated areas was insignificantly different from those during the first growing season (2020) during the whole study days ($P \ge 0.05$, Fig. 1). Chlorpyrifos caused 81% initial reduction in *S. littoralis* larval population. A gradual increase in population reduction was achieved reaching 86.36, 92.36, and 97.18% three, seven, and ten days post-treatment, respectively.

On the other hand, *B. bassiana* reduced *S. littoralis* larval population by 45.19, 70.36, and 84.17% at three, seven, and ten days post-treatment, respectively, compared to the untreated area (Fig. 1). In comparison with season 2021, the overall reduction of *S. littoralis* larvae was lower than that recorded in 2020. The overall (cumulative) reduction of the *S. littoralis* larval population upon treatment with chlorpyrifos and *B. bassiana* was 96.54 and 59.92%, respectively, during the first season (2020). The reduction in pest population decreased to 91.96% upon chlorpyrifos application while it reached 66.57% upon application of *B. bassiana* (dF=3, F=5.36, P=0.026, Fig. 2).

Associated predators population reduction

Studying the effect of insecticides on the pest-associated predators' number in sugar beet fields is a deterministic parameter of insecticide safety to nontarget pests. Regarding the impact of treatment on the number of related arthropod predators (*S. interruptus, C. carnea* and formicide ants), the actual predators' numbers were recorded during season 2020 (Fig. 3) and season 2021(Fig. 4).

Surprisingly, during the first season (2020) a complete reduction in *C. carnea* and formicide ants' population density was recorded just one day post-treatment indicating a detrimental effect of the recommended chlorpyrifos dose against those predators. However, by day 10, post-treatment, only 5% of *C. carnea* population was able to compensate for the insecticide toxic effect (Fig. 3). In contrast, treatment with *B. bassiana* reduced the predators' number, but it did not eradicate them; however, it supported the existence of the predators to an extent that guaranteed their sustainable role in pest population control. After only 3 days of treatment, a complete eradication effect of almost all the associated predators was observed during the first season, which is considered as a negative back withdraw for chlorpyrifos application.

The same pattern of the dramatic reduction happened in the next season (2021) with a complete reduction in *C. carnea* and formicide ants upon treatment with chlorpyrifos (Fig. 4). Moreover, *B. bassiana* treatment supported the previous hypothesis in slightly affecting the predators' numbers, thus maintaining a continuous population in a dynamically stable profile.

Considering the overall predators population reduction, *S. interruptus* population was reduced by 41.83 and 59.96% upon treatment with *B. bassiana* during the first and second seasons, respectively (Fig. 5). In contrast, chlorpyrifos application reduced its population by 96.25 and 98.7% during the first and second seasons, respectively (dF=3, F=26.0 P=0.03).

During the two seasons, *B. bassiana* significantly reduced *C. carnea* populations by 38.06 and 56.11% during the first and second growing seasons, respectively, than the non-treated plots. On the other hand, chlorpyrifos drastically suppressed its population with 98.14 and 100% overall reduction in first and second seasons, respectively. Both treatments differed significantly over the two growing seasons (dF=3, F=32.83, P=0.03).

Formicide ants were the most susceptible to the chlorpyrifos recommended dose; the population reduction reached 100%. In contrast, *B. bassiana* reduced the ant population by 65.1 and 36.15% during the first and second seasons, respectively (dF=3, F=11.64, P=0.05, Fig. 5).

Sugar beetroot yield and sugar content

Both the conventional chemical insecticide (chlorpyrifos) and the entomopathogenic fungus (*B. bassiana*) significantly increased beetroot weight ($P \le 0.05$) (Ton/feddan) than the control during both growing seasons (2020–2021). However, there was insignificant difference in root yield between the two treatments during the same season as well as when comparing the first and second season ($P \ge 0.05$, Table 1). Quantitatively, there were about 2.2-fold increase in beetroot weight in terms of tons produced per feddan and kilograms produced per replicate during the first and second growing seasons comparing treatments to control.

Table 1 Effect of the entomopathogenic fungus Beauveriabassiana and the insecticide chlorpyrifos treatments on the sugarbeetroot weight during the two consecutive growing seasons(2021 and 2022)

Treatments	Seasons	Sugar beetroot weight	
		(ton/feddan) Mean±S.E	(kg/replicate) Mean±S.E
Second	231.50 b±0.50	22.04 a±0.05	
Chlorpyrifos	First	$235.50 a \pm 0.28$	22.42 a±0.03
	Second	234.00 ab±0.91	22.28 a±0.08
Control	First	105.00 c±0.91	9.99 b±0.08
	Second	104.75 c±0.63	10.10 b±0.14

Means of the same column followed by different alphabetical letters are significantly different, $P \le 0.05$

Table 2 Effect of the entomopathogenic fungus *Beauveria bassiana* and chlorpyrifos treatments on the beetroot sugar content during two consecutive growing seasons (2021 and 2022)

Treatments	Seasons	Beetroot total sugar content	
		(Kg/feddan)	Percentage (%)
		Mean±S.E	Mean
Beauveria bassiana	First	3.760 c±0.01	17.03 c
	Second	3.814 b±0.02	17.30 ab
Chlorpyrifos	First	$3.854 \text{ ab} \pm 0.01$	17.19 b
	Second	3.880 a±0.03	17.41 a
Control	First	1.303 d±0.02	13.04 d
	Second	1.296 d±0.01	12.99 d

The means of the same column followed by different alphabetical letters are significantly different, $P \le 0.05$

Considering the total sugar content (Kg/feddan), both treatments significantly increased the sugar yield by about 2.9-fold than the control during both the first and second growing seasons ($P \le 0.05$), with a nonsignificant difference between sugar productions per feddan during the two seasons upon chlorpyrifos treatment ($P \ge 0.05$, Table 2). The sugar concentrations also significantly increased by 1.3-fold upon treatment during the two growing seasons ($P \le 0.05$).

Discussion

Sugar beet crop protection, through applying chemical pesticides, is an element in its integrated pest management program. However, environmental impact assessments of this strategy emphasize the detrimental effects on human health, environmental pollution, biodiversity including pests and nontarget natural enemies (Hafeez et al. 2022).

Polyphagous plant defoliators like the cotton leaf worm (*S. littoralis*) cause considerable damage during the initial stages of the vegetation season (Shalaby et al. 2011). Larvae start their feeding superficially yet others enter the root for feeding on the latest instar larvae. Both feeding habits are considered detrimental to the net root yield, especially in the case of sugar beetroot production. Loss due to secondary infections by plant pathogenic microbes is also a cause of impaired yield production through decreasing the physiological process of sugar storage in the root (Hegyi et al. 2007). Thus, proper pest monitoring and management are mandatory in maximizing yield together with other integrated pest management practices.

Herein, the comparative impact of chlorpyrifos insecticide and *B. bassiana* on *S. littoralis* pest population reduction and their impact on the sugar beetroot yield as an indirect effect of pest reduction were studied. The data analysis revealed that the recommended dose of chlorpyrifos should be lowered due to its high potency, severe population reduction within only 3 days as well as its long-lasting residual effect. In all the previous treatments, the total number of *S. littoralis* larvae decreased up to 10 days post-treatments, an effect that is not favored for an insecticide in order not to disrupt the normal balance of pest–natural enemy population that should be synchronized as the natural enemy depends on the pest population in building his population.

The entomopathogenic fungus B. bassiana is an effective tool in biological control programs against infestation with S. littoralis, along with chlorpyrifos insecticide under field conditions. In case the initial degree of infestation was modest, biological management programs using fungal-based biopesticide, in particular, were more successful in stopping pest damage. The effectiveness of fungal-based bioinsecticides depends on the environmental conditions at the time of application, fungal strain, the application method, and the initial pest population density. Recent studies showed a variable fungal potency and hence median lethal times and concentrations in different fungal isolates. A progressive increase in S. litura larval mortality, starting from day 3 of treatment, with B. bassiana was recorded recently assuring its effect (Islam et al. 2023). The fungal-based bioproducts are usually fast in their action as they contain toxins together with fungus spores and conidia. In the present study, the field application of B. bassiana fungal-based bioinsecticide reduced the S. littoralis infestation in sugar beet fields. There was a gradual decline in larval population after 3–7 days. This indicates that the selected product was efficient enough in significantly reducing the pest population, and the biopesticide is a promising tool for acquiring a sustainable pest management in sugar beet cultivation. This was in line with El-Fergani (1999) who mentioned that selected insecticide groups can be used in IPM programs along with biopesticides, which provide good and safe biocontrol strategies for early infestation of *S. littoralis* in the field of sugar beet plants.

Also, chlorpyrifos significantly reduced *Spodoptera exigua* larval populations in sugar beet fields causing an 88 to 92% pest reduction during the growing seasons (2019/20 and 2020/21), respectively (Mansour and Abou-Elkassem 2022). The sugar beet moth (*Scrobipalpa ocellatella*) population was reduced in sugar beet fields upon application of chlorpyrifos and *B. bassiana* (Fergani et al. 2022). The detrimental effect of chlorpyrifos in sugar beet fields extended to cause about 85% population reduction in the tortoise beetle (*Cassida vittata*) and an 89% reduction in its associated predator population (*Coccinella undecimpunctata*) at Kafr El-Sheikh governorate (Anter et al. 2020).

Pest management strategies depend on factors, such as the pest type and its associated predators, farming practices, environmental conditions, and ecological components. Fungal-based bioinsecticides proved to be more potent than various microbial pesticides against many lepidopterous insects such as S. littoralis, S. exigua, and aphids (Zaki and Abdel-Raheem 2010). It may lead to increasing pest mortality and morphogenetic abnormalities in various life stages. It is an efficient substitute for conventional insecticides. It is eco-friendly with minimal side effects on pest-associated arthropods (Fergani and Refaei 2021). Lacewing populations treated with conventional insecticides do not usually exhibit prominent levels of tolerance due to some physiological, morphological, and behavioral characteristics (Cordeiro et al. 2010). A significant advantage of these fungal-based biopesticides is their ability to control pests with minimal side effects on their natural enemies. Therefore, they are recommended to be included in IPM programs.

The indirect impact of the beetroot pest control strategies (conventional and non-conventional) on the root yield and the sugar content was promising. Earlier, the Egyptian agricultural pesticide committee (APC 2020) issued a periodical guide of the recommended insecticides for beetroot pests' management (www.APC.gov. eg). An array of insecticides was recommended for application including chlorpyrifos. Treatments include seed treatments and foliar application. Seed and foliar applications of neonicotinoids reduced sugar beet crop loss due to defoliators feeding activity and curly top disease that is responsible for major crop loss. The root yield and the sucrose content increased by 55–95 and 6.5–7.2%, respectively (Strausbaugh et al. 2012).

Herein, both chlorpyrifos and *B. bassiana* treatments significantly increased the pest population reduction. As an indirect result, the beetroot production and the sugar

content (%) were increased than the control with insignificant differences between both treatments. In addition, chlorpyrifos and the bacterial biopesticide proved their role in providing crop protection of the sugar beet fields infested with *S. exigua* and hence increasing the beetroot and sugar yield (Mansour and Abou-Elkassem 2022).

Conclusion

Although conventional pesticides like chlorpyrifos have a significant lethal impact on the sugar beet pest, S. littoralis, they also have a significant lethal impact on their associated predators. Both chlorpyrifos and B. bassiana reduced the pest-caused beetroot damage, and the total crop and sugar yield was increased. Thus, if used in accordance with the recommended dose and timing of application with standard agricultural practices, fungal-based bioinsecticides should be considered in pest management programs as a safe alternative to suppress the cotton leafworm in Egyptian sugar beet fields. Achieving the sustainable development goals by crop protection for zero hunger, stopping the use of hazardous chemical insecticides for mitigating climate change, and finally the use of eco-friendly biopesticides for protecting the biodiversity in terrestrial life are all outcomes of the present work.

Abbreviations

S. littoralis	Spodoptera littoralis
B. bassiana	Beauveria bassiana
C. carnea	Chrysoperla carnea
S. interruptus	Scymnus interruptus
S. exigua	Spodoptera exigua
W.P.	Wettable powder
CFU	Colony-forming unit
FAO	Food and Agriculture Organization
E.C.	Emulsifiable concentrate
ARC	Agricultural Research Centre

Acknowledgements

Not applicable.

Author contributions

YAF drafted the work, and edited and revised the manuscript. EAR acquired the data. NMF acquired the data and edited the manuscript. HMH contributed to experimental design, statistical analysis, and interpretation of data. All authors read and approved the final manuscript.

Funding

Not applicable.

Availability of data and materials Not applicable.

Declarations

Ethics approval and consent to participate Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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Received: 8 July 2023 Accepted: 27 September 2023 Published online: 02 October 2023

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