Combinations of Bio and Natural Compounds with Trichogramma evanescens (Westwood) for Pieris rapae Management in Cauliflower

Amany Nady Siam and M. Y. El-Kholy

Department of Cutworms and Mole Crickets, Plant Protection Research Institute, Agricultural Research Center, Cairo, Egypt.

Department of Pests and Plant Protection, National Research Center, Dokki, Cairo, Egypt.

Authors’ contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

Article Information

DOI: 10.9734/AJRAF/2023/v9i32216

Open Peer Review History:

This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/100788

ABSTRACT

The efficacy of a bio compound, Protecto (Bacillus thuringiensis) and botanicals extract (mixture of ginger, garlic, hot red pepper extracts and aloe vera) either in combination with Trichogramma evanescens releases or alone was evaluated in comparison to an insecticide, Uphold 36% SC (Methoxyfenozide/ Spinetoram) on Pieris rapae on cauliflower Brassica oleracea. Field experiments were conducted at Atfih District, Giza Governorate, Egypt in September 2020 and October 2021. Results revealed that, all the tested compounds combined with Trichogramma releases were more effective in reducing P. rapae population than when used alone. B. thuringiensis with Trichogramma releases caused 96.91% and 95.32% reduction in larval population in 2020 and 2021 respectively while the reduction in Pieris pupal pupation was 70.87% and 71.59% respectively during these two trials. Similar results were observed with botanical extracts. Parasitization by T.
Keywords: Compatibility; Trichogramma evanescens; Bacillus thuringiensis; botanicals extract; cauliflower; Pieris rapae; sustainable production.

1. INTRODUCTION

Agriculture faces several challenges like rapid exploitation of the natural resources, the huge use of insecticides and the sudden changes in the global climate, in addition to the need for the production of safe, pesticides-free agricultural products mainly the edible vegetables [1,2]. All these challenges push researchers and those responsible for public health to work on new, safe, available and eco-friendly alternatives to the synthetic pesticides for suppressing pests invading agricultural crops [3]. One of the important vegetable crops is the cauliflower (Brassica oleracea). This economically important crop is seriously attacked by the cabbage white butterfly Pieris rapae L. (Family: Pieridae). The pest is the most abundant during all the seasons, where the female lays solitary, yellowish eggs, hatching into nasty, green larvae which considered the most serious and destructive stage causing great economic loss in the yield [4] and [5,6]. Continuous application of broad spectrum synthetic insecticides has resulted in the development of the pest resistance, destruction of beneficial insects and contamination for the environment [7]. Currently, great attention has been drawn to the use of various biological agents such as the parasitoids and entomopathogens instead of synthetic insecticides [8], [9], [10] and [11]. Trichogramma parasitoids are considered the most efficient and promising organisms in biological control programs [12]. As the damage caused by P. rapae reduces the crop production, the intervention with Trichogramma parasitoids with compatible bio and natural compounds is imperative to conserve naturally occurring bioagents and to harvest healthy and safe produce [13,14,15]. The entomopathogen Bacillus thuringiensis is considered as an efficient biological control agent resulting in significant reduction in the pest and in the use of chemical insecticides [16]. The components of a botanical extract [17] such as ginger, garlic, and hot red pepper, and aloe vera possess an insecticidal effect against many insects [18], [19], [20] and [21]. The botanicals mixture was found to be effective against certain pests [17] and was safe to Trichogramma evanescens Westwood under laboratory conditions [22]. The integration of the parasitoids with bio and natural compounds was earlier evaluated by Oatman et al. [23] and Losey et al. [24] who found that, T. evanescens with B. thuringiensis was effective against Helicoverpa zea Boddie and Ostrinia nubilalis (Hubner). Because of the legal restrictions on the edible fresh vegetables and the periodical sprays of different insecticides, alternatives have to be introduced to suppress P. rapae [25,26]. Therefore, this work was carried out to evaluate the efficacy of B. thuringiensis, the botanicals mixture extract and combinations with T. evanescens releases against P. rapae infesting cauliflower crop.

2. MATERIALS AND METHODS

Experiments were carried out to evaluate the integration of T. evanescens Westwood releases with certain bio and natural compounds for the management of P. rapae on cauliflower. Comparisons were made with an insecticide. Preparation of the botanicals extract and the rearing processes were carried out at the Trichogramma Mass Rearing Laboratory of Fayoum Governorate, Plant Protection Research Institute, Agricultural Research Center, Egypt.

2.1 Rearing T. evanescens

The rearing of the host Sitotroga cerealella and the parasitoid T. evanescens were conducted at 25±2 °C and 70±5% R.H. The host S. cerealella Olivier was reared as the modification of Hassan [27]. The parasitoid was multiplied on S. cerealella eggs.

2.2 Preparation of the Botanicals Extract

The botanicals extract was prepared as per the methods described by Siam and Othman [17]. About one kilogram of garlic bulbs, dry hot pepper, fresh ginger rhizomes were cleaned,
ground well, soaked in ten liters water for 48-72 h and filtered. About one kilogram of aloe vera leaves were cut into small pieces, blended and added to the filtrate. This mixture was diluted to 100 liters water for spraying.

2.3 Field Experiments

Field experiments were carried out at Atfih District, Giza Governorate, Egypt in cauliflower seasons of 2020 and 2021. The experiments were conducted on 20 replicates randomized block design with six treatments and the control with three replications. Each replicate was ¾ feddan and each plot was separated with three rows as a buffer zone from the adjacent plot to reduce interference of the treatments. The experimental plots were planted on cauliflower seedlings (Colonia variety) on 25th September. Each plot had 10 rows of cauliflower plants which was 50 cm a side within the rows. Plants were inspected for the presence of eggs, larvae and pupae of *P. rapae* every 10 days until harvest on 5th December. Pre-treatment population count was made randomly from half of the rows. Plants had eggs of *P. rapae* was marked with coloured paper cards. Larvae and pupae were also counted. The first release of *T. evanescens* was done after 20 days of transplanting the seedlings. Hard paper envelopes housing the parasitoids were hung in the canopy of the cauliflower plants at the rate of 60 envelopes/ feddan (about 90000 parasitoids/each release). Each envelope had three pieces of different developmental stages of *T. evanescens*. The envelopes were hung every 10 days at the sunset. Normal cultivation practices were followed in cauliflower field.

The treatments were: Releasing *T. evanescens* alone, spraying *B. thuringiensis* alone, spraying the botanicals extract alone, combination of *T. evanescens* with *B. thuringiensis*, combination of *T. evanescens* with the botanicals extract, the chemical insecticide Uphold 36% SC and the control (without any treatment). Back motor sprayer (Agranomodo- China) of 20 liters was used for spraying the compounds. Unparasitized and parasitized *Pieris* eggs, number of both larvae and pupae of *P. rapae* were the criteria used for the evaluation of the treatments. Upon appearance of *Pieris* larvae, sprays of the tested compounds were applied in the early morning with the recommended rates as following:

1) *Bacillus thuringiensis* sub sp kurstaki 9.4% WP at the rate of 300 gm/200 L. (Product of Plant Protection Research Institute Agricultural Research Center, Egypt.)
2) The prepared botanicals extract at the rate of 1L/100L.
3) Uphold 36% SC at the rate 250cm/200L. (Commonly used by farmers in the neighboring fields in case of heavy infestation with the pest)

2.4 Data Collection and Statistical Analysis

The *Pieris* eggs collected from the plots were observed every 10 days to record the parasitism (the black color of the eggs). The efficacy of the treatments was assessed through the percent reduction of *Pieris* population. It was calculated according to Henderson and Tilton [28].

\[
\text{Reduction percentage} = \left(1 - \frac{\text{Population of pest after treatment}}{\text{Population of pest before treatment}}\right) \times 100
\]

Obtained data of treated plots were compared with those of Uphold insecticide and control plots. Statistical analysis was carried out by IBM-SPSS for windows (v.25). Variance Analysis (ANOVA) was used to process the obtained data. Duncan's multiple range test [29] was used for separation of means [30].

3. RESULTS

3.1 Efficiency of Tested Compounds on *P. rapae*

3.1.1 Reduction percentages of *P. rapae* larvae

The bio compound *B. thuringiensis* combined with *T. evanescens* releases was significantly more effective in reducing *P. rapae* larvae population in 2020 and 2021 (P<0.05) with reduction percentages of 96.91% and 95.32% respectively. This was followed by the natural compound the botanicals extract combined with the parasitoids releases causing the reduction in *Pieris* larvae population by 96.31% and 94.89% in 2020 and 2021 respectively. The chemical insecticide Uphold was the least effective compound in reducing the larvae population with respective reductions of 37.85% and 47.85% (Fig. 1, 3).
3.1.2 Reduction percentages of *P. rapae* pupae

The tested compounds significantly differed in its effectiveness against the population density of *P. rapae* pupae (P<0.05). The botanicals extract accompanied with *T. evanescens* releases was effective in reducing pupae of *P. rapae* on cauliflower plants in 2020 and 2021 growing seasons with reduction percentages of 77.4% and 72.63% respectively, followed by the treatment of combination of *B. thuringiensis* with *Trichogramma* releases with reduction of 70.87% and 71.59% in 2020 and 2021 respectively. The least reductions of 26.12% and 14.04% in *P. rapae* pupal population in 2020 and 2021 respectively were recorded in plots sprayed with the chemical insecticide Uphold. (Fig 2, 4).
3.1.3 Parasitism percentage

There was no parasitism rate recorded before any treatment in 2020 and 2021. With the successive releases of *T. evanescens*, increase in the parasitism was observed in parasitoid-released plots. Results presented in Tables 1, 2 revealed that the plots treated with *T. evanescens* and *B. thuringiensis* had the parasitism rates of 92.56% (2020) and 92.17% (2021). Also, *T. evanescens* releases with the botanicals extract recorded parasitism rates of 91.95% (2020) and 92.77% (2021). Results revealed that releases of *Trichogramma* parasitoids alone without any compound recorded parasitism rates of 91.03% (2020) and 89.99% (2021). The remaining treatments plots showed no parasitism rate as there no released parasitoids (Tables 1, 2) proving no natural parasitism.
Table 1. Parasitism by *T. evanescens* on *P. rapae* eggs (2020)

<table>
<thead>
<tr>
<th></th>
<th><em>B. thuringiensis</em></th>
<th>Botanicals extract</th>
<th><em>T. evanescens</em> + Bt</th>
<th><em>T. evanescens</em> + botanicals extract</th>
<th>Insecticide</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>unparas.</td>
<td>P.</td>
<td>%Paras</td>
<td>unparas.</td>
<td>P.</td>
<td>%Paras</td>
<td>unparas.</td>
</tr>
<tr>
<td>7.66</td>
<td>0</td>
<td>8.33</td>
<td>0</td>
<td>8</td>
<td>0</td>
<td>8.67</td>
</tr>
<tr>
<td>3</td>
<td>8.66</td>
<td>74.27</td>
<td>5.67</td>
<td>0</td>
<td>0</td>
<td>5.66</td>
</tr>
<tr>
<td>2</td>
<td>12.33</td>
<td>86.04</td>
<td>6.33</td>
<td>0</td>
<td>0</td>
<td>6.67</td>
</tr>
<tr>
<td>0.67</td>
<td>0</td>
<td>12.33</td>
<td>94.85</td>
<td>5.67</td>
<td>0</td>
<td>6</td>
</tr>
<tr>
<td>0</td>
<td>0</td>
<td>12.33</td>
<td>100</td>
<td>6</td>
<td>0</td>
<td>6</td>
</tr>
<tr>
<td>0</td>
<td>13.33</td>
<td>100</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>1.67</td>
</tr>
<tr>
<td>2.22</td>
<td>9.83</td>
<td>91.03</td>
<td>5.67</td>
<td>0</td>
<td>0</td>
<td>5.67</td>
</tr>
</tbody>
</table>

Table 2. Parasitism by *T. evanescens* on *P. rapae* eggs (2021)

<table>
<thead>
<tr>
<th></th>
<th><em>B. thuringiensis</em></th>
<th>Botanicals extract</th>
<th><em>T. evanescens</em> + Bt</th>
<th><em>T. evanescens</em> + botanicals extract</th>
<th>Insecticide</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>unparas.</td>
<td>P.</td>
<td>%Paras</td>
<td>unparas.</td>
<td>P.</td>
<td>%Paras</td>
<td>unparas.</td>
</tr>
<tr>
<td>3.33</td>
<td>7.67</td>
<td>69.73</td>
<td>7.33</td>
<td>0</td>
<td>0</td>
<td>7.67</td>
</tr>
<tr>
<td>2.67</td>
<td>13.11</td>
<td>83.08</td>
<td>7.11</td>
<td>0</td>
<td>0</td>
<td>8.33</td>
</tr>
<tr>
<td>0.33</td>
<td>15.33</td>
<td>97.89</td>
<td>6.67</td>
<td>0</td>
<td>0</td>
<td>5.67</td>
</tr>
<tr>
<td>0</td>
<td>15.76</td>
<td>100</td>
<td>7.33</td>
<td>0</td>
<td>0</td>
<td>6.67</td>
</tr>
<tr>
<td>0</td>
<td>16.33</td>
<td>100</td>
<td>7.33</td>
<td>0</td>
<td>0</td>
<td>7.67</td>
</tr>
<tr>
<td>2.44</td>
<td>13.62</td>
<td>90.14</td>
<td>7.24</td>
<td>0</td>
<td>0</td>
<td>7.34</td>
</tr>
</tbody>
</table>

144
4. DISCUSSION

The impact of integration between synthetic or natural compounds with bio control agents like Trichogramma parasitoids on the latter should be considered. The present work in 2020 and 2021 growing seasons indicates that incorporating either B. thuringiensis or the botanicals mixture extract might be compatible with T. evanescens for suppressing P. rapae populations infesting cauliflower plantations. Obtained results revealed that releasing T. evanescens with B. thuringiensis could suppress P. rapae larvae and pupae populations and resulted in the highest parasitism rates of Pieris than those treated by releasing Trichogramma parasitoids alone, that's because Trichogramma females parasitizes fresh eggs of the pest and B. thuringiensis kills larvae of the pest. Our results are in concurrence with those of Lundgren et al. [31] who evaluated releasing Trichogramma parasitoids with and without B. thuringiensis; their results confirmed that releasing only Trichogramma parasitoids was less effective than releasing them with B. thuringiensis for controlling Lepidopteron pests. Also, Suzanne et al. [32] supported our observations with their work on controlling P. rapae in organic cabbage crop. They reported that B. thuringiensis with Trichogramma releases was the most effective treatment as it caused high parasitism rate of Pieris eggs with a remarkable reduction in larval populations which resulting in a significant increase in the crop yield over the treatment with Trichogramma alone or controls. Other authors worked on the combined use of different species of Trichogramma parasitoids and B. thuringiensis on different crops aiming at the control of different Lepidopteron pests. Losey et al. [24] proved high efficiency of the combination of T. nubilalis and Bt., for controlling Ostrinia nubilalis H. (Lepidoptera: Pyralidae) infesting sweet corn. The combination of T. evanescens with Agerin (B. thuringiensis) was highly effective against Cryptolabes gnidiella Mill infesting grape orchards as it caused the highest parasitism than those with the parasitoids only [33]. AlSaedi et al. [34] reported that B. thuringiensis was not harmful for the parasitoids as proven by the highest parasitism rates of two species of Trichogramma parasitoids on Tuta absoluta infesting tomato in greenhouse. T. evanescens was highly effective on suppressing the Cydia pomonell (L.) in apple plantations when the parasitoid releases were combined with B. thuringiensis than either of the agents being used alone [35-37]. The results from the current study revealed that B. thuringiensis had no adverse effect on Trichogramma. Earlier studies agreed with our findings as those of Hwang et al. [38] who stated that, the combination of Trichogramma ostriniae (Peng and Chen) and Bt reduced the pest population significantly over either of the bio agent alone [25,26]. Ksentini et al. [39] found that among B. thuringiensis, deltamethrin and Spinosad, only Bt was harmless to the immature developmental stages of T. cacoeciae Marchal, T. bourarache Pintureau and T. evanescens Westwood in pomegranate orchard. Spore mixture of B. thuringiensis did not affect the parasitization performance or longevity of the T. evanescens adults [40]. Similarly, [41] found that B. thuringiensis was safe to T. evanescens and was able to suppress T. absoluta infesting tomato fields at Fayoum Governorate, Egypt by releasing T. evanescens and spraying B. thuringiensis. The present study declared that, the use of B. thuringiensis as alone recorded reduction in Pieris larvae and pupae populations. Our findings are consistent with an earlier studies by Varma et al. [42], Khan et al. [43], Furlong et al. [44] and Maxwell and Fadamiro [45] who reported that B. thuringiensis was significantly effective against Pieris larvae while El-Fahharany and Hendawy [46] reported that Bio Guard (B. thuringiensis) was the least effective compound against P. rapae.

In this work, the combination of the botanicals extract with T. evanescens releases did not affect the parasitism despite its lethal effect on Pieris larvae. Siam and El-Genaidy [22] reported that the botanicals extract was safe to the emergence, fecundity and the general productivity of the T. evanescens. The same botanicals extract had a bio pesticide property [17], effective against mango scale insect and was recommended as safe , easy prepared extract and an alternative to synthetic insecticides.

5. CONCLUSION

Either B. thuringiensis or the botanicals extract in combination with T. evanescens releases has the potential to suppress P.rapae populations on cauliflower plants and could be used as alternative for synthetic insecticides.

COMPETING INTERESTS

Authors have declared that they have no known competing financial interests or non-financial interests or personal relationships that could have appeared to influence the work reported in this paper.
REFERENCES


33. Tohamy TH, et al. Biological control of the Honey Dew Moth Cryptoblabes gniidiella Mill in grape orchards using the local egg parasitoid Trichogramma evanescens (Westwood) and Agerin (Bacillus thuringiensis) compared with recommended insecticides in Middle Egypt. Assiut.J.of Agric.Sci. 2007;38(3):93-111.


42. Varma GC, Bindrs OS, Singh D, Darshan S. Comparative efficacy of Bacillus thuringiensis Berliner formulations against Pieris brassicae Linnaeus. Current Science, Department of Entomology, Punjab University, Ludhiana. India. 1974;43:734-735.

44. Furlong MJ, Ju KH, Su PW, Chol KJ, Chang II, Zalucki P. Integration of endemic natural enemies and Bacillus thuringiensis to manage insect pests of Brassica crops in North Korea. Agriculture, Ecosystems & Environment. 2008;125(1-4):223-238.
