Journal of Population Therapeutics & Clinical Pharmacology

RESEARCH ARTICLE
DOI: 10.53555/jptcp.v31i4.5811

EFFICACY OF DIFFERENT IPM TOOL AGAINST PYRILLA PERPUSILLA UNDER LABORATORY AND FIELD CONDITIONS

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ABSTRACT

Sugarcane is among the chief cash crops and sole sugar crop in Pakistan. Burgeoning population has increased the sugar demand that needs more sugarcane yields. The attack of insect's pests is one of the reasons that creates a grave hurdle to enhance the yields. *Pyrilla perpusilla* is one of the major insects of sugarcane that needs management to achieve the goal. This study was planned to minimize the threat of *P. perpusilla* by utilizing different control tools i.e., cultural, bioagents (*E. melanoleuca*), botanical extracts (Neem, Sophora and Tobacco) and synthetic chemicals (Acetamiprid) in laboratory and field. During lab experiment when the *E. melanuleuca* was tested against the different life stages of *P. perpusilla*, which result showed that the younger nymphs had significantly higher nymphal mortality, which decreased as the host grew older. When the parasitoid was introduced to the first stage host nymphs, they died completely. However, nymphal death rates were 95.52, 78.55, 41.04, and 40.44 percent in the second, third, fourth, and fifth nymphal stages, respectively. Furthermore, in the latter phases, the survival of surviving nymphs was greatly extended. None of the parasitized nymphs in the first to fourth stages grew up to be normal adults. Although 18.0% of parasitized fifth nymphal stage adults were normal, their lifespan was much shorter than that of their un-parasitized counterparts. When male and female leafhopper adults were fed to the parasitoid, mortality of parasitized and non-parasitized individuals was found to be significantly different in both sexes. There was also a parasitoid-induced increase in the longevity of the parasitized adults who survived. In laboratory when *P. perpusilla* was treated with botanical extracts and acetamiprid for 24 hours. Acetamiprid killed all the insects. Among the botanical extracts, Sophora extract resulted in maximum mortality than Neem and Tobacco. When same spraying of chemical and botanical extracts was performed on the insects in the field, the chemical (Acetamiprid) resulted in highest mortality percentage followed by Saphora, Neem and Tobacco after 15 days of spray. The best performing botanical extract was used in combination with biological and cultural control comparing with other practices. The combination of cultural and biological practices with Saphora extract and Acetamiprid separately performed statistically better than all other combinations in declining pest population and enhancing cane yield. These findings depicts that the control of *P. perpusilla* at initial stage with the use of *E. melanoleuca* results in declining the population while the use of eco-friendly botanical extracts for the control of insects at field level is a better management approach. These findings are recommended to incorporate in future IPM programs.

Key word: *Pyrilla perpusilla*, Sugarcane, *Epicarnia melanuleuca*, IPM

Introduction

Sugarcane is one of the world's most significant crops, accounting for approximately one-third of global sugar production (Dotaniya and Datta 2014; Choudhary *et al*., 2016). It is cultivated in more than 109 countries on an area of 26.9 million hectares (M ha), with an annual harvest of 1.91 billion tonnes (bt) (Factfish, 2015). Pakistan ranks 5th among top cane producers in the world (Sarwar *et al*., 2010) and it is a major cash crop in Pakistan. Cane and sugar production are expected to reach 87.67 million tonnes and 7 million tonnes, respectively, in 2021-22, on an area of around 1.2 million hectares (FCA, 2021; USDA, 2021).

Yield reductions of almost 80-85% in Pakistan are precisely attributed to insect and pest attacks especially the top-borer (15-20%), Gurdaspur borer (10-20%), and more peculiarly Pyrilla (30-35%) (Zubair *et al*., 2006). *Pyrilla perpusilla* Wlk. recently been declared endemic pest (Rasul *et al*., 2014; Yaseen *et al*., 2021) and is a grave threat to Pakistan's sugar economy. Both larvae and adults of Pyrilla sucks cell sap, suggestively impacting sugarcane productivity (Kumar and Yadav, 2006). During the winter, Pyrilla survives by feeding the alternate hosts i.e., wheat, barley, and oats. Pest have 3-4 generations and remains active throughout the year with the peak of activity during July to September (Shah and Saleem, 2002). The yield is harmed by an early infestation during the cane's peak growth period, whereas a late infestation, beginning in September, mostly impacts the sugar content of the crop (Puri and Siddharth, 2001).

Sugarcane losses can only be reduced by protecting the cane crop from insect pests with a scientifically developed IPM program. IPM is the application of the most appropriate approaches (Mechanical, cultural, varietal, biological, chemical, sex-pheromone, and light trap procedures) while causing the least amount of disruption to the ecosystem. Pesticides are used when needed, along with cultural practices, resistant cultivars, and the introduction and conservation of natural enemies. Pesticides is significant component of IPM but will be used sparingly because of resistance development in insects, environmental and health hazards. Because of insecticide hazards the attention is being diverted to find other solutions to this complex dilemma.

The abundance of bio-active chemical compounds found in plants makes botanical insecticides a viable alternative to synthetic chemical insecticides (Miresmailli and Isman, 2014; Pavela, 2016). Aside from their insecticidal potential, they are less harmful to the environment and human health than synthetic pesticides (Pavela, 2014; 2016). The pesticidal characteristics of plant compounds have been proved to be specific to specific target species, biodegradable to non-toxic products, and possibly suited for use in an integrated pest management (IPM) program (D'Incao *et al*., 2013). Many of these plants are employed by farmers in impoverished nations (Walia and Koul, 2008). Furthermore, only a small number of these plants have been tested for insecticidal efficacy, and many of the studies were incomplete or employed improper bioassay methodologies (Isman, 2013).

Biological control agents against *P. perpusilla* have been found all over the world. *Epicarnia melanoleuca* (Fletcher) is one of them. It is an important nymphal and adult ectoparasitoid of *P. perpusilla*, which is found in India, Sri Lanka, and Pakistan (Srikanth *et al*., 2016; Ganehiarachchi and Fernando 2006; Yaseen *et al*., 2021). *P. perpusilla* nymphs and adults are caught and held by its hooked claws when they pass by. Their sharp mandibles pierce through the host's skin and they eat the body fluid. As soon as the larvae are done with their host body, they spin white, oval-shaped cocoons on the leaves before they become adults. The host (*P. perpusilla*) dies as soon as the parasitoid comes out and eats it (Rajak *et al*., 2007).

Pyrilla perpusilla has a negative impact on the sugarcane crop, as evidenced by the above facts. Therefore, it is need of time investigate several safe management practices for the control of *P. perpusilla*. The novelty of study is to control *P. perspusilla* effectively and safely by using different plant extracts (especially *Sophora alopecuroides* which is so far not used against *P. perpusiila*, insect predators and chemical insecticides in integrated pest management strategy.

Materials and Methods

Different experiments were performed to test the efficacy of Cultural, Biological botanical extracts and their different combinations under laboratory and field conditions during the year of research study (2021).

Laboratory experiments

Laboratory experiment was carried out in Biological Control Laboratory (Insectary) at NARC from July to September 2021. A little modified methodologies used by (Bal *et al*., 1990; Anwar *et al*., 1992; Sharma and Shera, 2021) was followed to test the efficacy of *Epicarnia melanuleuca,* botanical extracts, and standard insecticides against *P. perpusilla* under lab conditions.

Host Plant Sugarcane culture

The sugarcane variety CP77/400 obtained from SCRI was brought to the insectary and sown in the greenhouse. The greenhouse was fully covered from all sides to trap insect inside. All the agronomic practices was kept constant throughout the experimental period. Irrigation was applied as per requirement and recommended application of NPK fertilizers was made at 2-month interval. Pests were not controlled chemically during the entire research period.

Host insect (*P. perpusilla*) culture

Pyrilla perpusilla eggs cluster along with leaf were collected carefully from the sugarcane field at SCRI. The collected eggs were shifted carefully to the insectary in small plastic boxes and were stapled on sugarcane leaves at the greenhouse. The eggs cluster were monitored daily (without disturbing) till the emergence of nymphs and adults. The emerged nymphs were left undisturbed to enhance the population which were used in further experiments.

Parasitoid culture (*Epicarnia melanuleuca)*

The *E. melanuleuca* (eggs) was collected from well-maintained culture. These eggs were kept in an insect growth chamber at 28^{o}C and 75±5 RH in insectary and were observed daily till the larval emergence. The emerged larvae were used for further subsequent studies.

Botanical extracts and insecticides

The botanical extracts *viz,* Neem root extracts, Sophora plant extracts, and tobacco extract were obtained from the already prepared stock solution from IPMP Laboratory while the insecticides (Acetamiprid) was obtained from the local market. All these Botanical extracts and acetamiprid were used @ 3ml/ liter of water in further laboratory and field experiment.

Experiment 1.

To investigate the efficacy of *Epicarnia melanuleuca* against *Pyrilla perpusilla* (nymphs and adults) under laboratory conditions

Efficacy of *Epicarnia melanuleuca* against *P. perpusilla* (nymphs)

Nymphs (from each of the five instars) were removed from the culture (3.2) and were placed into rearing sets with an average of eight individuals per set. The insects were parasitized artificially using glass vials containing parasitoid larvae that had been raised in the lab (as mentioned in 3.3) and placed in the rearing setups for a period. A different group of nymphs showing symptoms of parasitism were segregated and raised for the purpose of documenting the mortality of host as nymphs, intermediates (nymph-adult or adult-nymph) or weak adults (not fully developed or less active), nymphal longevity and survival as normal adults. A set of un-parasitized nymphs was also kept for comparison.

Efficacy of *Epicarnia melanuleuca* against *Pyrilla perpusilla* (Adults)

Adults were parasitized using the same approach as for nymphs to examine the effect of the parasitoids on the adults. To compare longevity, 10 pairs of each of the parasitized and healthy adults, one pair per set, was released in rearing sets at a time

Data Analysis

All data are presented as mean ± standard error of the mean (SE). The percentage data on mortality of parasitized nymphs and survival as normal adults were transformed prior to analysis of variance (ANOVA) for normal distribution and meet the assumptions of ANOVA. Data on longevity of parasitized nymphs were normally distributed (goodness-of-fit test) and were analyzed using ANOVA. The means were separated using LSD test @ 0.05% probability. All statistical tests were carried out using "IBM SPSS 16.0 version.

Experiment 2

Testing the efficacy of Botanical extracts and standard insecticide against *Pyrilla perpusilla* Nymphs (4th instar) under laboratory conditions

The efficacy of Botanical extracts and standard insecticide against *Pyrilla perpusilla* (4th Nymphs) were tested under laboratory conditions. The sugarcane leaves measuring 8 x 2 cm was cut (10 for each treatment) from sugarcane culture and were kept vertically in small plastic vial containing water wet cotton swab. The cut leaf surfaces were completely treated with the appropriate plant extracts and pesticides by camel hairbrush painting technique while the control set were kept untreated. These vials containing treated leaves were further kept in transparent cage having mesh cloth at both ends. In each cage 10 nymphs (collected from 3.2) were released and were allowed to feed on treated leaves. This experiment was repeated 10 times for each treatment and were arranged in Completely Randomized Design. Data were recorded by observing the mean percent mortality at 12-hour intervals for total of 36 hours.

Statistical analysis

A statistical software package (IBMS-SPSS) was used to analyze the recorded data, and the means was compared using LSD with a 95 percent probability.

Experiment 3

To test the efficacy of Botanical extracts and standard insecticide against *Pyrilla perpusilla* (Nymphs and adults) under Field conditions

This experiment was be performed in the vicinity of Agriculture Research Station, Harichand, Charsadda during the year 2021. The methodology used by Chand *et al*., (2019) was followed with slight modifications.

Field preparation

The plot size (19 × 9) meters was prepared. The whole plot was sub divided into three equal blocks. Each block was further divided into five equal sub-plots size (3.8 × 2.4) meters. There were four rows in each subplot (having equal distance) and a total of eight sugarcane cut buds were sown of sugarcane variety (CP-77/400) `in all plots. The treatments (Acetamiprid, Neem root extracts, Sophora plant extracts, and tobacco extract and control) were randomly applied to each subplot. The experimental design was randomized complete block design with three replications. The mentioned treatments were applied with recommended rates when the pest population reached an economic threshold level in the field (July-September). The data regarding the mean percent mortality was collected from all the plots after 3-, 5-, and 7-days post-treatment application respectively.

Data Analysis

The recorded data was subjected to analysis by using statistix software 8.1 version and means were compared by using LSD test @ 0.05 probability.

Experiment 4

Integrated management of *Pyrilla perpusilla*

This experiment was also performed in the research area of Agriculture Research Station, Harichand, during July-August 2022. The methodology used by Rasul *et al*., 2014 was followed with slight modifications.

Field preparation

The plot size (47 × 12) meters was prepared. The whole plot was sub divided into three equal blocks which was further divided into 10 equal sub-plots size (3.8 × 2.4) meters. There was buffer zone of 2 meters between each block and each sub plot. Each sub plot was consisting of four rows having equal distance (74 cm) and a total of eight sugarcane cut buds were sown of sugarcane variety (CP-77/400) `in each row of all plots in the month of February 2020.

Treatments applications

The following treatments were applied alone and with possible combination

1. **Culture Control (T1):** Manual detaching/dtrashing of eggs culture with camel hairbrush, Fortnightly removal of dry leaves, destructions of weeds/ alternative host
2. **Biological Control (T2):** Releasing/ placing of *Epicarnia melanuleuca* cocoon (near to emergence) @ 2800- 3000 ha^{-1} one a month (July-August)
3. **Chemical control (T3):** Chemical (Acetamiprid) application @ 3ml $litre^{-1}$ with 15 days interval during the July and August
4. **Botanical extracts (T4):** Application of Sophora Extracts 3% with 15 days interval during the July and August
5. **Cultural + Biological (T5)**
6. **Culture + Chemical (T6)**
7. **Culture + Botanical Extracts(T7)**
8. **Cultural + Biological+ Chemical (T8)**
9. **Cultural + Biological+ Botanical (T9)**
10. **Control (T10)**

Yield data

At the end of the season the cane yield was obtained from the all treated plots in Kg ha^{-1} which was then converted in Tons ha^{-1}.

Experimental design and Analysis

All the treatments were randomly arranged in RCB Design with three replications. The data was recorded fortnightly and were analyzed with SPSS. The means were compared using LSD test with 95% probability.

RESULTS

To investigate the efficacy of Epicarnia melanuleuca against Pyrilla perpusilla (nymphs and adults) under laboratory conditions

Mortality as nymphs, intermediate forms, or weak adults

There was a statistically significant difference in nymphal mortality between nymphs of different ages that were fed to the parasitoid. The illustration against nymphs depicts the first nymphal stage where mortality was 100%, followed by stages where mortality was 95.52, 78.58, 41.05 and 40.44% respectively. The mortality of intermediate form depicts the fifth nymphal stage where mortality was 56.34 percent. After nymphal stage three it was 19.74%; after nymphal stage four, it was 15.23%; after stage two, it was 4.84%. The death of parasitized nymphs as weak adults was also observed after the nymphal stage was completed, suggesting they perished as weak adults after the nymphal period was over. It was estimated that 32.27% of nymphs died by the fourth stage, 14.72% by the fifth, 9.36%

by the third, and only 4.88% by the second nymphal stage due to age-related mortality as a weak adult (Fig. 1).

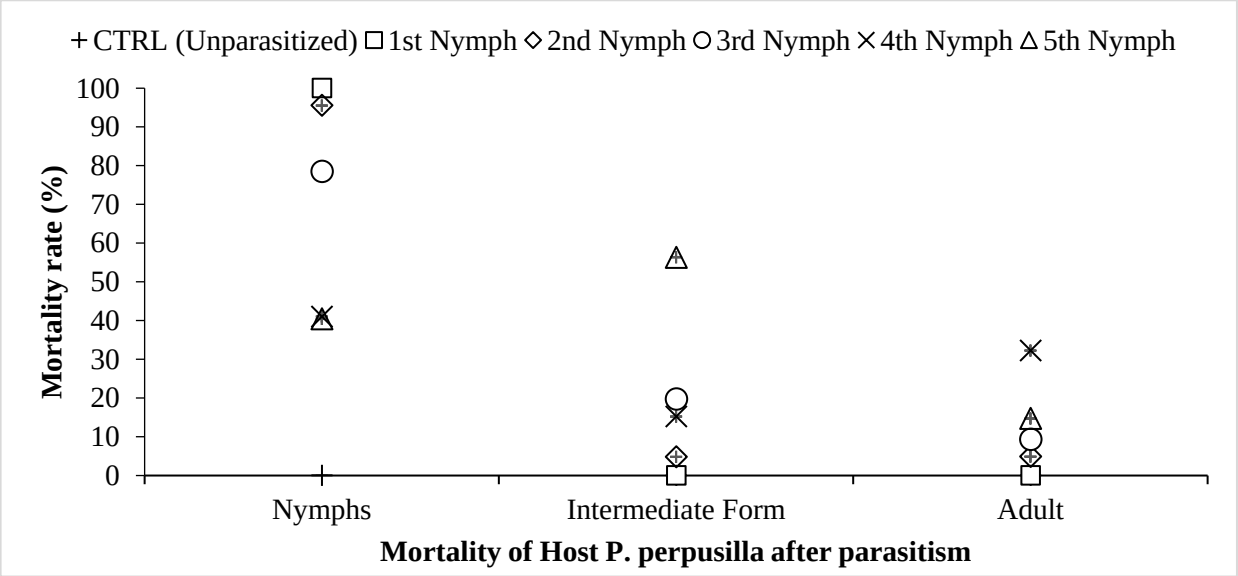

Fig. 1 Mortality of host *P. perpusilla* nymphs, intermediate forms (nymph-adult or adult nymph) or weak adults on parasitism

Nymphal Longevity

There were significant differences between nymphs that were parasitized in the third, fourth, and fifth stages of development, with the fourth parasitized nymphs having the highest duration (+ 7.59 days). Compared to unparasitized nymphs, nymphs parasitized in the second stage had a significantly shorter development time (8.15 days) than nymphs that were not parasitized.

Table 4.1Effect of parasitism by *E. melanoleuca* on the longevity of *P. perpusilla* nymphs

Parasitism Host (nymphs) stage	**Longevity of nymphs in days (mean ± SE)**	**Increase/ decrease (+)/ (−) days over control**
CTRL (Unparasitized)	27.21 ± 0.12	--
1st Nymph	**	**
2nd Nymph	19.06 ± 01.6	-8.15
3rd Nymph	32.38 ± 0.24	+5.17
4th Nymph	34.80 ± 0.20	+7.59
5th Nymph	32.99 ± 0.04	+5.78

Survival as normal adults

No nymphs parasitized at the first, second, third, or fourth stages were able to mature into normal adult nymphs. As nymphs, nymph-adult intermediates, or adults, they all died as frail creatures. Adults of the fifth parasitized nymphs exhibited a normal morphology in 19.91 percent of the cases. In contrast to unparasitized nymphs, parasitized nymphs had a zero percent survival rate which was statistically significant (Fig. 2).

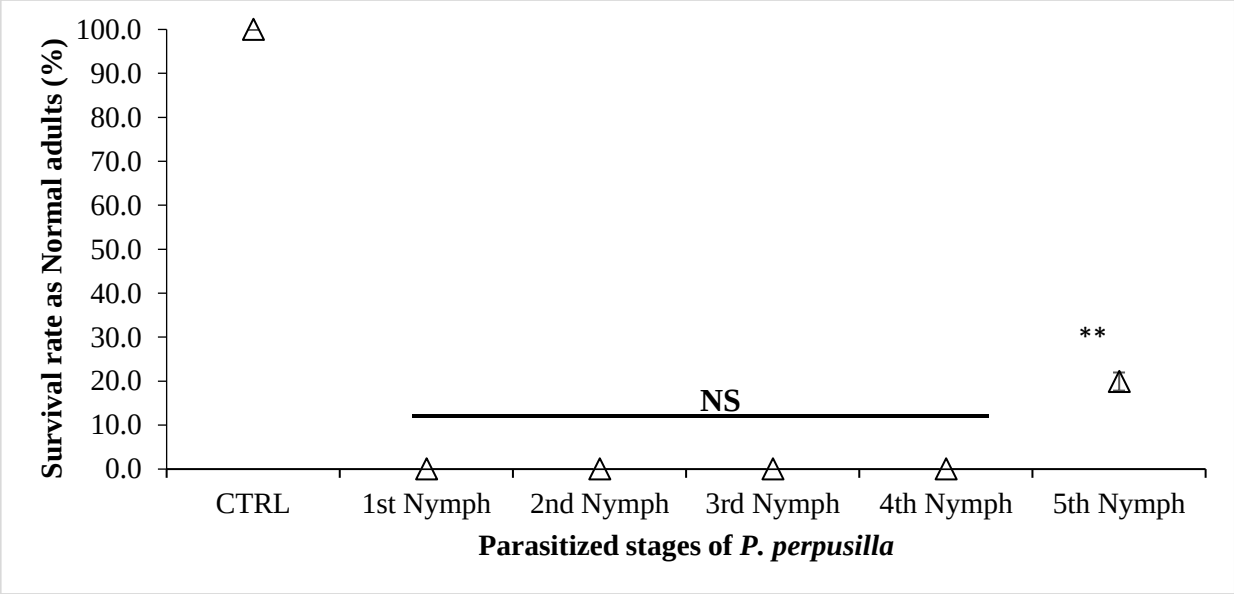

Fig. 2 Survival of host *P. perpusilla* as normal adults on parasitism at different stages

Longevity of survived Nymphs (as adults)

Regardless of the host stage, females outlived males by a significant margin (Fig. 3). Parasite-infested adult females from the 5th nymphal stage had a considerably lower lifetime than their uninfected counterparts. When comparing the parasitized and unparasitized 5th nymphal stages, there was no statistically significant difference in the length of life expectancy of adult males.

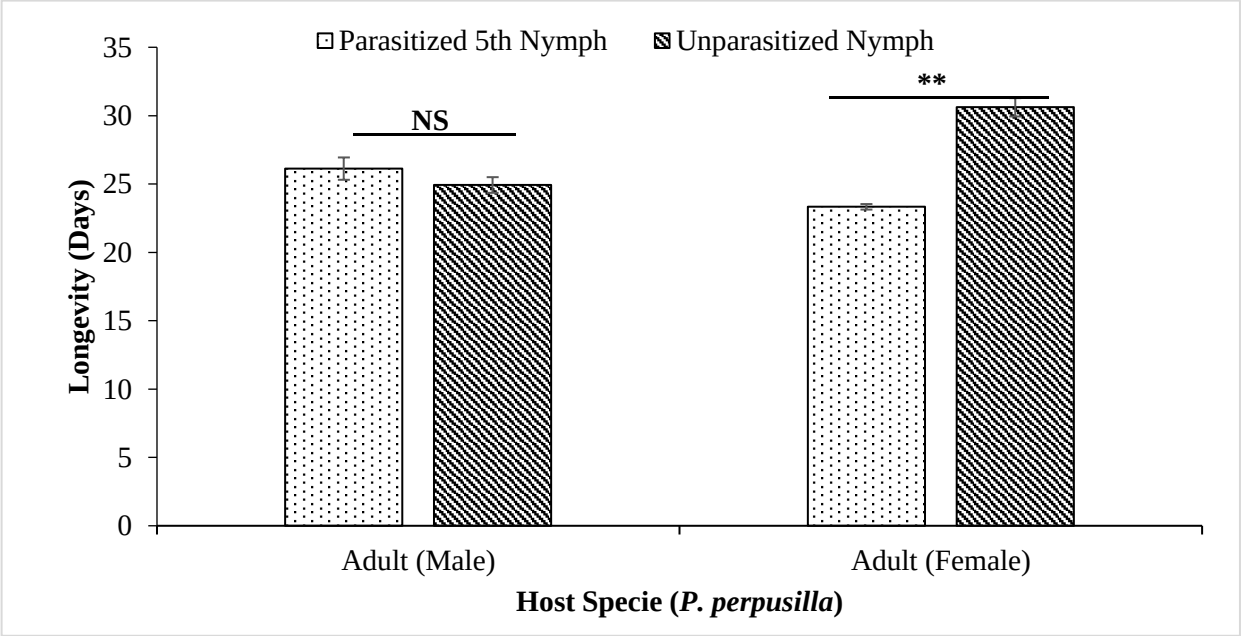

Fig. 3 Longevity of surviving adults (male and female) of *E. melanuleuca* parasitism effects on *P. perpusilla* adults

Adult Mortality

Male and female pyrilla adults were offered the parasitoid, and significant mortality disparities were observed in both male and female pyrilla when the parasitoid was offered to male and female pyrilla adults. Because of parasitic infection, adult male and females perished at a rate of 94.01 percent and 85.43 percent, respectively, in the study. Adults that were not parasitized died at a rate of zero (Fig. 4) in both sexes, according to the findings.

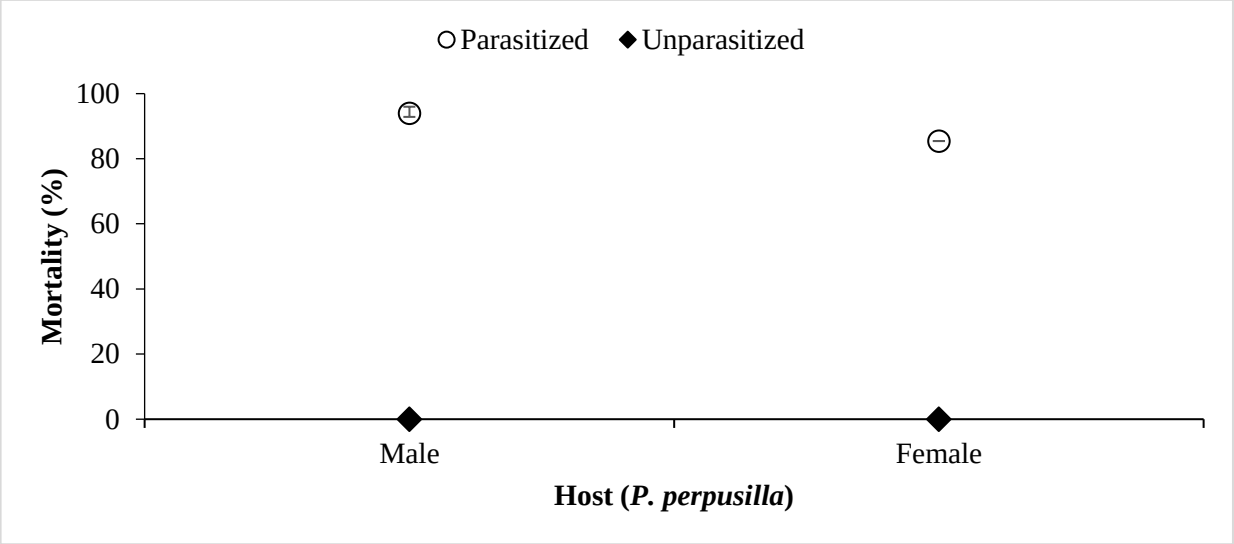

Fig. 4 Mortality of host *P. perpusilla* when male and female adults parasitized with *E. melanuleuca*

Adult longevity

Regardless of the host sex, the parasitized host females lived significantly longer than the parasitized host males. Male and female surviving host adults lived significantly longer lives after contracting the parasitism than after contracting the parasitism. The average lifespan of parasite-infected male was 6.38 days longer than that of their non-infected counterparts. In a similar vein, parasitized females outlived their unparasitized counterparts by 4.83 days on average, according to the study (Table 2).

Table 2 Effect of parasitism by *E. melanoleuca* on the longevity of *P. perpusilla* adults

Host stage	Longevity (Mean ± SE)		Increase (+) in days
	Parasitized	Un-parasitized	
Adult male	29.60 ± 1.11	23.22 ± 0.66	+6.38
Adult female	33.31 ± 0.58	28.47 ± 0.69	+4.83

Experiment 2

To test the efficacy of Botanical extracts and standard insecticide against *Pyrilla perpusilla* (Nymphs 4th instars) under Laboratory conditions

The figure 5 Shows significant difference in the efficacy of Botanical extracts and standard insecticide against *Pyrilla perpusilla* (Nymphs 4th instars) after 8, 16 and 24hrs under laboratory conditions. After 8hrs, acetamiprid had the highest percentage of population reduction (40.93 ± 0.65%), followed by Sophora, neem, and tobacco extracts, which had percentages of population reduction of 33.59 ± 0.87%, 32.09 ± 0.74%, and 27.81 ± 1.22%, respectively, (Fig. 5). Acetamiprid was the most effective pesticide in terms of population reduction (40.93 ± 0.65%). According to the findings, acetamiprid has the highest percentage of population reduction (40.93 ± 0.65%). Control treatment in the study resulted in the lowest population reduction ever recorded (0.00) percent.

All the plant extracts and acetamiprid were found to have a significant effect on the population of *P. perpusilla* after 16 hours of treatment. Acetamiprid was associated with the highest mortality rate (74.74%), followed by Sophora, neem, and tobacco extracts, which were associated with mortality rates of 54.83, 53.45, and 48.12%, respectively. The control treatment had the lowest population reduction (0.00) (Fig. 5).

Furthermore, after 24 hours, all of the plant extracts and the acetamiprid had great impact on the mean number of *P. perpusilla* in the area. Most organisms who took acetamiprid died, but Sophora, neem, and tobacco extracts had death rates of 74.79, 70.79 and 66.79%, respectively. In the study, the control treatment had the minimum population decline (0.00%) (Fig. 5).

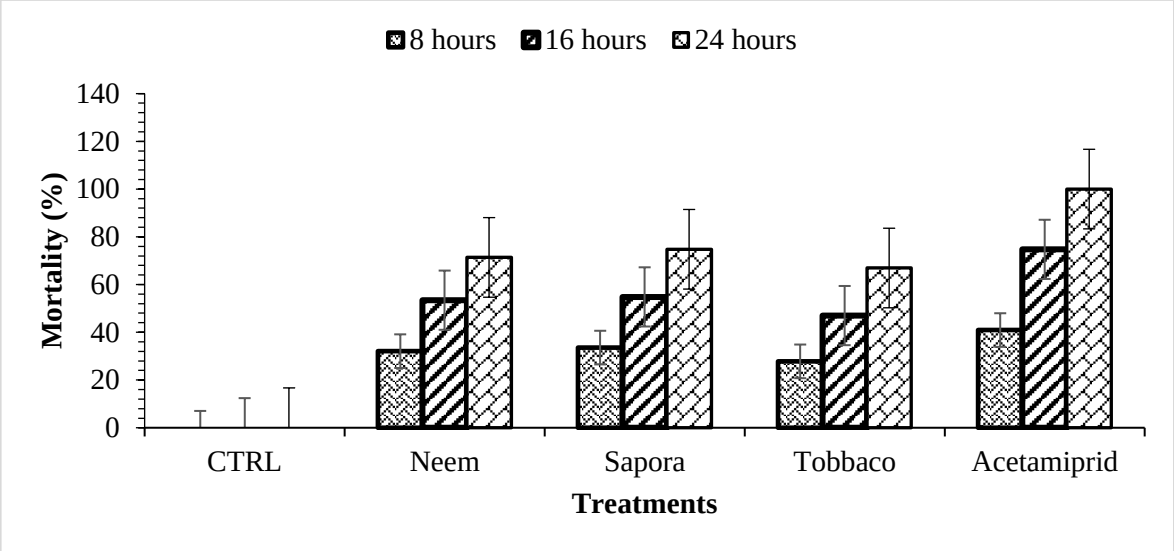

Fig. 5 Efficacy of Botanical extracts and standard insecticide against *Pyrilla perpusilla* (Nymphs 4th instars) under Laboratory conditions Sophora

Experiment 3

To test the efficacy of Botanical extracts and standard insecticide against *Pyrilla perpusilla* (Nymphs 4th instars) under field conditions

The Figure 6 shows significant difference the efficacy of Botanical extracts and standard insecticide against *Pyrilla perpusilla* (Nymphs 4th instars) under field conditions after 3, 7 and 14 (DAS). Acetamiprid had the highest percentage of population reduction (68.74 ± 0.56%), followed by Sophora, neem, and tobacco extracts, which had percentages of population reduction of 62.07 ± 0.62%, 57.37 ± 0.32%, and 50.24 ± 1.49%, respectively, (Fig. 6). Acetamiprid was the most effective pesticide in terms of population reduction (68.74 ± 0.56%). Control treatment in the study resulted in the lowest population reduction ever recorded (8.48 ± 0.39) percent.

All the plant extracts and acetamiprid were found to have a significant effect on the population of *P. perpusilla* after 7 DAS. Acetamiprid was associated with the highest mortality rate (66.83 ± 0.58%), followed by Sophora, neem, and tobacco extracts, which were associated with mortality rates of 59.79, 55.73, and 49.58%, respectively. In the study, the control treatment had the smallest population reduction (8.13 ± 0.30%), which was considered the smallest population decline (Fig. 6).

They found that after 15 DAS, all the plant extracts and the acetamiprid had a big impact on the number of *P. perpusilla* in the area. Insects who took acetamiprid died at the rate of (62.47 ± 0.53%), but saphora, neem, and tobacco extracts had death rates of 57.19, 54.80 and 48.98%, respectively. In the study, the control treatment had the minimum population decline (7.85 ± 0.11%).

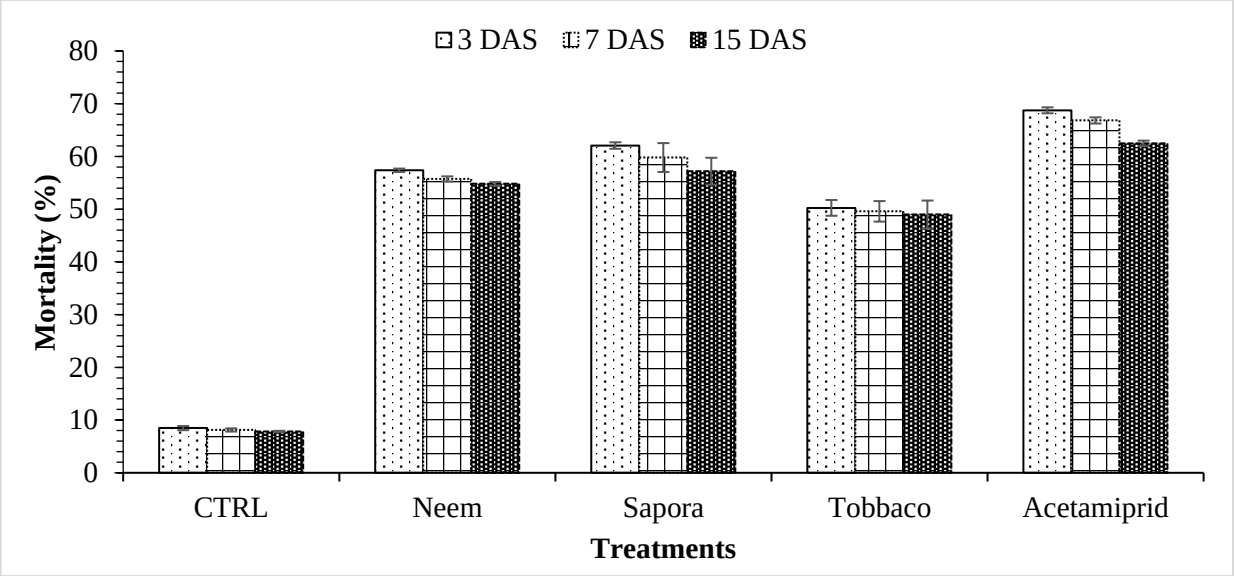

Fig. 6 Mortality of *P. perpusilla* with application of synthetic and natural pesticides under field conditions

Experiment 4

Efficacy of cultural, chemical, botanical extracts, biological control, and their different combinations on the population of *Pyrilla perpusilla* at ARI, Harichand

The data present in table (3) shows significant difference between the efficacies of different applied treatments @ P-value =≤ 0.05. The overall means data shows that the lowest mean population (0.46 *P. perpusilla* leaf^{-1}) was recorded from T_8 that follows T_9, T_3, T_7, T_4, T_5, T_2 and T_1 with mean population (0.66, 1.24, 1.39, 1.555, 1.93, 2.13 and 3.06 *P. perpusilla* leaf^{-1}) respectively, while the highest mean population (4.24 *P. perpusilla* leaf^{-1}) were recorded from T_{10}.

Similar trend regarding the efficacy of different treatments against population *P. perpusilla* were also recorded on 16-July where the minimum mean population (0.70 *P. perpusilla* leaf^{-1}) were recorded from T_8, that followed T_9, T_6, T_3, T_5, T_7, T_4, T_2, and T_1 (0.85, 1.28, 1.50, 1.75, 175, 1.95, 2.10 and 2.85 *P. perpusilla* leaf^{-1}), respectively. While the maximum mean population (3.34 *P. perpusilla* leaf^{-1}), were recorded from T_{10}.

The mean significant difference among treatments were also observed on 31-July. The highest mean (3.75 *P. perpusilla* leaf^{-1}) were noted from t10, followed by T_1 (3.33 *P. perpusilla* leaf^{-1}), T_2 (2.50 *P. perpusilla* leaf^{-1}), T_5 (1.70 *P. perpusilla* leaf^{-1}), T_4 (1.62 *P. perpusilla* leaf^{-1}), T_7 (1.49 *P. perpusilla* leaf^{-1}), T_3 (1.35 *P. perpusilla* leaf^{-1}), T_6 (1.20 *P. perpusilla* leaf^{-1}) and T_9 (0.85 *P. perpusilla* leaf^{-1}), whereas, the minimum mean (053 *P. perpusilla* leaf^{-1}) were noted from T_8.

Moreover, the data recorded on 16-August shows significant difference. The lowest (0.43 *P. perpusilla* leaf^{-1}) were observed from T_8, that followed T_9, T_6, T_3, T_7, T_4, T_5, T_2 and T_1 (0.65, 1.05, 1.20, 1.27, 1.42, 1.87, 2.80, 3.30 *P. perpusilla* leaf^{-1}), respectively**.** While the highest mean (4.75 *P. perpusilla* leaf^{-1}) were observed from T_{10}.

The data recorded on 31-August present in the table showed that the maximum (5.08 *P. perpusilla* leaf^{-1}) were recorded from T_{10} followed T_1 (2.53 *P. perpusilla* leaf^{-1}), T_2 (2.24 *P. perpusilla* leaf^{-1}), T_5 (1.48 *P. perpusilla* leaf^{-1}), T_4 (1.22 *P. perpusilla* leaf^{-1}), T_7 (1.05 *P. perpusilla* leaf^{-1}), T_3 (1.05 *P. perpusilla* leaf^{-1}), T_6 (0.91 *P. perpusilla* leaf^{-1}) and T_9 (0.35 *P. perpusilla* leaf^{-1}). The minimum (0.18 *P. perpusilla* leaf^{-1}) were recorded from T_8.

Table 3 Efficacy of cultural, chemical, botanical extracts, biological control, and their different combinations on the population of *Pyrilla perpusilla* at ARI, Harichand

Treatments	16-July	31-July	16-August	31-August	Mean
T_1	2.85 b	3.33 b	3.30 b	2.53 b	3.06 b
T_2	2.10 c	2.50 c	2.80 c	2.24 c	2.13 c
T_3	1.50 f	1.35 f	1.20 f	0.92 f	1.24 g
T_4	1.95 d	1.62 de	1.42 e	1.22 e	1.55 e
T_5	1.75 e	1.70 d	1.87 d	1.48 d	1.93 d
T_6	1.28 g	1.20 g	1.05 g	0.91 f	1.11 g
T_7	1.75 e	1.49 e	1.27 f	1.05 f	1.39 f
T_8	0.70 i	0.53 i	0.43 i	0.18 h	0.46 i
T_9	0.85 h	0.85 h	0.65 h	0.35 g	0.66 h
T_{10}	3.34 a	3.75 a	4.75 a	5.08 a	4.24 a
LSD (0.05)	0.0902	0.1367	0.1076	0.1518	0.1350

Mean followed by different alphabetic letters are statistically significant @ P-value= ≥0.05

Effect of different treatments on sugarcane yield (t ha^{-1})

The results present in figure 7 shows significant difference on mean yield of sugarcane (t ha^{-1}). The maximum yield 64.91 were obtain from plot treated with T_8 followed by T_9, T_7, T_6, T_3, T_5, T_4, T_2, and T_1, with 64.40, 64.20, 64.00, 63.80, 63.70, 63.44, 63.36 and 63.16 cane yield t ha^{-1}. While, the minimum yield 62.13 was obtain from control plot.

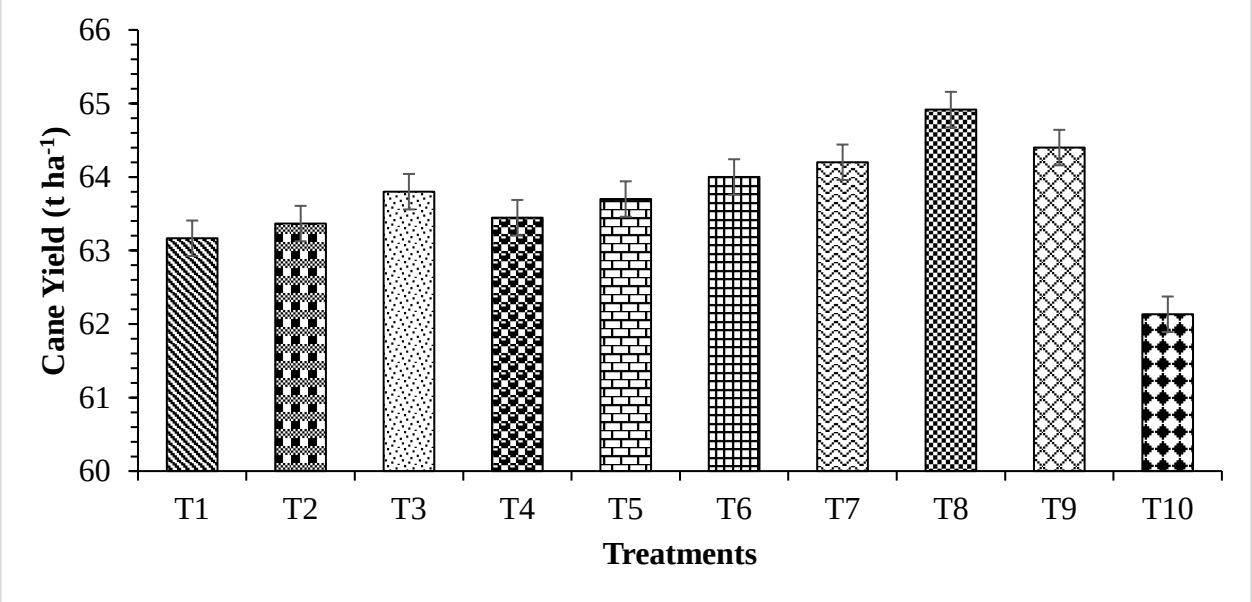

Figure 7: Effect of different treatments on sugarcane yield t ha^{-1}

Discussion

The studies were conducted for the safe management of sugarcane *Pyrilla perpusilla* by using different control strategies i.e., using of cultural, biological control, chemical, botanical extracts, and their different possible combinations. At first the biological control agent (*Epicarnia melanuleuca*), chemical and botanical extracts were tested un laboratory conditions for finding the influence on the *P. perpusilla* and after the confirmation of their effect on the pest these *Epicarnia melanuleuca*, chemical and botanical extracts were evaluated under field conditions.

In present research work the Biocontrol agent *Epicarnia melanuleuca* were tested against the various stages of *Pyrilla perpusilla* which resulted indicated that there was a statistically significant difference in nymphal mortality between nymphs of different ages that were fed to the parasitoid. The illustration against nymphs depicts the first nymphal stage where mortality was 100%. The mortality of

intermediate form depicts the fifth nymphal stage where mortality was 56.34 percent. After nymphal stage three it was 19.74%; after nymphal stage four, it was 15.23%; after stage two, it was 4.84%. The death of parasitized nymphs as weak adults was also observed after the nymphal stage was completed, suggesting they perished as weak adults after the nymphal period was over. It was estimated that 32.27% of nymphs died by the fourth stage, 14.72% by the fifth, 9.36% by the third, and only 4.88% by the second nymphal stage due to age-related mortality as a weak adult. Development of a biocontrol strategy that is both ecologically benign and successful, it is essential to understand the host-parasite relationship. I believe that this is a very important step in the process. Due to its short life cycle, high reproduction rate, and intense searching capacity, the parasitoid *E. melanoleuca* poses a considerable danger to the population of the nymphs and adults of Perpusilla in Indian field conditions (Seneviratne and Kumarasinghe 2002). To put it another way, it has the potential to be used in sugarcane leafhopper biocontrol in the field. Studies on *P. perpusilla* adults and advanced nymphal stages studied parasitism's influence on fitness features to discover whether parasitism affects fitness attributes at any stage of nymphal growth. Mortality is more common in nymphs when they are younger, but as the host ages, the probability of death lowers even further. One or more parasitoid-infected wounds experienced during exposure may have caused the first nymphal stage's death two days after exposure. It was shown that parasitism was linked to the early nymphal death of the host. During their investigation, (Sharma and Shera 2021) identified a high death rate associated with parasitoids. The early nymphs may have been less tolerant of parasitoid feeding than the later nymphs, which may have resulted in their demise as nymphs rather as adults.

For the first to fourth nymphal stages of the parasitoid, we found that all parasitized individuals died as weak nymphs, intermediates, or weak adults. This is critical for maximising the parasitoid's field-based augmentative release. Researchers Misra and Krishna (1986) found that parasites do not kill their hosts immediately, but rather over time. Infected nymphs could complete their development, but they were unable to mature into adults, as (Bal *et al*., 1990) had previously discovered.

In comparison to the general population, only 14% of parasitized fifth nymphal stage adults showed typical morphologies. This ectoparasitoid, *E. melanoleuca*, is unique because it permits both the host and the parasite to grow. Due to its maturity, it is probable that the final stage of the larval stage was able to resist parasitoid feeding and moult like an adult. There was also a finding that older hosts' immune systems were more effective (Strand and Pech, 1995). Another option is for the parasitoid to detach and pupate once it has gotten enough nutrients during the fifth nymphal stage of development. A small percentage of nymphs parasitized during the fifth stage of development can survive and mature into normal adults because of this. *P. perpusilla*'s fifth nymphal stage of *F. melanoleuca* was shorter than the penultimate stage of the fourth nymphal stage of the species, according to another study's findings (Sharma and Shera 2021). Some parasitized nymphs in the advanced stages of development did not reveal parasitism, as discovered by (Bal *et al*., 1990), demonstrating that a parasitoid may continue developing on following adults until symptoms manifest as well. According to Bindra and Brar's (1978) findings, parasitized penultimate nymphs may still mature into healthy adults.

This study confirmed prior findings that parasitized nymphs in the third to fifth stages (as opposed to un-parasitized nymphs) have a longer development period and a shorter lifespan. When a parasite initially attacks, the length of time it takes to kill the host depends on several factors, including the host's age and size, as well as the host's signal to the juvenile parasitoid that it has enough resources to finish growing (Vuts *et al*., 2012). In the interim, until the parasitoid can sustain itself, the host must be kept alive at all costs. There are two probable reasons for the decreased feeding rate of parasitized nymphs: the parasitoid-induced increase in longevity and the parasitism-induced weakening of the parasitized nymphs' immune systems. Koinobiont larvae can decrease or lengthen their development time greatly depending on the quality of the host they're parasitizing. Infected penultimate nymphs took longer to progress into adults, according to studies by Bindra and Brar (1978).

Those females who became adults after being parasitized as fifth nymphs survived for a shorter duration than those who became adults after being uninfected as nymph phases. When it comes to

adult fertility, parasitized and non-parasitic populations differed statistically in this study (Bal *et al.*, 1990). First through fourth nymphal stages were found to be completely dead after being parasitized; no survivors grew into normal adults. Both sexes of *P. perpusilla* adults died when the parasitoid was offered to the adults, compared to when the parasitoid was not offered to the adults.

Even though parasitized pyrilla adults may have lower eating rates than normal throughout the course of their long lives, this is most likely due to the parasites' weakened state. Solitary parasitoids can significantly reduce their host's consumption of food in comparison to healthy hosts when it comes to biological control (Rohlfs and Mack, 1983). When compared to parasite-free females and men, parasitized females had lower fecundity, and parasitized males had lower fecundity as well (25%). Parasitic infections do not appear to shorten the lives of those sick, according to Brar and Bains (1979). Furthermore, the botanical extrcats and chemical (acetamiprid) were applied against the *Pyrilla perpusilla* Nymphs (4^{th} instar) under laboratory conditons and field conditions which results shows that acaetamprid showed significantly more effectiveness in the mortality of *P. perpusilla* (Nymphs) followed by Saphora, neem and tobacco extracts. These results are in line with Chand *et al*. (2019) who reported the effectiveness of acetamiprid againd *P. perpusilla*. The efficacy of the Saphora extarcts was reported against several insects. The effectiveness of the Neem extracts was reported by Anwar *et al*. (1992) who tested various botanical extacts against *P. perpusilla* under laboratory conditions and find the Neem extacts more effective in the mortality of this pest. These findings support the notion that saphora extract has the potential to be employed as a botanical insecticide against *P. perpusilla* in the future.

There are several advantages to using botanical pesticides over synthetic pesticides, such as minimal mammalian toxicity, the absence of the possibility of building pest resistance, and the fact that botanical extracts are more easily available and less expensive than synthetic pesticides (Isman 2013). The efficacy of the plant leaf extracts such as Vitex, Pongamia, and Calotropis was shown to be superior (Anand Prakash *et al*., 2008). When 5% Vitex leaf extracts were sprayed, (Rajappan *et al*., 2000) found a decrease in hopper population. Mariappan *et al*. (1988) found that pongamia leaf extract inhibited the survival of green leafhoppers and the emergence of planthoppers. Pupal deformity of rice insect pests was discovered by Sukumaran *et al*. (1987) after exposure to Vitex leaf extract. Vitex leaf extract at 5% concentration was found to be effective against hoppers and leaf folders, according to Mahapatra *et al*. (2009).

The active ingredients in neem plants are Saphora, azadirachtin, salanin, meliantriol, and neem. The azadirachtin chemical is supposed to destroy insects. The leaves and seeds of neem contain phenols, quinones, alkaloids, acids, and terpenes. The terpene chemicals neem (nimbinen), thionemon, meliantriol, azadirachtin, and salanin are believed to be bioactive components in vegetable insecticides (Rukmana and Yuyun, 2002). Nimbilin, melanitrol, salanin, and neem are secondary metabolites from neem plants. Azadirachtin is a contact, gastrointestinal, and pesticide toxin. It can be utilised to manage pests like Helopelthis sp. (long caterpillar), Aphis sp. (spyder caterpillar), Nilarvata sp. and Sitophilus sp. (Rukmana and Yuyun, 2002). Insect pests near the neem tree can be killed by azadirachtin chemicals that diminish hunger, egg formation and hatching, increase mortality, and activate infertility (act as antifertile). The neem seed's azadirachtin inhibits the action of ecdyson hormone, which is involved in insect metamorphosis (Prakash and Rao, 1997). Salanin reduces hunger, which reduces insect destructive capacity, even when the insects are not dead. Insect pests exposed to neem seed powder will spread out and their destructive power will be considerably diminished (Indiati and Marwoto, 2008). Meliantriol acts as a deterrent to pest insects, preventing them from approaching plants. The use of neem seed extract can result in 60% mortality of red mite *Tetranychus urticae*, compared to dicofol which reaches 80% (Singh *et al*., 2005). The neem leaves contain a toxin that interferes with digestion by suppressing intestinal contractions (Nurtiati and Widya, 2001). Botanical leaf extracts can be absorbed through the body walls, acting as a contact poison (Ardiyansyah *et al*., 2002). Nathan *et al*. (2007) reported that the use of 0.5 ppm neem extract ensures significantly high % nymphal mortality of plant hopper.

Moreover, the effectiveness of cultural, biological agent (*Epicarnia melanuleuca*), acetamiprid and plant extracts (Saphora) and different combinitions were used to inhibit the population of *Pyrilla*

perpusilla varied greatly and was found to be statistically significant. The combination of cultural, biological, chemical was found the most effective in reducing the *P. perpusilla* population followed by the combination of cultural, biological and saphora extracts. Although the difference between were significant from each other but the overall mean difference was not far away from each other. These results are at par with Rasul *et al.* (2014) who concluded that interation of of cultural, biological, and chemical is the most effective to keep the pest population below the economic threshold level. The results regarding the yield obtained from different treated plots indicated significant difference. The maximum yield t ha^{-1} was obtained from plot treated with the combinations of cultural, biological, and chemical treated plot followed by the plots treated with cultural, biological and botanical extracts. The lowest yield was obtained from the control plots. It is obvious from the data that the combinations of different treatments for managing the pest is more helpful for high yield production. Similar study was conducted by Rasul *et al*. (2014) who used different combination of cultural, biological, and chemical and recorded maximum yield compared other treatments. These results are agreed with those finding of Rana *et al*. (2002), who managed the infestation of *P. perpusilla* with the application of biocontrol agent, combined with pesticide applications and reported the best management of this pest. The findings of the present results are agreed with those of Anwar *et al.* (1992) and Khan *et al*., (2017) who reported the best control of different sugarcane insect pest while using different integrated management practices with enhanced in yield capabilities.

Conclusion

It was concluded from the study that mortality rate of nymphs was significantly greater in younger nymphs and dropped significantly in older nymphal stages. Except for the fifth nymphal stage, all parasitized *P. perpusilla* nymphs died as nymphs, nymph-adult intermediates, or weak adults. However, it took a longer time to develop into an adult than those who were not parasitized. Additionally, the parasitoid resulted in a considerable increase in the death rate of pyrilla adults. In both sexes, parasitized adults lived longer than unparasitized individuals. Because the parasitoid *F. melanoleuca* had a severe effect on host fitness, it was an important biocontrol agent in decreasing the *Pyrilla* population, as most parasitized individuals died before contributing to the following generation. These findings regarding the host-parasitoid relationship may aid in developing an augmentative biological control programme against *P. perpusilla*. The combination of cultural, chemical, and biological are a more effective in reducing the pest population. Along with this the combination of cultural, biological, and botanical extracts were also find viable alternative to synthetic insecticides since they are inexpensive, readily available, and reasonably safe for natural enemies and other non-target species. As a result, by sacrificing little yield, it is recommended to utilise integration of indigenous cultural, biological agent and botanical insecticides to manage *P. perpusilla* in sugarcane yield in a sustainable manner.

Acknowledgement

All the authors highly acknowledge department of Plant and environmental Protection, National Agricultural Research Centre, Islamabad for providing all the research facilities during the experimental period.

Novelty statement

The novelty of the study is the safe management of *Pyrilla perpusilla* in sugarcane crop. This study assessed the effectiveness of different control tools, including cultural practices, bioagents, botanical extracts, and synthetic chemicals, against *P. perpusilla* in laboratory and field experiments. These findings will provide valuable insights for future IPM Programs against *P. perpusilla* infestation in sugarcane fields.

Conflict of interest

The authors declare no conflict of interest.

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