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Screening of biopesticides and their combinations against sucking insect pest complex in *Bt* cotton

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ABSTRACT

Sucking pests like whitefly, aphid, thrips, and leafhopper pose a serious threat to cotton crop, affecting yield and quality. Implementing integrated pest management strategies, which involve the use of biopesticides is essential for successful control. The present experiment was conducted to evaluate the efficacy of different biopesticides and their combinations against sucking insect pests in Bt cotton at Khalsa College, Amritsar, Punjab during kharif 2022. Considering the Economic Threshold Level (ETL) of pests, four sprays were given at interval of 10 days. The data obtained revealed that a combination of neem oil and Verticillium lecanii showed a maximum reduction in population of sucking pests viz. whitefly (64.88%), aphid (70.59%), thrips (54.23%) and leafhopper (62.73%) at seven days after the end of last spray. The present study highlights the significance of microbial biopesticides in promoting eco-friendly and sustainable pest management.

Keywords: *Bt cotton, biopesticides, neem oil, sucking pests, Verticillium lecanii*

INTRODUCTION

Bt cotton is a genetically modified type of cotton achieved by introducing genes from soil dwelling bacterium, *Bacillus thuringiensis* (Berliner) through genetic engineering [1]. These genes encode for the production of crystal insecticidal proteins which are toxic to lepidopteran pests of cotton, particularly bollworms, and therefore reducing the need of chemical insecticides [2]. In India, *Bt* cotton was introduced in 2002 and following its success, the area and production of cotton increased significantly with an expanse of 11.7 million hectares under cultivation [3]. In Punjab, *Bt* cotton was released for cultivation in 2005 [4]. The rate of adoption of *Bt* cotton has reached 95 percent with 2.49 lakh hectares of area under cultivation and total production of 6.39 lakh bales in 2022 [5].

Despite being effective against bollworms, *Bt* cotton has attracted a wide range of other insect pests, especially sucking pests due to the reduction in usage of insecticides at early stages [6]. Leafhopper, *Amrasca biguttula bigutulla* (Ishida), aphid, *Aphis gossypii* (Glover), whitefly, *Bemisia tabaci* (Gennadius), thrips, *Thrips tabaci* (Lindeman), red cotton bug, *Dysdercus cingulatus* (Fabricius), mealy bug, *Phenacoccus solenopsis* (Tinsley) and dusky cotton bug, *Oxycarenus laetus* (Kirby) are common sucking pests of cotton causing damage from seedling emergence to harvest with significant decline in yield by sucking sap from plants and making them weaker. Around 28.13 percent of avoidable yield losses are caused by major sucking pests in cotton [7]. More than 90 percent of area is under *Bt* cotton which is susceptible to sucking pests, which cause considerable damage and need to be controlled [8].

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Until now, chemical control is the most popularized method used by farmers to control the sucking insect pests in cotton. The highest share of pesticide consumption, nearly 40-56 per cent of total pesticides is received by cotton in our country [9]. However, the repeated use of these synthetic insecticides give rise to serious hazards to the environment and human health [10]. Therefore, different alternative methods such as biopesticides and biorationalsare being introduced to provide economical and eco-friendly management of insect pests, lowering the problems associated with insecticides [11]. Among botanicals, neem oil is widely used due to its diverse mode of action against a wide range of insects. Entomopathogenic fungi *viz*. *Beauveria bassiana* (Balsamo), *Metarhizium anisopliae* (Metchnikoff) and *Verticillium lecanii* (Zimmerman) and bacteria are also gaining importance due to their target specificity and safety to the environment [12]. Combination of botanical having insecticidal action and entomopathogen is a unique technique to combat insect pest resistance and recurrence, resulting in effective management comparable to synthetic insecticides [13]. Therefore, an attempt was made to assess the compatibility of various entomopathogens and neem oil alone against sucking insect pests of cotton and their combinations with neem oil to investigate potential synergies.

MATERIALS AND METHODS

To evaluate the efficacy of biopesticides against sucking insect pests, the present investigation was carried out during kharif 2022 at Khalsa College, Amritsar in Randomized Block Design (RBD). The total experimental area of 325 square meters (26×12.5m) was divided into three blocks having 1-meter space between them and each block served as a separate replication. Further, each block was divided into six plots with individual plot sizes of 3.5 × 3.5 m. *Bt* cotton hybrid US 71 (BG II) was grown in the field as per the Package of Practices by Punjab Agricultural University, Ludhiana. Six treatments including neem oil, *Beauveria bassiana* (Balsamo), *Verticillium lecanii* (Zimmerman), neem oil + *B. bassiana*, neem oil + *V. lecanii* and untreated control were evaluated (Table 1).

Application of all the treatments within each replication was done in random manner. All the treatments were replicated thrice. First spray of biopesticides was done when the pest population reached its Economic Threshold Level (ETL) and subsequent sprays were given at interval of 10 days. The observations recorded were subjected to statistical analysis.

Table 1. Detail of treatments

Treatment	Biopesticide
T_1	Neem oil (5 ml/l)
T_2	*Beauveria bassiana* (10 g/l)
T_3	*Verticillium lecanii* (10 ml/l)
T_4	Neem oil (5 ml/l) + *Beauveria bassiana* (10 g/l)
T_5	Neem oil (5 ml/l) + *Verticillium lecanii* (10 g/l)
Control	

Considering the ETL of pests during the experimental period four sprays were given at an interval of 10 days. The observations were recorded from five randomly selected plants from each plot. Three leaves per section (upper, middle and bottom) of each selected plant were observed for a population of nymphs and adults of each major sucking pests. Pre-treatment observations were recorded one day before spraying of any treatment and post-treatment observations were recorded 1,3, 5 and 7 days after each spray. The mean count of sucking pests per 3 leaves was recorded in the data.

RESULTS

Whitefly

Results revealed that there was no significant difference of the whitefly population among the treatments before spraying. Maximum percent reduction in whitefly population over control at seven days after spray was recorded in the combination of neem oil and *V. lecanii* (48.42%) followed by the combination of neem oil and *Beauveria bassiana* (37.93%), *V. lecanii* (29.77%), neem oil (24.29%) and *B. bassiana* (18.54%). Similar results were found after second spray also. Combination of neem oil and *V. lecanii*recorded highest percent reduction of whitefly population (53.25%) whereas, lowest per cent reduction was observed in *B. bassiana* (27.75%). The same trend of efficacy of biopesticides against whitefly was observed during third spray. A combination of neem oil and *V. lecanii* was found superior at seven days after spraying with a maximum per cent reduction in whitefly population over control (59.97%). After fourth spray, the results revealed that treatments were found effective in a similar trend at seven days after spray also. A combination of neem oil and *V. lecanii* was found to be most effective with highest per cent reduction in whitefly population over control (64.88%). The next best treatment was the combination of neem oil and *B. bassiana* (55.17%) followed by *V. lecanii* (48.12%), neem oil (42.47%) and *B. bassiana* (34.64%) (Table 2). However, whitefly population was found to be highest before spraying and start decreasing with application of treatments and was found to be lowest at the end of fourth spray. Combination of neem oil and *V. lecanii* maximum reduction from 15.41 to 13.38 whiteflies per three leaves (Fig 1).

Table 2. Efficacy of biopesticides against whitefly population on Bt cotton

Sr. No.	Treatment	Dose	Per cent reduction in whitefly population over control at 7 DAS			
			1st Spray	2nd Spray	3rd Spray	4th Spray
T_1	Neem oil	5 ml/l	24.29	34.46	42.39	42.47
T_2	*Beauveria bassiana*	10 g/l	18.54	27.75	36.12	34.64
T_3	*Verticillium lecanii*	10 ml/l	29.77	41.84	45.50	48.12
T_4	Neem oil + *Beauveria bassiana*	5 ml/l + 10 g/l	37.93	45.53	52.44	55.17
T_5	Neem oil + *Verticillium lecanii*	5 ml/l + 10 ml/l	48.42	53.25	59.97	64.88

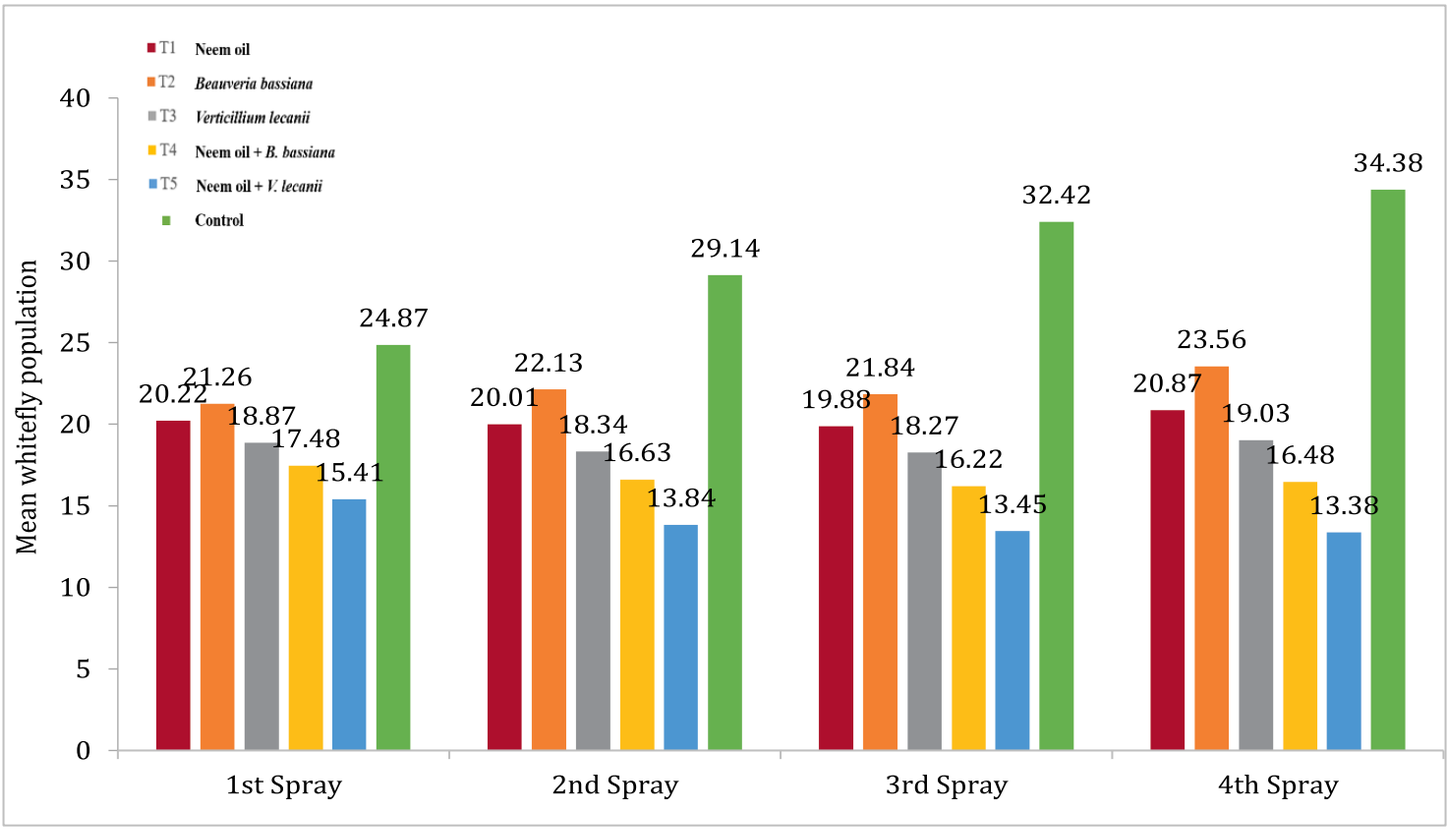

Fig 1. Effect of biopesticides based on mean population of whitefly

Aphid

There was no significant difference in aphid population among the treatments before spraying. Significant decline in population was observed at seven days after spray. The data on per cent reduction of aphid population over control at seven days after spray revealed that maximum reduction of aphid population was observed in the combination of neem oil and *V. lecanii* (42.11%) followed by a combination of neem oil and *B. bassiana* (35.21%), *V. lecanii* (33.14%), neem oil (29.90%) and *B. bassiana* (24.27%). During the second spray, the data inferred that again a maximum reduction in aphid population over control at seven days after spray was recorded in a combination of neem oil and *V. lecanii* (58.97%) while, *B. bassiana* showed minimum reduction (38.33%) among the treatments. Similarly, after third spray the results showed that all the treatments were found superior over control in controlling the aphid population.

The data recorded at seven days after spray also showed a combination of neem oil and *V. lecanii* as best among the other biopesticides with highest per cent reduction of aphid population (67.32%) over control while, the treatment *B. bassiana* was found as least effective with lowest reduction in the population of aphid (46.43%). After fourth spray, efficacy of biopesticides was observed in same trend which showed the maximum per cent reduction in population of aphid over control at seven days after spray in combination of neem oil and *V. lecanii* (70.59%) followed by a combination of neem oil and *B. bassiana* (67.72%), *V. lecanii* (63.13%), neem oil (58.29%) and *B. bassiana* (52.72%) (Table 3). Lowest population of whitefly population from 13.51 to 8.04 aphids per three leaves was recorded in plots treated with combination of neem oil and *V. lecanii* after fourth spray (Fig 2).

Table 3. Efficacy of biopesticides against aphid population on Bt cotton

Sr. No.	Treatment	Dose	Per cent reduction in aphid population over control at 7 DAS			
			1st Spray	2nd Spray	3rd Spray	4th Spray
T_1	Neem oil	5 ml/l	29.90	45.85	54.26	58.29
T_2	*Beauveria bassiana*	10 g/l	24.27	38.33	46.43	52.72
T_3	*Verticillium lecanii*	10 ml/l	33.14	48.51	62.57	63.13
T_4	Neem oil + *Beauveria bassiana*	5 ml/l + 10 g/l	35.21	54.14	64.55	67.72
T_5	Neem oil + *Verticillium lecanii*	5 ml/l + 10 ml/l	42.11	58.97	67.32	70.59

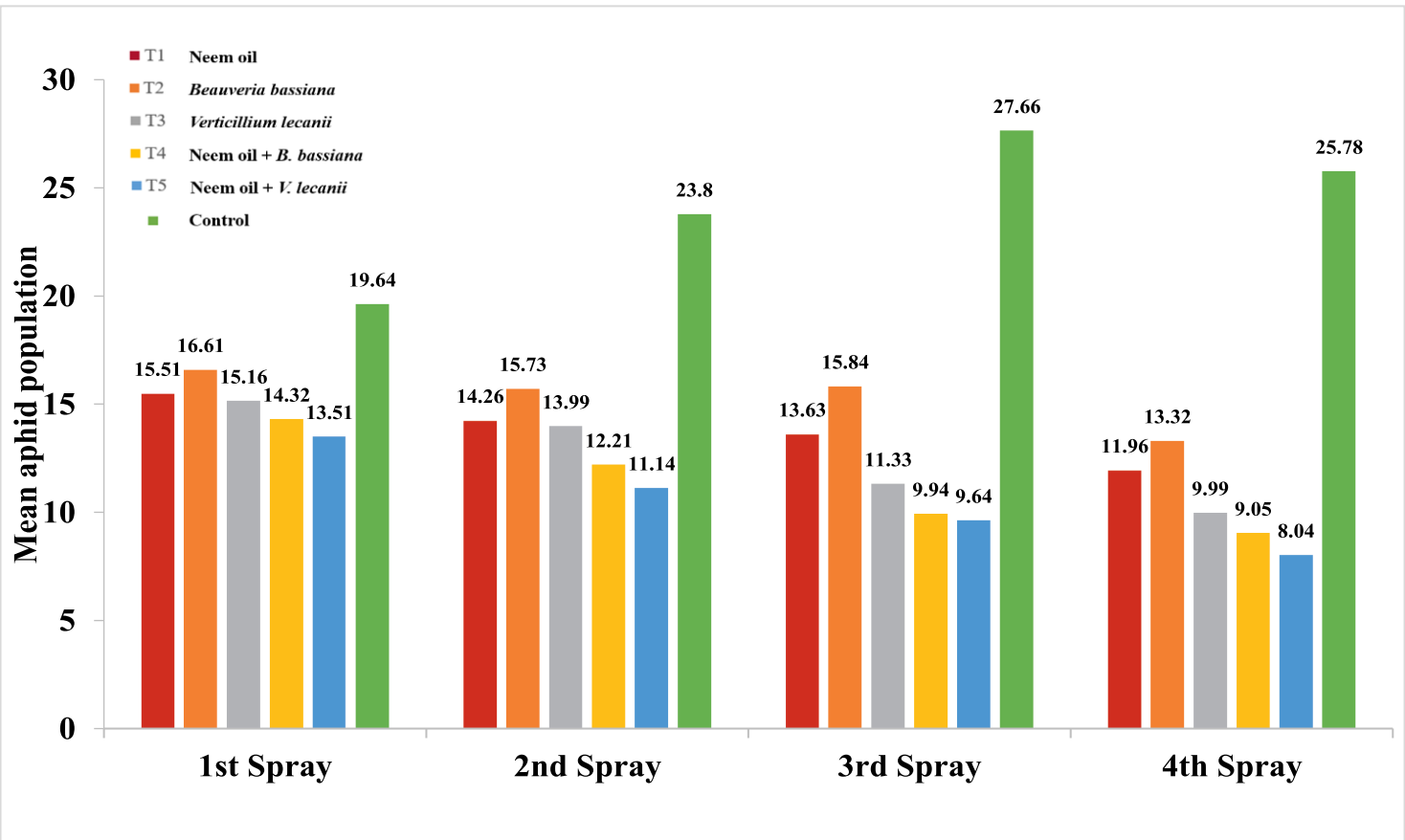

Fig 2. Effect of biopesticides based on mean population of aphid.

Thrips

In case of thrips, the data on efficacy of biopesticides after first spray revealed that no significant difference was observed among the treatments before spray. Maximum per cent reduction in thrips over control at seven days after spray was observed in combination of neem oil and *V.lecanii* (23.31%) while, a minimum percent reduction was seen in *B. bassiana* (11.14%). After second spray, the results on per cent reduction of thrips population at seven days after spray revealed that a combination of neem oil and *V. lecanii* (36.98%) was found most promising in reducing the population of thrips. Minimum reduction in thrips population was observed in *B. bassiana* (22.40%). A similar trend of efficacy of biopesticides was observed after third spray also. Combination of neem oil and *V. lecanii* was found superior above all the treatments at seven days after spray with a maximum per cent reduction in population of thrips over control (45.41%) while a minimum reduction was observed in *B. bassiana* (27.33%). A significant decline in the population of thrips was observed after fourth spray. Combination of neem oil and *V. lecanii* recorded the maximum per cent reduction in population of thrips over control (54.33%) followed by the combination of neem oil and *B. bassiana* (45.63%), *V. lecanii* (38.13%), neem oil (34.89%) and *B. bassiana* (30.80%) (Table 4). The data on the mean population of thrips revealed that the application of neem oil and *V. lecanii*showed minimum population of thrips from 35.53 to 25.37 thrips per three leaves at end of the fourth spray (Fig 3).

Table 4. Efficacy of biopesticides against thrips population on Bt cotton

Sr. No.	Treatment	Dose	Per cent reduction in thrips population over control at 7 DAS			
			1st Spray	2nd Spray	3rd Spray	4th Spray
T_1	Neem oil	5 ml/l	11.97	24.69	34.33	34.89
T_2	*Beauveria bassiana*	10 g/l	11.14	22.40	28.18	30.80
T_3	*Verticillium lecanii*	10 ml/l	16.14	26.86	35.73	38.13
T_4	Neem oil + *Beauveria bassiana*	5 ml/l + 10 g/l	18.09	32.38	39.88	45.63
T_5	Neem oil + *Verticillium lecanii*	5 ml/l + 10 ml/l	23.31	36.98	45.41	54.53

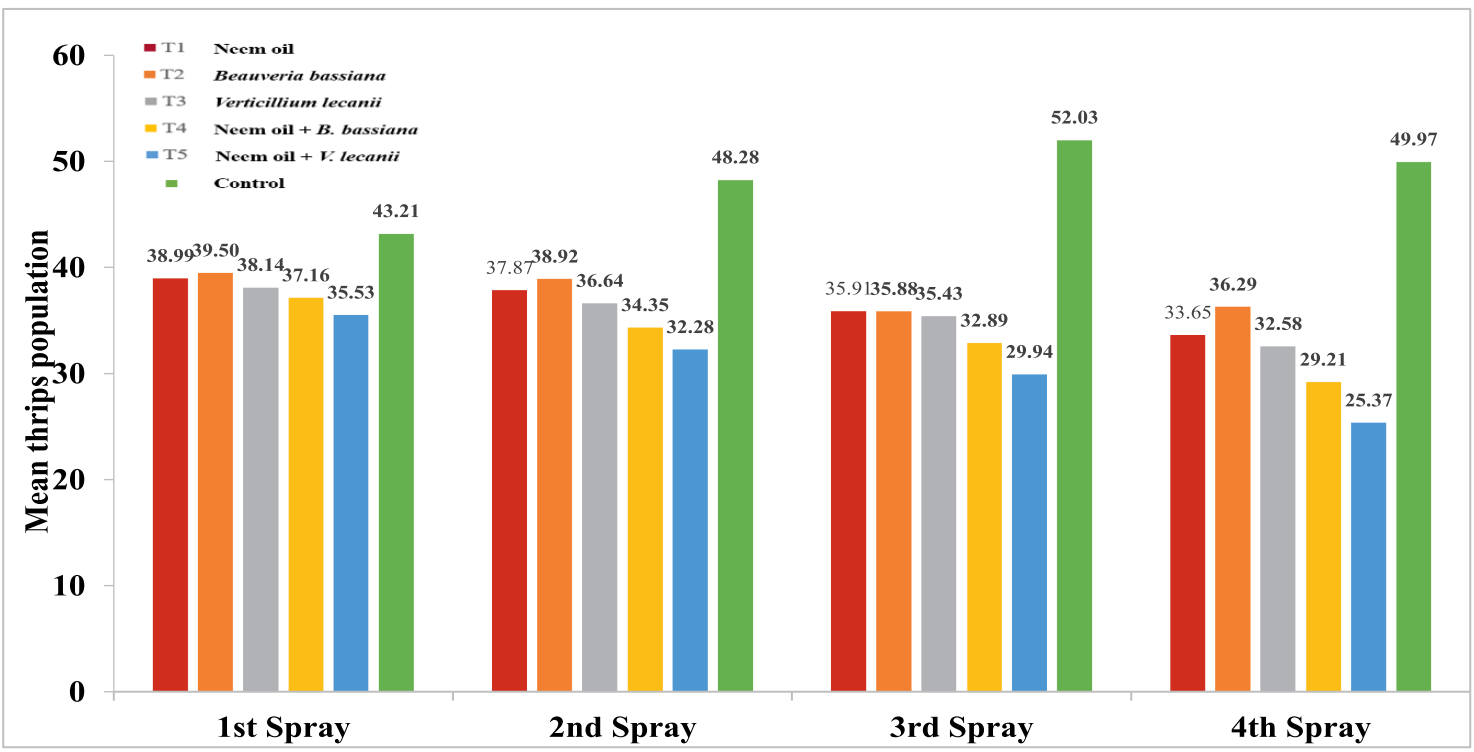

Fig 3. Effect of biopesticides based on mean population of thrips

Leafhopper

There was no significant difference in leafhopper population among the treatments before spraying. During first spray, the combination of neem oil and *V. lecanii* recorded highest per cent reduction in leafhopper population at seven days after spray (27.45%) whereas *B. bassiana* showed the minimum per cent reduction (15.80%). The data on per cent reduction of leafhopper population after second spray revealed that same efficacy pattern was found among the treatments. Combination of neem oil and *V. lecanii* recorded a maximum per cent reduction (43.70%) whereas, *B. bassiana* showed minimum reduction in population of leafhopper (31.67%). Similarly, after third spray, highest reduction was recorded in combination of neem oil and *V. lecanii* (52.39%) and lowest reduction was found in treatment *B. bassiana* (41.75%). Ultimately after fourth spray, the efficacy of different biopesticides against leafhopper was observed in the same pattern. Combination of neem oil and *V. lecanii* was found significantly superior among the treatments with maximum percent reduction in leafhopper population (62.73%) at seven days after spray. The next best treatment was the combination of neem oil and *B. bassiana* (55.47%) followed by *V. lecanii* (49.77%) and neem oil (45.92%) while *B. bassiana* showed a minimum reduction in leafhopper population (42.51%) (Table 5). Minimum population of leafhopper was recorded from 16.83 to 13.23 leafhopper per three leaves in combination of neem oil and *V. lecanii* from first to last spray (Fig 4).

Table 5. Efficacy of biopesticides against leafhopper population on Bt cotton

Sr. No.	Treatment	Dose	Per cent reduction in leafhopper population over control at 7 DAS			
			1st Spray	2nd Spray	3rd Spray	4th Spray
T_1	Neem oil	5 ml/l	19.12	34.16	42.95	45.92
T_2	*Beauveria bassiana*	10 g/l	15.80	31.67	41.75	42.51
T_3	*Verticillium lecanii*	10 ml/l	22.70	40.10	45.33	49.77
T_4	Neem oil + *Beauveria bassiana*	5 ml/l + 10 g/l	25.52	40.73	47.08	55.47
T_5	Neem oil + *Verticillium lecanii*	5 ml/l + 10 ml/l	27.45	43.70	52.39	62.73

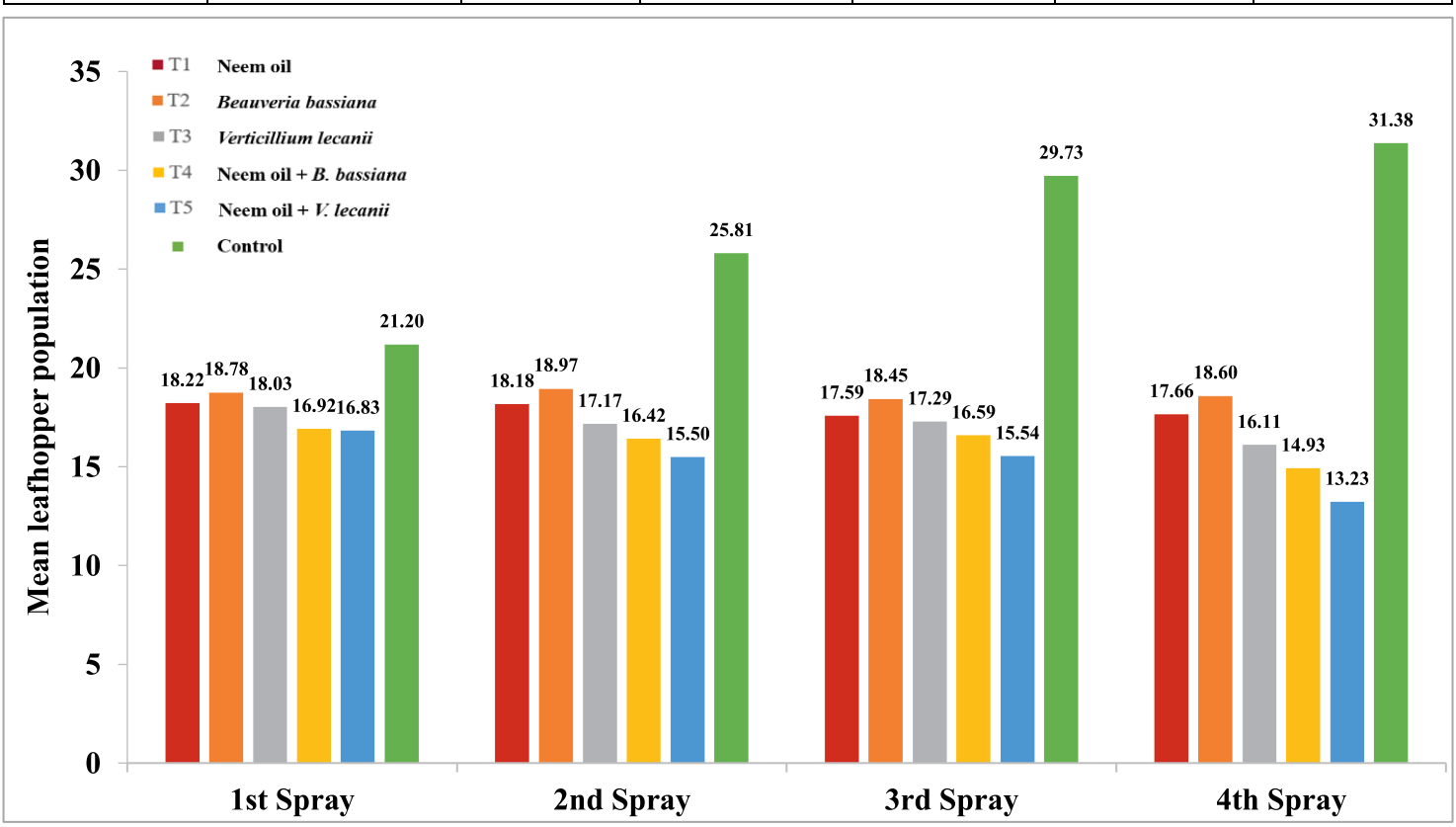

Fig 4. Effect of biopesticides based on mean population of leafhopper

DISCUSSION

In the present investigation,all the biopesticides and their combinations were found effective against sucking pests in *Bt* cotton. It was found that a combination of neem oil and *V. lecanii*and combination of neem oil and *B. bassiana* was found superior among all treatments in controlling sucking pests such as whitefly, aphid, thrips and leafhopper. Our results are in conformity with the findings of [14] who reported combination of Neem seed kernel extract (NSKE) and *V. lecanii* as highly effective in reduction of whitefly (from 0.75 to 0.47/3 leaves), aphid (from 4.77 to 4.67/3 leaves), thrips(from 15.13 to 3.85/3 leaves) and leafhopper (from 1.82 to 0.87/3 leaves) when compared to other microbial biopesticides. The results of [15] strongly support our results as they revealed that among different entomopathogenic fungals, *Lecanicillium lecanii* alone and its combination with neem oil were the best in recording highest percent reduction of whitefly (50.74% and 52.22%) and jassid (62.22% and 65.15%), respectively. [16] acknowledged *V. lecanii* as best in recording minimum population of aphid and thrips followed by *B. bassiana*. [17] observed that among biopesticides tested, neem oil, *V. lecanii*, *B. bassiana* and *Metarhizium anisopliae* were found effective for the control of major sucking pests. [18] reported the compatibility of neem product and microbial biopesticides. Many botanicals such as azadirachtin is have a diverse mode of action. Combining two effective biopesticides (neem oil and microbials) was found synergistic and show the enhanced effect which allows the higher reduction of pest population [19].

CONFLICT OF INTERESTS: Authors declare that they have no conflict of interests.

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