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Synergistic effects of insecticide and lactic acid bacteria formulation on *Spodoptera litura* and beneficial spider in cauliflower under field condition

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Abstract

Cauliflower (*Brassica oleracea* var. botrytis) is an important vegetable crop that is susceptible to various insect pests, including the polyphagous lepidopteran pest, *Spodoptera litura*. This research paper presents an integrated initiative of managing *S. litura* in cauliflower using a combination of selective insecticides and the co-application of lactic acid bacteria (LAB). Field experiments were conducted to evaluate the bio-efficacy of emamectin benzoate 05 % SG, tolfenpyrad 15 % EC and chlorantraniliprole 18.5 % SC, both with and without the addition of LAB @ 5 % during kharif and rabi seasons of 2024. The results demonstrated that the co-application of chlorantraniliprole 18.5 % SC at 10 g a.i. ha^{-1}, with LAB 5 %, was the most effective strategy for managing *S. litura*, as it led to a significant reduction in leaf damage (14.64-16.85 %) ($p<0.05$) and an enhanced natural enemy population. Additionally, the application of LAB alone was found to be effective in attracting predatory spiders (8.66-8.27 spiders per ten plants) ($p<0.05$) to the cauliflower plants through the induction of volatile compounds by the epiphytic microbiome. The experimental data were subjected to ANOVA analysis using SPSS software with square root transformation for spider population data and arc sine transformation for *S. litura* data. The treatments were found to be statistically significant. These findings suggest that integrating selective insecticides and lactic acid bacteria can provide a cost- effective and sustainable approach to managing *S. litura* in cauliflower production, offering a promising alternative to the overreliance on chemical pesticides.

Keywords

insecticides; lactic acid bacteria formulation; *S. litura*; spiders

Introduction

Cauliflower (*Brassica oleracea* L. var. *botrytis*) is a vegetable crop with edible curd consisting of a compact shoot system with reduced internodes, branches and apices. Nutritionally, cauliflower is rich in vitamins A, B, C and K, along with essential minerals like phosphorus, potassium, calcium, sodium and iron and 100 g serving provides 25 kcal, 4.97 g carbohydrates, 0.3 g fat, 1.92 g protein and 2 g dietary fibre (1). Its high vitamin C content (48.2 mg) is widely consumed in curries, soups and pickles. India leads global cauliflower and cabbage production, cultivating cauliflower over 13.7 lakh ha, yielding 9.53 million metric tonnes annually, with a productivity of 19.6 metric tonnes per hectare in 2024 (2).

Insect pests are crucial in agricultural losses, inflicting significant economic damage on crops. Estimates of annual global yield losses caused by DBM have been reported to exceed $4-5 billion, with exceptionally high impacts in regions that are major crucifer producers, such as India, where crop losses can be as high as 52 % due to caterpillar damage under severe infestation conditions (3). Among various pests ravaging the plant, *Spodoptera litura*, considered as highly destructive cosmopolitan insect pest species that poses significant challenges to cauliflower cultivation worldwide. *S. litura* larvae are notorious for rapidly defoliating crops, feeding on leaves and damaging floral structures, leading to weakened plants, reduced yields and severe economic losses for farmers. The increasing prevalence of *S. litura* has made effective management critical, as conventional pest control methods, heavily reliant on synthetic insecticides, have raised concerns regarding environmental sustainability, pest resistance and ecological imbalance.

The overuse of chemical insecticides has not only contributed to the development of insecticide resistance in *S. litura* populations but also led to the accumulation of harmful residues on crops, posing risks to human health and the environment. Additionally, indiscriminate use of insecticides disrupts agroecosystems, often harming non-target organisms, including natural enemies such as parasitoids and predators. This disruption can exacerbate pest issues by triggering secondary pest outbreaks, developing resistance and diminishing the resilience of agricultural systems (4).

Researchers focus on innovative pest management strategies that integrate chemical and biological control methods to overcome these challenges. One promising approach involves the synergistic application of insecticides and beneficial microorganisms, such as lactic acid bacteria (LAB). LABs are known for their antimicrobial properties, including producing organic acids, hydrogen peroxide and bacteriocins, which can exhibit insecticidal activity (5). Additionally, LAB enhances plant health and resilience, fostering a more robust agricultural system.

Studies indicate combining insecticides with LAB can improve pest suppression while minimizing negative environmental impacts (6). This integrated approach reduces the ecological footprint of pest control and supports the population of natural enemies, maintaining a balanced predator-prey dynamic. LAB can improve the gut health of natural enemies, enhancing their predatory efficiency against pests like *S. litura*. Similarly, plant growth-promoting microorganisms, such as actinomycetes, have been shown to boost crop yields and contribute to pest and pathogen management, further advancing sustainable agricultural practices.

In this study, an attempt has been made to combine the LAB formulation with insecticides to provide a synergistic effect in managing *S. litura* infecting cauliflower. By understanding the interactions between *S. litura*, insecticides and beneficial microorganisms, researchers and farmers can develop holistic solutions that enhance pest control, protect helpful organisms and promote long-term agricultural sustainability. This study explores the potential of integrating insecticides with LAB as a biocontrol agent for managing *S. litura* in cauliflower fields while maintaining ecological balance and enhancing crop productivity. The objectives include evaluating the synergistic effect of insecticides and LAB formulation on *S. litura* management and assessing their influence on the orientation of beneficial spiders within the cauliflower ecosystem.

Materials and Methods

Field experiment

Field experiments were conducted to evaluate the effectiveness of insecticides and LAB formulation against *S. litura* and natural enemies in cauliflower. The study was performed in Vellimalaipattinam at two different locations, location-1 (Lat 10.984034° & Long 76.791493 °) and location-2 (Lat 10.979909 ° & Long 76.76946 °) during two distinct seasons, kharif and rabi of 2024. The trials were arranged in a randomized block design (RBD) with eight treatments, including an untreated control and each treatment was replicated three times (Table 1). Following standard agronomic practices, cauliflower seedlings were raised in well-prepared nursery beds. Before transplanting, the experimental field was thoroughly ploughed with a tractor one month in advance, followed by three cross-ploughings using a power tiller. The soil was then pulverized and levelled. Fertilizers were applied at a rate of 100:80:60 kg/ha of NPK, using urea, single superphosphate and muriate of potash as nutrient sources. Thirty-day-old seedlings of the cauliflower variety CFL-1522 were transplanted at a spacing of 60 × 40 cm within the plots. Irrigation was provided immediately after transplanting and subsequently as needed. Weed management was performed manually three times at 30, 60 and 75 days after transplanting to maintain a weed-free environment.

The spray solution containing insecticides, with and without LAB formulation, was prepared individually for each treatment at the required concentrations. The lactic acid bacterial formulation was made from different sources, including a semisolid product of 100 g of milk powder, 1.0 kg of cane jaggery, 100 mL of one-day fermented grape juice and beaten egg in a selective fermentation process, curd and milk by performing serial dilutions (ranging from 10^4 to 10^6). This semisolid product containing LAB was cultured in agar plates and inoculated in MRS (de Mann Rogosa Sharpe) broth after three days of incubation in Petri plates (6). The cultured bacterial colonies were further studied for molecular identification from the total genomic DNA extracted in isolates using the standard cetyl hexadecyl - trimethyl ammonium bromide (CTAB) method and published in NCBI to obtain accession numbers. The formulated microbial consortia of LAB comprise *Lactococcus lactis* strain LAB 1-PP474431, *L. lactis* strain LAB 2-PP732186, *Lactobacillus paracasei* strain LAB 3-PQ469952 and *Lactiplantibacillus plantarum* strain LAB 4-PQ470018. These LAB-containing microbial consortia were subsequently utilized for experimental investigations. The LAB culture was prepared by inoculating the bacterial culture into MRS broth and left for three days to allow growth and fermentation. Once this incubation period was complete, the cultured broth was ready for immediate use. The microbial consortia containing LAB culture was prepared at a 5 %

Table 1. Treatments applied in the experiments and their corresponding dosages

Treatments	Dosage (mL/lit)
T1-Emamectin Benzoate 05 %SG	0.4 g/ L
T2-Tolfenpyrad 15 %EC	2 mL/L
T3- Chlorantraniliprole 18.5 % SC	0.1mL/L
T4-Emamectin Benzoate 05 % SG+LAB	0.4 g/L + 5 % LAB broth culture
T5-Tolfenpyrad 15 %EC +LAB	2 mL/L 5 % +5 % LAB broth culture
T6-Chlorantraniliprole 18.5 % SC +LAB	0.1mL/L +5 % LAB broth culture
T7- LAB alone	5 % LAB broth culture
T8- Untreated control	-

SG- Soluble granule; EC- Emulsifiable Concentrate; SC- Suspension concentrate; LAB- Lactic acid bacteria

concentration for the spraying application, corresponding to 50 mL of the cultured broth per 1 L of spray fluid. The treatments in the recommended dosage were applied using a battery-operated knapsack sprayer, utilizing 500 L of spray fluid per hectare. This concentration was maintained for all treatments in the field experiment study. Precautions were taken to prevent the spray solutions' drift between plots and ensure the plants' uniform coverage. For untreated control plots, only plain water was applied to the plants (7).

Field efficacy of treatments was recorded as the % leaf damage caused by *S. litura*. Ten plants were randomly selected in each plot and sampled at the top, middle and bottom canopy of three leaves from each plant to cover all strata of the plant. Insecticidal and LAB formulation treatments also influenced the populations of naturally occurring generalist predatory spiders, which were quantified by the counts of individuals found on 10 randomly selected plants within each plot during the dawn period of the day. The % leaf damage and natural enemy populations, particularly beneficial spiders, were monitored at 7-day intervals after each treatment application, with assessments conducted over two spray cycles..

Statistical analysis

The experimental data were subjected to statistical analysis following an RBD as outlined by (8). The experimental data were subjected to ANOVA analysis using SPSS software (version 16.00; SPSS Inc., USA) and mean values were compared using the least significant difference (LSD) method. To ensure appropriate statistical analysis, data transformations were performed, applying square root transformation for spider population data and arc sine transformation for *S. litura* data.

Results

Effect of insecticides and LAB formulation on % leaf damage induced by S. litura under field evaluation

The study assessed the effectiveness of insecticide treatments, with or without LAB, on % leaf damage caused by *S. litura* in cauliflower across two seasons under field condition, emphasizing the potential of combining chemical insecticides with biological agents like LAB. In season-1, chlorantraniliprole 18.5 % SC combined with LAB (T_6) emerged as the most effective treatment, achieving the lowest mean leaf damage (14.64 %). Similarly, tolfenpyrad 15 % EC with LAB (T_5) showed significant efficacy, outperforming Tolfenpyrad alone. Emamectin Benzoate 05 % SG with LAB (T_4) also improved pest control over its standalone counterpart. In contrast, LAB alone (T_7) demonstrated limited efficacy, while the untreated control (T_8) recorded the highest leaf damage (69.78 %) (Table 2). In season-2, the trends were consistent, with chlorantraniliprole 18.5 % SC combined with LAB (T_6) maintaining superior performance by achieving the lowest mean leaf damage (16.85 %). Tolfenpyrad 15 % EC with LAB (T5), followed by emamectin benzoate and 05 % SG with LAB (T_4), with moderate efficacy. LAB alone (T_7) showed limited pest suppression and the untreated control (T_8) recorded the highest leaf damage (70.57 %). Across both seasons, treatments that combined insecticides with LAB consistently outperformed standalone applications, providing a significant synergistic effect that enhanced pest suppression (Table 3). The pooled data analysis also showed that insecticide

Table 2. Effect of insecticides and LAB on crop damage by *S. litura* (Season-1)

Treatments	PTC	% leaf damage#						Overall mean	PRC (%)
		1st spray			2nd spray				
		7 DAS	14 DAS	Mean	7 DAS	14 DAS	Mean		
T1-Emamectin Benzoate 05 % SG	33.53 (35.38)	23.00 (28.63)b	27.78 (31.67)c	25.39 (30.15)c	27.98 (31.98)c	28.31 (31.61)bc	28.15 (31.80)c	26.77 (30.97)c	61.63
T2-Tolfenpyrad 15 % EC	34.34 (35.87)	21.24 (27.35)b	21.93 (27.89)cde	21.59 (27.62)cd	18.21 (25.22)d	23.33 (28.85)c	20.77 (27.03)cd	21.18 (27.32)cd	69.64
T3- Chlorantraniliprole 18.5 % SC	34.46 (35.94)	19.39 (26.24)b	19.65 (26.12)cde	19.52 (26.18)cde	19.14 (25.93)cd	20.35 (26.81)c	19.75 (26.37)cd	19.63 (26.27)de	71.86
T4-Emamectin Benzoate 05 % SG+LAB	34.48 (35.95)	17.12 (24.40)b	24.09 (29.34)cd	20.61 (26.87)cde	20.52 (26.85)cd	20.61 (26.75)c	20.57 (26.80)cd	20.59 (26.89)d	70.49
T5-Tolfenpyrad 15 % EC +LAB	34.89 (36.20)	15.93 (23.47)b	16.92 (24.24)de	16.43 (23.86)de	14.87 (22.56)d	17.61 (24.74)c	16.24 (23.65)d	16.33 (23.75)de	76.58
T6-Chlorantraniliprole 18.5 % SC +LAB	34.75 (36.12)	14.84 (22.58)b	14.92 (22.48)e	14.88 (22.53)e	13.70 (22.78)d	15.08 (21.70)c	14.39 (22.24)e	14.64 (22.39)e	80.03
T7- LAB alone	34.65 (36.06)	49.12 (44.49)a	51.11 (45.63)b	50.12 (45.06)b	49.43 (46.92)b	53.33 (44.67)b	51.38 (45.79)b	50.75 (45.43)b	27.27
T8- Untreated control	34.48 (35.95)	58.19 (50.00)a	68.33 (55.98)a	63.26 (52.99)a	74.49 (59.83)a	78.09 (65.77)a	76.29 (62.80)a	69.78 (57.89)a	-
SE.d	NS	4.04	3.03	2.25	3.14	3.08	3.12	1.93	-
CD (0.05)	NS	8.49	6.37	4.74	6.60	6.47	6.56	4.06	-
P- value @ 0.05	-	0.0037	0.0034	0.0038	0.0016	0.0070	0.0047	0.0053	-

LAB: Lactic acid bacteria; DAS: Days after spraying; PTC: Pre-treatment count; SG: Soluble granule; EC: Emulsifiable concentrate; SC: Suspension concentrate. Values in parentheses are *arc sine* values. #Mean of three replications.

Table 3. Effect of insecticides and LAB on crop damage by *S. litura* (Season-2)

Treatments	PTC	% leaf damage#						Overall mean	PRC (%)
		1st spray			2nd spray				
		7 DAS	14 DAS	Mean	7 DAS	14 DAS	Mean		
T1-Emamectin Benzoate 05 % SG	32.46 (34.72)	26.56 (33.19)bc	30.15 (31.02)c	28.36 (32.10)c	26.50 (30.96)c	27.19 (31.42)c	26.85 (31.19)c	27.60 (31.64)c	62.33
T2-Tolfenpyrad 15 % EC	32.58 (34.80)	21.69 (27.72)cd	23.66 (28.99)cd	22.68 (28.36)cd	21.01 (27.26)cde	25.86 (30.54)cd	23.44 (28.90)cd	23.06 (28.63)d	67.10
T3- Chlorantraniliprole 18.5 % SC	32.87 (34.98)	19.14 (25.90)cd	22.35 (28.17)cd	20.75 (27.03)d	19.15 (28.53)cd	22.92 (25.93)cd	21.04 (27.23)de	20.89 (27.13)de	70.48
T4-Emamectin Benzoate 05 % SG+LAB	32.33 (34.65)	20.39 (27.74)cd	21.67 (26.71)cd	21.03 (27.22)cd	16.56 (23.95)e	24.05 (29.32)cd	20.31 (26.64)de	20.67 (26.93)de	71.51
T5-Tolfenpyrad 15 % EC +LAB	32.64 (34.84)	19.14 (25.90)cd	19.62 (26.22)cd	19.38 (26.06)d	19.15 (26.45)de	19.89 (25.93)cd	19.52 (26.19)de	19.45 (26.13)de	72.60
T6-Chlorantraniliprole 18.5 % SC +LAB	33.12 (35.13)	16.27 (24.15)e	16.77 (23.78)d	16.52 (23.96)d	16.55 (24.94)de	17.79 (23.99)d	17.17 (24.47)e	16.85 (24.21)e	81.90
T7- LAB alone	32.74 (34.90)	40.99 (39.79)b	47.87 (43.74)b	44.43 (41.77)b	37.89 (37.98)b	41.39 (39.97)b	39.64 (38.98)b	42.04 (40.37)b	44.37
T8- Untreated control	32.66 (34.85)	63.89 (61.32)a	75.86 (53.24)a	69.88 (57.28)a	70.30 (58.32)a	72.22 (57.33)a	71.26 (57.82)a	70.57 (57.55)a	-
SE.d	NS	3.65	3.43	2.39	2.11	3.21	1.81	1.42	-
CD (0.05)	NS	7.67	7.22	5.03	4.45	6.75	3.81	2.99	-
P- value @ 0.05	-	0.00723	0.00153	0.00169	0.0034	0.00697	0.0013	0.0035	-

LAB: Lactic acid bacteria; DAS: Days after spraying; PTC: Pre-treatment count; SG: Soluble granule; EC: Emulsifiable concentrate; SC: Suspension concentrate. Values in parentheses are *arc sine* values. #Mean of three replications.

treatments combined with LAB were more effective than standalone insecticides in management of *S. litura* (Table 4). In pooled analysis, per-cent reduction over control (PRC) analysis showed that chlorantraniliprole 18.5 % SC + LAB (T_6) had the highest efficacy (80.71 %) in reducing *S. litura* damage, followed by tolfenpyrad 15 % EC + LAB (T_5) at (74.00 %). Chlorantraniliprole 18.5 % SC (T_3) alone achieved (70.02 %), while emamectin benzoate + LAB (T_4) and emamectin benzoate (T_1) had moderate effectiveness (71.59 % and 61.26 % respectively). The field evaluation of insecticides and LAB formulation on % leaf damage caused by *S. litura* revealed statistically significant effects (P<0.05) among the treatments (Table 2-4). These findings highlight the importance of integrating biological agents like LAB with synthetic chemical insecticides to achieve sustainable pest management, reduce crop damage and support agricultural productivity effectively.

Effect of insecticides and LAB formulation on the population of spiders under field evaluation

Across both seasons, significant differences in spider populations were observed among treatments, highlighting the influence of various pest management strategies on natural enemy conservation. In Season 1, the highest spider population was recorded in LAB alone (T_7), with a mean of 8.66 spiders/10 plants, showcasing its ability to create a favourable environment for beneficial arthropods. Similarly, chlorantraniliprole 18.5 % SC + LAB (T_6) supported a relatively higher spider population (6.79 spiders/10 plants), indicating the potential of integrating LAB with insecticides to maintain predator populations while achieving pest suppression. Treatments combining insecticides and LAB, such as tolfenpyrad + LAB (T_5) and emamectin benzoate + LAB (T_4), showed intermediate spider populations, with means of 6.35 spiders/10 plants and 6.25 spiders/10 plants, respectively, representing a balanced approach for supporting natural enemies (Table 5).

Table 4. Effect of insecticides and LAB on crop damage by *S. litura* (Season-1& 2) (Pooled)

Treatments	% leaf damage#						Overall mean	PRC (%)
	1st spray			2nd spray				
	7 DAS	14 DAS	Mean	7 DAS	14 DAS	Mean		
T1-Emamectin Benzoate 05 % SG	24.78 (30.91)c	28.97 (31.35)c	26.87 (31.13)c	27.24 (31.47)c	27.75 (31.51)c	27.50 (31.49)c	34.05 (35.52)c	61.98
T2-Tolfenpyrad 15 % EC	18.81 (27.53)cd	20.29 (28.44)cd	19.55 (27.99)cd	17.94 (26.24)d	21.74 (29.70)c	19.84 (27.97)cd	26.55 (31.44)d	67.10
T3- Chlorantraniliprole 18.5 % SC	20.19 (26.07)cd	22.14 (27.14)cde	21.17 (26.61)d	18.68 (27.23)cd	23.13 (26.37)cd	20.90 (26.80)de	25.57 (29.28)e	70.48
T4-Emamectin Benzoate 05 % SG+LAB	18.76 (26.07)cd	22.88 (28.02)cd	20.82 (27.05)d	18.54 (25.40)d	22.33 (28.03)cd	20.44 (26.72)de	33.54 (35.31)c	71.51
T5-Tolfenpyrad 15 % EC +LAB	17.83 (23.36)d	18.21 (23.13)e	18.02 (23.25)e	17.85 (23.86)d	19.07 (22.85)e	18.46 (23.35)e	20.61 (28.73)e	72.60
T6-Chlorantraniliprole 18.5 % SC +LAB	16.99 (24.69)d	17.27 (25.23)de	17.13 (24.96)de	16.43 (24.50)d	17.49 (25.34)cd	16.96 (24.92)de	20.34 (25.50)f	81.90
T7- LAB alone	45.06 (42.14)b	49.49 (44.69)b	47.27 (43.41)b	43.66 (42.45)b	47.36 (42.32)b	45.51 (42.38)b	46.39 (42.90)b	44.37
T8- Untreated control	61.04 (55.66)a	72.10 (54.61)a	66.57 (55.14)a	72.40 (59.07)a	75.16 (61.55)a	73.78 (60.31)a	71.33 (57.87)a	-
SE.d	2.70	2.03	1.59	2.04	3.24	1.82	0.89	-
CD (0.05)	5.67	4.28	3.35	4.29	6.82	3.84	1.87	-
P- value @ 0.05	0.0045	0.0036	0.0042	0.0015	0.0072	0.0047	0.0068	-

LAB: Lactic acid bacteria; DAS: Days after spraying; PTC: Pre-treatment count; SG: Soluble granule; EC: Emulsifiable concentrate; SC: Suspension concentrate. Values in parentheses are *arc sine* values. #Mean of three replications.

Table 5. Effect of insecticides and LAB on spider population (Season-1)

Treatments	PTC	% leaf damage[#]						Overall mean	% reduction/ increase over control
		1st spray			2nd spray				
		7 DAS	14 DAS	Mean	7 DAS	14 DAS	Mean		
T1-Emamectin Benzoate 05 % SG	4.33 (2.08)	4.08 (2.01)c	4.93 (2.43)c	4.51 (2.21)d	4.11 (2.20)d	5.33 (2.30)c	4.72 (2.17)d	4.61 (2.14)e	-40.28
T2-Tolfenpyrad 15 % EC	4.25 (2.06)	4.13 (2.03)bc	5.47 (2.38)c	4.80 (2.19)d	4.20 (2.04)cd	5.67 (2.32)c	4.94 (2.23)cd	4.87 (2.20)de	-38.52
T3- Chlorantraniliprole 18.5 % SC	4.86 (2.20)	4.47 (2.11)bc	6.13 (2.34)c	5.30 (2.30)d	4.60 (2.14)cd	6.07 (2.46)c	5.34 (2.31)bcd	5.32 (2.30)d	-34.56
T4-Emamectin Benzoate 05 % SG+LAB	4.67 (2.16)	4.83 (2.19)bc	7.07 (2.75)ab	5.95 (2.43)c	4.87 (2.20)cd	6.40 (2.50)bc	6.14 (2.37)bc	6.25 (2.40)c	-23.12
T5-Tolfenpyrad 15 % EC +LAB	4.73 (2.17)	5.00 (2.23)ab	7.13 (2.75)ab	6.07 (2.43)bc	5.40 (2.38)ab	7.87 (2.80)ab	6.64 (2.57)a	6.35 (2.52)b	-21.89
T6-Chlorantraniliprole 18.5 % SC +LAB	4.45 (2.11)	5.20 (2.28)a	8.00 (2.86)a	6.60 (2.56)ab	5.87 (2.42)ab	8.07 (2.83)a	6.97 (2.64)a	6.79 (2.60)ab	-16.48
T7- LAB alone	4.69 (2.17)	8.60 (2.84)ab	8.86 (2.97)ab	8.77 (2.96)ab	8.66 (2.94)abc	8.45 (2.90)a	8.56 (2.92)a	8.66 (2.94)ab	6.51
T8- Untreated control	4.83 (2.20)	7.93 (2.81)a	8.20 (2.93)a	8.07 (2.86)a	8.13 (2.85)a	8.27 (2.86)a	8.20 (2.83)a	8.13 (2.84)a	-
SE.d	NS	0.15	0.10	0.09	0.12	0.14	0.08	0.05	-
CD (0.05)	NS	0.33	0.23	0.19	0.24	0.31	0.17	0.12	-
P- value @ 0.05	-	0.0048	0.0050	0.0043	0.0011	0.0291	0.0025	0.0018	-

LAB: Lactic acid bacteria; DAS: Days after spraying; PTC: Pre-treatment count; SG: Soluble granule; EC: Emulsifiable concentrate; SC: Suspension concentrate. Values in parentheses are *square root (x + 0.5)* values. [#]Mean of three replications.

In Season 2, the trend was consistent, with the highest spider populations recorded in LAB alone (T_7) (8.27 spiders/10 plants) and untreated control (T_8) (8.09 spiders/10 plants), likely due to natural occurrences and the influence of volatiles from LAB that attract beneficial predators. Chlorantraniliprole 18.5 % SC + LAB (T_6) and tolfenpyrad 15 % EC + LAB (T_5) continued to demonstrate considerable spider populations, with means of 6.32 spiders/10 plants and 6.07 spiders/10 plants, respectively. Statistical analysis confirmed significant variations among treatments, underscoring the compatibility of LAB with chemical treatments in fostering natural enemy populations (Table 6). Pooled analysis revealed substantial variation in spider populations, with LAB alone (T_7) and untreated control (T_8) recording the highest means of 8.46 and 8.11 spiders/10 plants, highlighting natural predator dynamics and LABs' volatile-mediated attraction. Treatments like chlorantraniliprole 18.5 % SC + LAB (T_6) and tolfenpyrad 15 % EC + LAB (T_5) showed substantial spider populations of 6.55 and 6.21 spiders/10 plants, reflecting their balance between pest control and predator conservation (Table 7).

The combined PRC analysis across both seasons revealed significant differences in spider populations, highlighting the impact of treatments on natural enemies. The LAB alone (T7) recorded the highest spider densities, suggesting minimal disruption to predator populations. Among insecticide treatments, chlorantraniliprole 18.5 % SC + LAB (T_6) retained the highest spider population (6.55 spiders/10 plants) with a PRC of 20.23 %, followed by tolfenpyrad 15 % EC + LAB (T_5) (6.21 spiders/10 plants, PRC 23.42 %). Chlorantraniliprole 18.5 % SC (T_3) showed a PRC of 34.40 %, while emamectin benzoate + LAB (T_4) and emamectin benzoate (T_1) exhibited PRCs of 28.48 % and 39.86 %, respectively, indicating moderate effects on spider populations. Notably, the LAB-alone treatment (T_7) had 4.57 % increase over control suggesting increased spider abundance compared to the control, emphasizing its role in natural enemy conservation (Table 7). The field evaluation of insecticides and LAB formulation on population dynamics of beneficial spiders revealed statistically significant effects ($P<0.05$) among the treatments. These results highlight the ecological benefits of LAB integration in pest management systems for enhancing predator conservation and promoting sustainable agriculture.

Discussion

Effect of insecticides and LAB formulation on % leaf damage induced by S. litura under field evaluation

The combined application of chlorantraniliprole 18.5 % SC with LAB demonstrated the highest effectiveness in mitigating *S. litura* damage, followed closely by tolfenpyrad 15 % EC with LAB. While chlorantraniliprole alone showed considerable impact, emamectin benzoate with and without LAB exhibited moderate control. These results underscore the significance of integrating microbial agents with chemical insecticides for enhanced pest suppression and sustainable crop protection. The research findings showed that the co-application of LAB at 5 % concentration with insecticides, including chlorantraniliprole, tolfenpyrad and emamectin benzoate, significantly enhances their effectiveness and demonstrates compatibility, establishing a promising integrated pest management approach (9). The relative toxicity of eight insecticides (indoxacarb, flubendiamide, chlorantraniliprole, chlorfenapyr, novaluron, emamectin benzoate, betacyfluthrin, quinalphos) against *S. litura* on soybean was assessed. The study found that chlorantraniliprole resulted in the lowest larval population (5.90 larvae/10 plants), followed by emamectin benzoate (9.86 larvae), indoxacarb (13.91 larvae) and flubendiamide (14.52 larvae). Insecticide treatments significantly reduced pest numbers compared to the untreated control (10).

Table 6. Effect of insecticides and LAB on spiders population (Season 2)

Treatments	PTC	No. of spiders/ ten plants#						Overall mean	% Reduction/ Increase over control
		1st spray			2nd spray				
		7 DAS	14 DAS	Mean	7 DAS	14 DAS	Mean		
T1-Emamectin Benzoate 05 % SG	4.45 (2.10)	4.33 (2.08)f	4.67 (2.15)e	4.50 (2.12)f	4.40 (2.10)d	5.47 (2.20)c	4.94 (2.22)e	4.72 (2.17)d	-39.65
T2-Tolfenpyrad 15 % EC	4.66 (2.15)	4.40 (2.10)ef	5.40 (2.32)d	4.90 (2.21)e	4.47 (2.12)d	5.20 (2.27)c	4.84 (2.20)e	4.87 (2.21)d	-38.80
T3- Chlorantraniliprole 18.5 % SC	4.53 (2.12)	4.47 (2.11)e	6.27 (2.50)c	5.37 (2.32)d	4.53 (2.13)d	6.00 (2.53)b	5.27 (2.30)d	5.32 (2.31)c	-34.23
T4-Emamectin Benzoate 05 % SG+LAB	4.43 (2.10)	4.60 (2.14)de	6.40 (2.53)c	5.50 (2.35)cd	5.47 (2.34)cd	6.73 (2.59)b	6.10 (2.47)cd	5.80 (2.41)c	-28.30
T5-Tolfenpyrad 15 % EC +LAB	4.56 (2.13)	5.20 (2.28)cd	6.53 (2.55)bc	5.87 (2.42)bc	5.13 (2.26)bc	7.40 (2.71)ab	6.27 (2.50)bc	6.07 (2.46)b	-24.96
T6-Chlorantraniliprole 18.5 % SC +LAB	4.87 (2.20)	5.62 (2.37)bc	7.13 (2.67)b	6.37 (2.52)b	5.60 (2.37)bc	6.93 (2.63)b	6.29 (2.51)bc	6.32 (2.52)b	-21.87
T7- LAB alone	4.66 (2.15)	7.73 (2.78)ab	8.47 (2.90)a	8.10 (2.85)a	8.40 (2.89)a	8.47 (2.90)a	8.44 (2.91)a	8.27 (2.87)a	4.73
T8- Untreated control	4.68 (2.16)	7.87 (2.86)a	8.07 (2.84)a	8.20 (2.86)b	7.79 (2.81)ab	8.20 (2.83)a	7.97 (2.82)b	8.09 (2.84)b	-
SE.d	NS	0.09	0.06	0.04	0.10	0.09	0.06	0.03	-
CD (0.05)	NS	0.19	0.13	0.10	0.21	0.20	0.14	0.08	-
P- value @ 0.05	-	0.0012	0.0220	0.0026	0.0035	0.0048	0.0025	0.0016	-

LAB: Lactic acid bacteria; DAS: Days after spraying; PTC: Pre-treatment count; SG: Soluble granule; EC: Emulsifiable concentrate; SC: Suspension concentrate. Values in parentheses are *square root (x + 0.5)* values. #Mean of three replications.

Table 7. Effect of insecticides and LAB on spiders population (Season-1 &2) (Pooled)

Treatments	No. of spiders/ ten plants#						Overall mean	% Reduction/ Increase over control
	1st spray			2nd spray				
	7 DAS	14 DAS	Mean	7 DAS	14 DAS	Mean		
T1-Emamectin Benzoate 05 % SG	4.21 (2.18)e	4.80 (2.14)d	4.50 (2.12)g	4.26 (2.06)b	5.40 (2.25)e	4.83 (2.20)c	4.67 (2.16)e	-39.86
T2-Tolfenpyrad 15 % EC	4.27 (2.30)de	5.44 (2.30)cd	4.85 (2.20)f	4.34 (2.08)b	5.44 (2.30)de	4.89 (2.21)c	4.87 (2.21)d	-38.85
T3- Chlorantraniliprole 18.5 % SC	4.47 (2.41)cd	6.20 (2.36)c	5.34 (2.31)ef	4.57 (2.14)b	6.04 (2.49)cd	5.30 (2.30)b	5.32 (2.31)c	-34.40
T4-Emamectin Benzoate 05 % SG+LAB	4.72 (2.42)bc	6.74 (2.59)b	5.73 (2.39)de	5.17 (2.27)b	6.57 (2.55)bc	5.87 (2.42)b	5.80 (2.41)c	-28.48
T5-Tolfenpyrad 15 % EC +LAB	5.10 (2.58)bc	6.83 (2.60)b	5.97 (2.44)cd	5.27 (2.30)a	7.64 (2.75)a	6.45 (2.54)a	6.21 (2.49)b	-23.42
T6-Chlorantraniliprole 18.5 % SC +LAB	5.40 (2.74)ab	7.57 (2.72)ab	6.48 (2.55)b	5.74 (2.40)a	7.50 (2.73)ab	6.62 (2.57)a	6.55 (2.56)b	-20.23
T7- LAB alone	8.17 (2.62)b	8.67 (2.71)ab	8.44 (2.90)bc	8.53 (2.73)a	8.46 (2.69)abc	8.50 (2.92)a	8.46 (2.91)b	4.57
T8- Untreated control	7.90 (2.81)a	8.14 (2.85)a	8.02 (2.83)a	8.07 (2.86)a	8.24 (2.84)a	8.09 (2.85)a	8.11 (2.85)a	-
SE.d	0.09	0.07	0.05	0.08	0.09	0.05	0.03	-
CD (0.05)	0.19	0.16	0.12	0.17	0.20	0.12	0.08	-
P- value @ 0.05	0.0029	0.0085	0.0045	0.0076	0.0032	0.0020	0.00100	-

LAB: Lactic acid bacteria; DAS: Days after spraying; PTC: Pre-treatment count; SG: Soluble granule; EC: Emulsifiable concentrate; SC: Suspension concentrate. Values in parentheses are *square root (x + 0.5)* values. #Mean of three replications.

Research indicates that chlorantraniliprole demonstrated the highest efficacy in suppressing *P. xylostella* larvae, achieving a 91.30 % reduction over the control, with an average of 1.02 larvae per plant (11). The effectiveness of chlorantraniliprole 600 g/L was studied with standard insecticides for controlling *S. litura* and *Helicoverpa armigera*. Two foliar applications at ten-day intervals (40 and 30 g a.i./ha) led to a significant decline in larval populations without inducing phytotoxicity in cotton (12). In the studies on the efficacy of various insecticides against *S. litura* in soybean, chlorantraniliprole at 30 g a.i./ha demonstrated the highest effectiveness, followed by spinetoram at 15 g a.i./ha. Emamectin benzoate (11 g a.i./ha), flubendiamide (48 g a.i./ha), indoxacarb (30 g a.i./ha) and lambda-cyhalothrin (15 g a.i./ha) exhibited comparable performance (13).

Effect of insecticides and LAB formulation on the population of spiders under field evaluation

Among the various insecticide treatments evaluated, chlorantraniliprole 18.5 % SC combined with LAB proved the most favourable for maintaining spider populations, highlighting its compatibility with beneficial predators. Tolfenpyrad 15 % EC combined with LAB also supported a relatively high spider presence, further emphasizing the potential of microbial additives in reducing the adverse effects of chemical insecticides. The standalone application of chlorantraniliprole 18.5 % SC demonstrated moderate impact, while emamectin benzoate, with and without LAB, showed a deviation in the orientation of spider populations towards the plant. Notably, the treatment with LAB alone resulted in the least disruption, reinforcing its role in conserving natural enemies and promoting ecological balance. The findings of the study closely corroborate the findings of field experiments conducted in rice to assess the impact of flubendiamide combined with a LAB formulation on the abundance of natural enemies (7). Coccinellids, spiders and rove beetle populations were significantly higher in plots treated solely with LAB, with densities recorded at 3.49, 6.96 and 8.66 per 10 hills, respectively. In comparison, plots treated with flubendiamide, alone or in combination with LAB, exhibited lower densities ranging between 1.47-2.48, 3.54-5.18 and 5.02-6.70 per 10 hills, respectively. This suggests that LAB applications provide a more conducive environment for natural enemy populations than flubendiamide treatments. The cyantraniliprole 10.26 OD, chlorantraniliprole 18.50 SC and flubendiamide 20 WG insecticides exhibited relatively lower toxicity towards natural enemies, particularly spiders and coccinellids. Consequently, cyantraniliprole 10.26 OD at 60 g a.i./ha, chlorantraniliprole 18.50 SC at 10 g a.i./ha and flubendiamide 20 WG at 18.24 g a.i./ha are promising candidates for inclusion in integrated pest management strategies targeting the conservation of natural enemies population especially, coccinellids and spiders in cole crops (14). The impact of insecticides on natural enemy populations while managing *S. litura* in soybean. Chlorantraniliprole (30 g a.i./ha) and spinetoram (15 g a.i./ha) were the least disruptive to beneficial arthropods, preserving predator abundance (15). In contrast, emamectin benzoate (11 g a.i./ha) and lambda-cyhalothrin (15 g a.i./ha) posed a more significant threat to natural enemies, indicating their potential adverse effects on ecological balance.

Conclusion

The underlying mechanism through which LAB contributes to the reduction of *S. litura* populations remains unclear; however, it is hypothesized that its volatile compounds significantly influence insect behaviour and plant-insect interactions. Notably, experimental plots treated with LAB exhibited a substantially higher abundance of beneficial arthropods than other treatments, including the untreated control, suggesting its potential to enhance biological control. Among the various treatment combinations, integrating chlorantraniliprole 18.5 % SC at 10 g a.i ha^{-1} with formulated LAB 5 % emerged as the most effective strategy for managing *S. litura*. This combination provided substantial suppression of pest populations and demonstrated greater compatibility with natural enemies, ensuring minimal disruption to beneficial insect communities. The findings underscore the significance of incorporating LAB-based formulation into pest management programs to optimize control efficacy while preserving ecological balance. This integrated approach offers a sustainable alternative to conventional insecticidal applications, emphasizing the need for strategies that simultaneously enhance pest suppression and support biodiversity conservation in agricultural ecosystems. Future research should focus on elucidating the precise biochemical pathways through which LAB-derived VOCs influence pest behaviour and predator orientation. Additionally, long-term field studies assessing the persistence of LAB in agroecosystems, its impact on soil microbiota and its potential role in pesticide residue degradation will further strengthen its application in sustainable agriculture. Advancing knowledge on LAB interactions with different insect pests and crop species will contribute to refining eco-friendly pest management approaches that minimize reliance on synthetic pesticides while enhancing agricultural biodiversity.

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Authors' contributions

MJ contributed to the conceptualization, methodology, data curation, visualization and writing of the original manuscript and provided lab study resources. YSJTE assisted with guidance in conducting experiments, contributed to the methodology, corrected the conceptual aspects, edited and assisted with data analysis. AS was involved in the method, writing, resource collection, revision and editing of the overall manuscript. MK contributed to the visualization of the methodology and assisted in revising the manuscript. RA provided guidance on bacterial species and supervised the study. RK offered supervision, contributed to the writing and revision of the manuscript and assisted with data analysis.

Compliance with ethical standards

Conflict of interest: The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper for the publication of this work.

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