



Original Article

Effectiveness of a nanoemulsion formulation of *Piper aduncum* L. extract and *Cymbopogon nardus* L. hydrosol in controlling the main pests of broccoli

Efektivitas formulasi nanoemulsi ekstrak *Piper aduncum* L. dan hidrosol *Cymbopogon nardus* L. untuk pengendalian hama utama tanaman brokoli

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ABSTRACT

A nanoemulsion formulation of a mixture of *Piper aduncum* extract and *Cymbopogon nardus* hydrosol was evaluated for its effectiveness against the major broccoli pests, *Crociodolomia pavonana* and *Plutella xylostella*. The study was conducted in Alahan Panjang, West Sumatra, using a completely randomized design with four treatments (the nanoemulsion formulation, *Bacillus thuringiensis*, cypermethrin, and a control) and seven replications. Treatments were applied weekly from 21 to 70 days after planting (DAP). Larval population density and plant damage were recorded, and the data were analyzed using analysis of variance (ANOVA) followed by a least significant difference (LSD) test at a 5% significance level. Infestation by *P. xylostella* occurred earlier at 21 DAP, whereas infestation by *C. pavonana* began at 28 DAP. Overall, the population density of *P. xylostella* was higher than that of *C. pavonana*. The nanoemulsion formulation exhibited insecticidal activity, although its effectiveness was lower than that of cypermethrin and *B. thuringiensis*. The effectiveness rates against *C. pavonana* were 71.49%, 48.54%, 33.33%, and against *P. xylostella* were 73.49%, 60.88%, and 57.52% for cypermethrin, *B. thuringiensis*, and the nanoemulsion formulation, respectively.

Key words: *Bacillus thuringiensis*, *Crociodolomia pavonana*, cypermethrin, insecticidal activity, *Plutella xylostella*

ABSTRAK

Formulasi nanoemulsi campuran ekstrak *Piper aduncum* dan hidrosol *Cymbopogon nardus* dievaluasi efektivitasnya terhadap hama utama brokoli, *Crociodolomia pavonana* dan *Plutella xylostella*. Penelitian ini dilakukan di Alahan Panjang, Sumatera Barat, menggunakan rancangan acak lengkap dengan empat perlakuan, yaitu formulasi nanoemulsi, *Bacillus thuringiensis*, cypermethrin, dan kontrol, dengan tujuh ulangan. Perlakuan diterapkan setiap minggu dari 21 hingga 70 hari setelah tanam (HST). Kepadatan populasi larva dan kerusakan tanaman dicatat, dan data dianalisis menggunakan analisis varians (ANOVA) diikuti dengan uji perbedaan terkecil yang signifikan (LSD) pada tingkat signifikansi 5%. Infestasi oleh *P. xylostella* terjadi lebih awal pada 21 HST, sedangkan infestasi oleh *C. pavonana* dimulai pada 28 HST. Secara keseluruhan, kepadatan populasi *P. xylostella* lebih tinggi daripada *C. pavonana*. Formulasi nanoemulsi menunjukkan aktivitas insektisida, meskipun efektivitasnya lebih rendah daripada sipermetrin dan *B. thuringiensis*. Tingkat efektivitas terhadap *C. pavonana* adalah 71,49%, 48,54%, 33,33%, dan *P. xylostella* adalah 73,49%, 60,88%, dan 57,52% masing-masing untuk sipermetrin, *B. thuringiensis*, dan formulasi nanoemulsi.

Kata kunci: aktivitas insektisida, *Bacillus thuringiensis*, *Crociodolomia pavonana*, cypermethrin, *Plutella xylostella*

INTRODUCTION

Broccoli (*Brassica oleracea* L. var. *italica*) contains high levels of fiber, minerals, and antioxidants, which are beneficial for health (Handayani & Ayustaningwarno 2014). According to Statistic Indonesia (BPS) (2023), cabbage and broccoli production in West Sumatra decreased by 17.62%, from 211,711 tons in 2020 to 174,387 tons in 2021. One of the contributing factors to this decline is the incidence of plant-disrupting organisms, including of plant pests, pathogens, and weeds. Pests and diseases are major causes of yield instability in cabbage and broccoli crops. Among the most damaging insect pests affecting these crops are cabbage head caterpillar (*Crocidolomia pavonana* Fab.) and the diamondback moth (*Plutella xylostella* Linn.), both of which are key pests of Brassicaceae crops (Jamtsho et al. 2021).

Farmers continue to rely on synthetic insecticides to control *C. pavonana* and *P. xylostella*, primarily due to their practicality, cost-efficiency, and quick action in reducing pest populations (Indiati & Marwoto 2017). However, if used unwisely, synthetic insecticides can have adverse effects such as pest resistance, pest resurgence, and mortality of non-target organisms (Indiati & Marwoto 2017). Insecticides, including carbamate and organophosphates, are extensively utilized in agriculture and have the potential to lethally affect various non-target organisms, such as earthworms, natural enemies (predators and parasitoids), and other ecologically significant species. Research indicates that the insecticides carbosulfan, carbofuran, and BPMC result in the mortality of the earthworm *Eisenia fetida*, with mortality rates ranging from 66% to 76% at elevated concentrations (300 mg/kg) (Kinasih et al. 2014). Therefore, botanical insecticides are considered a promising alternative, as they offer a more environmentally friendly approach and help mitigate the negative consequences associated with synthetic insecticide use.

Botanical insecticides offer several advantages, such as biodegradability, environmental friendliness, and a lower risk of inducing pest resistance (Yusuf 2012). Several plant species possess the potential to be developed into effective insecticides. According to previous reports, spiked pepper (*Piper aduncum* L.) contains secondary metabolites such as dillapiole, alkaloids, flavonoids, phenolics, triterpenoids, steroids, saponins, and coumarins. *P. aduncum* acts as a contact poison, a stomach poison, and an antifeedant (Arneti 2012). The secondary metabolites produced by *P. aduncum* are known to possess insecticidal activity (Safrida et al. 2020). Spiked pepper has been utilized as

a plant-based insecticide in conventional formulations such as emulsifiable concentrate (EC) and wettable powder (WP). Botanical insecticides are generally considered environmentally friendly because they are biodegradable and tend to degrade rapidly in the environment after application. However, in EC and WP formulations still present several limitations, including phytotoxicity, the presence of residues on plant surfaces after application, and reduced adhesion or persistence on foliage (Lina 2014). To overcome these limitations, nanoemulsion formulations have been developed to create more effective botanical insecticidal products.

In nanoemulsion systems, two phases are involved: an organic phase and an aqueous phase. The organic phase contains plant-derived active compounds dissolved in a suitable solvent, while the aqueous phase consists of water and emulsifying agents to stabilize the emulsion. These two phases are mixed using a spontaneous emulsification technique to produce stable nanoemulsion (Lina et al. 2021). In addition to *P. aduncum*, citronella (*Cymbopogon nardus* L.) is also a potential source of plant-based insecticides. The distillation of citronella grass produces two types of liquid fractions: the essential oil itself and a by-product known as hydrosol. In this study, citronella hydrosol was used as the aqueous phase in the nanoemulsion formulation because it is environmentally friendly, readily biodegradable, and may enhance insecticidal activity compared with formulations using plain water. Hydrosol is a water-based distillation product that contains small amounts of active compounds originating from citronella essential oil, such as citronellal, citronellol, and geraniol (Saidi et al. 2015). Although present in low concentrations, these compounds exhibit biological activity, including repellent and antifeedant effects against insect pests (Lina et al. 2021). Therefore, the use of hydrosol not only serves as a carrier in the aqueous phase but also contributes additional bioactive compounds to the nanoemulsion system.

Plant-based insecticides, particularly those developed using nanotechnology, must be formulated in a stable form to ensure their efficacy. Nanoemulsion technology enables the reduction of particle size, thereby enhancing the efficiency and effectiveness of the active ingredients (Noveriza et al. 2017). Nanoemulsion formulations have been developed to improve the performance of botanical insecticides compared with earlier and commonly used formulations like EC and WP. These nanoparticle-based formulations offer several advantages, including enhanced penetration into leaf tissues, extended shelf life, resistance to sedimentation, low volatility, and improved physicochemical stability

(Shakeel et al. 2008). A nanoemulsion formulation of *P. aduncum* extract has been shown to cause significant mortality in *C. pavonana* larvae, with an LC₉₅ value of 1.02% (Erlina et al. 2020). Furthermore, Lina et al. (2023a) reported that nanoemulsion formulations of *P. aduncum* showed high larval mortality against *Spodoptera frugiperda* Smith. Recent studies have demonstrated that nanoemulsion-based botanical insecticides enhance the stability and bioactivity of active compounds, leading to increased larval mortality and physiological disruption in insect pests, including *S. frugiperda* (Campos et al. 2020; de Oliveira et al. 2021).

Prior to commercialization, botanical insecticides must undergo thorough testing. Efficacy testing is a critical step in evaluating the performance of a pesticide product intended for commercial use (Fitriasari et al. 2009). This study aimed to evaluate the effectiveness of a nanoemulsion formulation containing *P. aduncum* extract and *C. nardus* hydrosol in controlling *C. pavonana* and *P. xylostella* on broccoli crops in Alahan Panjang.

MATERIALS AND METHODS

Time and place

This study was conducted in Nagari Alahan Panjang, Lembah Gumanti District, Solok Regency, from July to October 2023. Botanical insecticide nanoemulsions were formulated at the Insect Bioecology Laboratory, Department of Plant Protection, Faculty of Agriculture, Andalas University.

Making hydrosols

Hydrosols, also known as hydrolates, are byproducts of the essential oil distillation process and typically contain trace amounts of volatile compounds derived from the essential oil, generally less than 0.02%. These compounds originate from citronella essential oil and remain dispersed in the aqueous distillate during the distillation process. In this study, citronella (*Cymbopogon nardus*) hydrosol was used as the aqueous phase of the nanoemulsion formulation. The hydrosol was obtained from Laing Farmers, Solok Regency, West Sumatra, Indonesia.

Preparation of *P. aduncum*

Piper aduncum was utilized as the primary source material for the plant-based insecticide. The *P. aduncum* fruits selected were green, hard-texture, and free of mold. The fruits were collected and transported to the Insect Bioecology Laboratory, Department of Plant Protection, Faculty of Agriculture, Andalas University. They were cut into small pieces and placed on a paper-lined tray with a diameter of 60 cm. The fruits were

air-dried at room temperature for approximately three weeks until the moisture content was reduced to below 12%. Finally, the dried *P. aduncum* fruits were pulverized into a fine powder using a blender (Lina et al. 2018).

Extraction of *P. aduncum*

The spiked pepper powder was extracted using the maceration method with ethyl acetate as the solvent. A total of 100 g of the powdered material was placed in an Erlenmeyer flask and soaked in ethyl acetate until the final volume reached 1 liter. The maceration process was carried out for 48 hours. The resulting extract was filtered using a glass funnel lined sequentially with Whatman No. 1 and No. 41 filter papers. The filtrate was then concentrated using a rotary evaporator at 45 °C and a pressure of 277 mbar (Lina 2014). The concentrated extract was transferred into a storage bottle and stored in refrigerator until further formulation.

Nanoemulsion formulation

The *P. aduncum* nanoemulsion was prepared using the spontaneous emulsification method, comprising an aqueous phase (90%) and an organic phase (10%). The aqueous phase, consisting of 87% lemon grass hydrosol and 3% Tween 80, was subjected to a magnetic homogenization process and stirred for 30 minutes at 2500 rpm. The organic phase was prepared by mixing the *P. aduncum* extract (5%) and 96% ethanol (5%) in an Erlenmeyer flask. To formulate the emulsion, the organic phase was slowly dripped into the aqueous phase using a Mohr pipette under continuous magnetic stirring for 45 min (Lina 2014). The obtained *P. aduncum* nanoemulsions were stored in a refrigerator until further use. Following formulation completion, droplet size was measured using a particle size analyzer, which confirmed that the nanoemulsion had a mean particle size of 104.2 nm (Lina et al. 2023b).

Site selection and land preparation

This efficacy trial began with a survey of potential locations. Nagari Alahan Panjang located in Lembah Gumanti District, Solok Regency, West Sumatra, was selected as the experimental site due to its highland characteristics, which are typical for cabbage and broccoli production centers. The land was prepared by clearing rocks, grasses, shrubs, and trees. The prepared area was divided into 28 experimental plots, and planting beds were constructed manually using hoes. Each bed measured 10 m in length, 1.5 m in width, and approximately 30 cm in height, with a 50 cm spacing between beds. Organic manure was applied at a rate

of 100 g per plant, and NPK fertilizer (16:16:16) was applied at a dose of 4 g per plant during transplanting.

Broccoli cultivation

Broccoli seeds of the Green Magic variety were used. At 21 days after sowing, the seedlings were transferred to the experimental plots, totalling 60 plants per plot. Broccoli seedlings were planted in each bed at a distance of 50 cm × 50 cm. Population monitoring and damage assessments for *C. pavonana* and *P. xylostella* were conducted on six randomly selected sample plants per plot. Sample plants were selected using a simple random sampling lottery technique.

Field applications

Spraying was initiated following the detection of *P. xylostella* larvae, which typically appear earlier in the growing season. Control actions to *P. xylostella* were taken once the pest population exceeded the economic threshold, defined as the presence of 0.5 larvae per plant or a cumulative total of five of third and fourth instar larvae per 10 plants (Winarto & Bayang 2015). The concentration of the botanical insecticide used in the field was determined based on prior laboratory toxicity tests, specifically at $2 \times LC_{95}$, equivalent to 2.04% (Erlina et al. 2020). For comparison, two commercial insecticides were applied at the manufacturer's recommended dosages: Thuricide HP (active ingredient: *Bacillus thuringiensis*, biological insecticide) and Instop 311 EC (active ingredient: cypermethrin 311 g/l, synthetic insecticide).

All insecticides were applied using a knapsack sprayer at weekly intervals. As determined by calibration, the spray volume varied depending on the plant growth stage. The application volume was 2 l per plot at the early growth stage, 5 l per plot from 28 to 42 days after planting (DAP), and 10 l per plot from 49 to 70 DAP. The insecticide concentration used in the spray solution was 20.4 ml/l.

Variables observed

Population of *C. pavonana* and *P. xylostella*. Larval populations of *C. pavonana* and *P. xylostella* were observed visually by counting and recording the number of larvae on the broccoli leaves, stems, and heads of the six sample plants per plot. Observations were conducted at weekly intervals from 21–70 DAP. Data were subsequently tabulated and graphed.

Percentage of infested plants. The percentage of infested plants was determined by counting the number of sample plants showing symptoms of damage or the

physical presence of *C. pavonana* and *P. xylostella* larvae. Observations were conducted in the morning at weekly intervals from 21 DAP to 70 DAP. The percentage of attacks can be calculated using the following formula:

$$P = \frac{A}{B} \times 100\%$$

where P: percentage of infested plants (%); A: number of infested plants; and B: number of plants observed.

Insecticide effectiveness level. Based on the efficacy standards, an insecticide is considered effective if its effectiveness value remains above 70% in at least half the observation periods plus one (Directorate of Fertilizers and Pesticides 2012). The effectiveness level of the tested insecticides (%) was calculated using Abbot's formula as follows:

$$EI = \frac{Ca - Ta}{Ca} \times 100\%$$

where EI: effectiveness of the insecticide tested (%); Ca: target pest population in the control plot; and Ta: target pest population in the treatment plot after insecticide application.

Data analysis

Larval population data and the percentage of plants infested by *C. pavonana* and *P. xylostella* were analyzed using analysis of variance (ANOVA). When significant differences were detected, the data were further analyzed using a least significant difference (LSD) test at the 5% significance level using Statistis 8.

RESULTS

Larval populations of *C. pavonana* and *P. xylostella*

Observations of the larval populations of *C. pavonana* and *P. xylostella* consistently showed that untreated (control) populations were higher than those exposed to insecticides. Statistical analysis revealed that the botanical insecticide nanoemulsion formulation—containing a mixture of *P. aduncum* and *C. nardus* hydrosol (INN)—along with the biological insecticide *B. thuringiensis* (Bt) and the synthetic insecticide cypermethrin (IS), each produced population densities that were significantly lower than that of the control. For *C. pavonana* population, these significant differences were observed from 49 DAP onwards through 70 DAP (Table 1). Meanwhile, for *P. xylostella* significant population increase on control were noted earlier, occurring continuously from 28 DAP to 70 DAP (Table 2).

The lowest overall average larval density of *C. pavonana* was recorded in the IS treatment (0.12 larvae per plant), followed by Bt (0.22 larvae per plant) and INN (0.23 larvae per plant). All three treatments

Table 1. Larval population density of *Crocidolomia pavonana* on broccoli under different insecticide treatments

Treatment	Population of <i>C. pavonana</i> larval (Mean ± SD)								Average
	21 DAP	28 DAP	35 DAP	42 DAP	49 DAP	56 DAP	63 DAP	70 DAP	
Control	0.00±0 a	0.09±1.51 a	0.16±2.64 a	0.26±1.51 a	1.40±9.93 a	1.21±6.07 a	1.02±3.48 a	0.69±3.21 a	0.60
INN	0.00±0 a	0.09±1.13 a	0.11±0.98 a	0.19±2.26 a	0.30±2.54 b	0.69±3.18 b	0.59±1.71 ab	0.35±1.21 b	0.23
Bt	0.00±0 a	0.07±0.78 a	0.07±0.78 a	0.11±1.25 a	0.26±1.90 b	0.54±4.07 b	0.61±1.88 b	0.16±0.81 b	0.22
IS	0.00±0 a	0.02±0.37 a	0.00±0 a	0.00±0 a	0.11±0.95 b	0.47±1.86 b	0.14±1.21 bc	0.28±1.38 b	0.12

DAP: day after planting; INN: nanoemulsion insecticide; Bt: *Bacillus thuringiensis* insecticide; IS: synthetic insecticide.

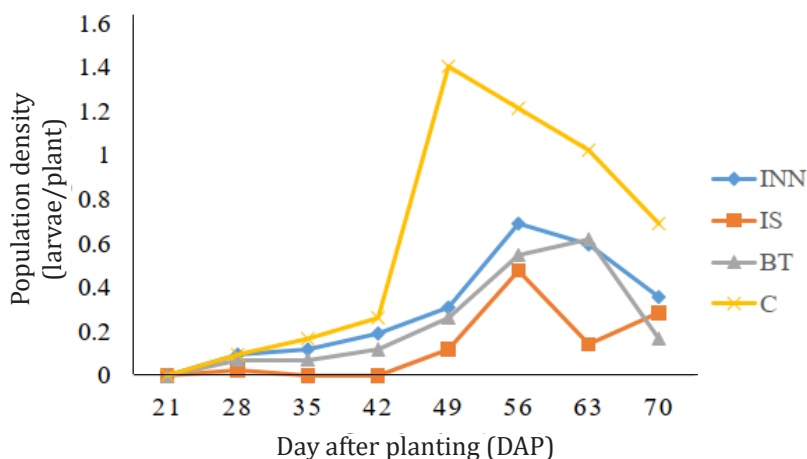
Numbers followed by the same letter within the same column are not significantly different according to the LSD tests at the 5% significance level.

Table 2. Larval population density of *Plutella xylostella* on broccoli under different insecticide treatments

Treatment	Population of larvae <i>P. xylostella</i> (Mean ± SD)								Average
	21 DAP	28 DAP	35 DAP	42 DAP	49 DAP	56 DAP	63 DAP	70 DAP	
Control	0.07±0.78 a	0.61±2.56 a	0.76±3.86 ab	2.80±5.11 a	8.73±4.82 a	8.16±12.71 a	6.61±12.48 a	4.02±10.18 a	3.97
INN	0.16±0.88 a	0.42±2.37 b	0.97±2.79 a	1.19±3.33 b	3.28±6.82 bc	3.04±10.54 b	0.78±2.05 b	0.73±1.90 b	1.32
Bt	0.21±1.88 a	0.59±2.76 b	0.45±1.79 b	1.50±4.72 b	3.52±10.10 b	2.69±5.78 b	1.23±7.11 b	0.28±2.05 b	1.30
IS	0.14±0.89 a	0.21±2.21 b	0.59±1.98 ab	0.97±1.67 b	2.02±7.05 c	2.61±10.95 b	0.38±3.68 b	0.42±2.37 b	0.91

DAP: day after planting; INN: nanoemulsion insecticide; Bt: *Bacillus thuringiensis* insecticide; IS: synthetic insecticide.

Numbers followed by the same letter within the same column are not significantly different according to the LSD tests at the 5% significance level.

**Figure 1.** Population development of the pest *Crocidolomia pavonana* after insecticide treatment. INN: nanoemulsion insecticide; IS: synthetic insecticide; Bt: *Bacillus thuringiensis* insecticide; C: control.

were significantly lower than the control, which had a density of 0.60 larvae per plant (Table 1). Similarly, for *P. xylostella*, the lowest average population density was found in the IS treatment (0.91 larvae per plant), followed by Bt (1.30 larvae per plant) and INN (1.32 larvae per plant), all of which were significantly lower than the control (3.97 larvae per plant) (Table 2).

Throughout the observation period (21 to 70 DAP), larval population densities of both *C. pavonana* and *P. xylostella* fluctuated across all treatments. However, the control consistently maintained the highest population densities than the treated plots.

The population density of *C. pavonana* larvae peaked at 49 DAP, reaching 1.40 larvae per plant in the control group, whereas densities in the insecticide-treated plots

remained significantly lower; 0.30, 0.26, and 0.11 larvae per plant in the INN, Bt, and IS treatments, respectively (Figure 1). Following this peak infestation, *C. pavonana* population densities generally decreased toward the end of the observation period (63 to 70 DAP) across all treatments, though the control group maintained the highest values.

Similarly, the population density of *P. xylostella* larvae reached its peak at 49 DAP with the control group recording 8.73 larvae per plant, while the treated plots remained significantly lower (Figure 2). At 49 DAP, the IS, INN, and Bt treatments 2.02, 3.28, and 3.52 larvae per plant, respectively. The *P. xylostella* larval population densities subsequently declined from 56 to 70 DAP. Throughout the entire observation period, the

control group had a higher population density than any of the insecticide treatments.

Percentage of infested plants

The percentage of infested plants by *C. pavonana* and *P. xylostella* larvae was simultaneously assessed. The results showed that all insecticide treatments (INN, Bt, and IS) significantly reduced plant infestation compared with the control group continuously from 28 to 70 DAP (Table 3). Averaged across all observation periods, the control group demonstrated the highest infestation rate (70.66%). Among the treatments, INN resulted in the lowest overall infestation rate (27.85%), followed by Bt (37.49%) and IS insecticide (38.01%).

Insecticide efficacy

Field evaluations indicated that the IS treatment was the most effective against *C. pavonana*, with an overall average efficacy of 71.49%. In contrast, INN treatment recorded the lowest efficacy among the treated groups at 33.33% with Bt showed intermediate efficacy at 48.54% (Table 4). A similar trend was observed for *P. xylostella*, the IS treatment was again the most effective, with an average efficacy at 73.70%, followed by Bt (60.88%), and INN treatment recorded the lowest average at 58.38% (Table 5). According to national efficacy standards, an insecticide is considered effective if its efficacy (EI) values exceeds 70% in more than half of the observations. The IS treatment met this criterion, exceeding 70% efficacy in five out of the

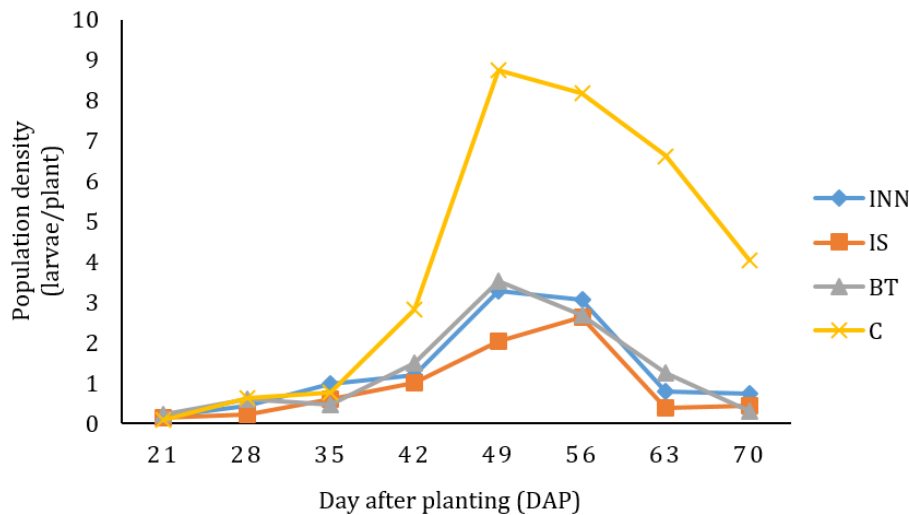


Figure 2. Population development of *Plutella xylostella* after insecticide treatment. INN: nanoemulsion insecticide; IS: synthetic insecticide; Bt: *Bacillus thuringiensis* insecticide; C: control.

Table 3. Percentage of infested plants by *Crociodolomia pavonana* and *Plutella xylostella* simultaneously on broccoli plants after treatment

Treatment	Percentage of infested plants by <i>C. pavonana</i> and <i>P. xylostella</i> (% ± SD)								Average (%)
	21 DAP	28 DAP	35 DAP	42 DAP	49 DAP	56 DAP	63 DAP	70 DAP	
Control	5.71±2.12 a	17.61±4.49 a	43.63±7.76 a	99.04±2.51 a	99.28±1.88 a	100±0.00 a	100±0.00 a	100±0.00 a	70.66
INN	5.47±1.58 a	10.23±1.78 b	28.81±9.09 b	46.68±10.3 b	51.44±8.32 b	52.88±7.14 b	53.59±7.28 b	55.02±7.43 b	27.85
Bt	5.23±1.78 a	9.52±1.85 b	25.95±5.25 b	45.60±5.56 b	52.51±8.45 b	53.22±8.25 b	53.22±8.25 b	54.89±5.93 b	37.49
IS	4.52±1.25 a	7.38±2.32 b	20.95±3.70 b	34.04±5.34 c	34.87±5.71 c	38.93±7.65 c	40.12±7.68 c	42.03±6.42 c	38.01

DAP: day after planting; INN: nanoemulsion insecticide; Bt: *Bacillus thuringiensis* insecticide; IS: synthetic insecticide. Numbers followed by the same letter within the same column are not significantly different according to the LSD tests at the 5% significance level.

Table 4. Field efficacy of insecticide treatments against *Crociodolomia pavonana* larvae on broccoli

Treatment	Insecticide efficacy at each observation period (%)								Average (%)
	21 DAP	28 DAP	35 DAP	42 DAP	49 DAP	56 DAP	63 DAP	70 DAP	
INN	0.00	0.00	28.57	27.27	77.96	43.13	41.46	48.27	33.33
Bt	0.00	25.00	57.14	54.54	81.35	54.90	39.53	75.86	48.54
IS	0.00	75.00	100	100	91.52	60.78	86.04	58.62	71.49

DAP: day after planting; INN: nanoemulsion insecticide; Bt: *Bacillus thuringiensis* insecticide; IS: synthetic insecticide.

Table 5. Field efficacy of insecticide treatments against *Plutella xylostella* larvae on broccoli

Treatment	Insecticide efficacy at each observation period (%)								Average (%)
	21 DAP	28 DAP	35 DAP	42 DAP	49 DAP	56 DAP	63 DAP	70 DAP	
INN	37.60	15.38	57.62	62.19	62.39	62.68	88.12	81.06	58.38
Bt	19.23	63.46	44.06	59.45	59.67	67.05	81.29	92.89	60.88
IS	65.38	51.92	64.44	77.26	76.83	70.55	94.24	89.34	73.70

DAP: day after planting; INN: nanoemulsion insecticide; Bt : *Bacillus thuringiensis* insecticide; IS: synthetic insecticide.

eight observation periods for both *C. pavonana* and *P. xylostella*. Consequently, the synthetic insecticide was deemed effective, whereas both the Bt and INN botanical treatments failed to meet this threshold and were classified as ineffective under these field conditions. These results suggested that the biological and botanical formulations provided only moderate pest suppression under field conditions tested.

DISCUSSION

In general, field observations of *C. pavonana* and *P. xylostella* larval populations, showed that untreated larval populations (control) were consistently higher than those subjected to insecticide treatments. Statistical analysis revealed that the INN, Bt, and IS treatments resulted significantly lower populations compared to the control group from 49 to 70 DAP for *C. pavonana*, and from 28 DAP to 70 DAP for *P. xylostella*.

Throughout the study, the larval populations of both pest species fluctuated significantly. Highest population occurred at 49 DAP, which aligns with findings by Kumarawati et al. (2013), who reported that the highest abundance of *C. pavonana* and *P. xylostella* larval populations on broccoli typically occurs between seven to eight weeks post-planting. Abiotic factors, particularly rainfall, influence these insect population dynamics (Murtiningsih et al. 2023). Low rainfall and intermittent drought periods when the plants were 42 - 56 DAP likely facilitated the increase in larval populations of *C. pavonana* and *P. xylostella*. Conversely, high rainfall intensity during the eighth week contributed to a decline in both pest populations by 63 DAP. Furthermore, larval abundance in the field is positively correlated with food availability, indicating that areas with more food sources tend to support higher larval populations (Kumarawati et al. 2013). At 49 DAP, broccoli plants experience the increase in leaf number. This vegetative growth increases in leaf number correlates with a heightened susceptibility to pest attacks, resulting in a rise in the populations of *C. pavonana* and *P. xylostella* during this observation period. Akter et al. (2025) reported that leaf number in broccoli increases progressively during the vegetative

stage, with observations commonly conducted up to around 49 days after planting (DAP).

The suppression of larval populations of *C. pavonana* and *P. xylostella* across treatments can be attributed to the specific active compounds in each formulation (Kumarawati et al. 2013). Dillapiole, a major active compound in *P. aduncum*, can inhibit the activity of the cytochrome P450 detoxification enzyme, which plays a role in reducing the toxicity of toxic compounds or metabolites in the larval body (Bernard et al. 1995). Previous studies have demonstrated that spiked piper extracts can cause 100% mortality in *C. pavonana* (Syahroni & Prijono 2013) and 98 % mortality in *P. xylostella* (Lina et al. 2018). Although, the major bioactive compounds of *C. nardus*, such as citronellal and geraniol, are primarily present in the essential oil fraction. However, small amounts of these compounds can be carried over into hydrosol during the distillations. These carried-over compounds likely contribute to the overall insecticidal activity of the formulation by disrupting cellular metabolism and respiratory processes in insect (Araújo et al. 2023). Therefore, the combined antifeedant and toxicological effects of *P. aduncum* extract and *C. nardus* hydrosol mixture can also cause a decline in the larval populations of *C. pavonana* and *P. xylostella* (Erlina et al. 2020).

The biological insecticide *B. thuringiensis* (Bt) contains δ -endotoxin crystals that are lethal and can reduce the populations of *C. pavonana* and *P. xylostella*. The toxin bind to specific receptors on midgut epithelial cell, leading to pore formation, cell lysis, and gut paralysis. According to Hariyani et al. (2014), *B. thuringiensis* formulations can suppress the larval populations of *C. pavonana* and *P. xylostella* by > 45%. The disadvantages of the insecticide *B. thuringiensis* are its sensitivity to ultraviolet light and ease of washing; therefore, its active ingredients do not last long in the field (Bahagiawati 2002). In contrast, the synthetic insecticides with the active ingredient cypermethrin (IS) are known to have high acute toxicity and persistence, and can rapidly kill *C. pavonana* and *P. xylostella* larvae, thereby reducing pest infestation. Cypermethrin is a synthetic pyrethroid insecticide that acts on the insect

nervous system by modifying the gating kinetics of voltage-gated sodium channels, resulting in prolonged depolarization, repetitive nerve firing, paralysis, and ultimately insect death (Field et al. 2017).

Application of INN, Bt and suppressed the attack of *C. pavonana* and *P. xylostella*. The results showed that the percentage of plants infested with *C. pavonana* and *P. xylostella* the same time, and broccoli plants were treated with a lower insecticide than those without insecticide treatment. The percentage of infested plants is closely related to the number of *C. pavonana* and *P. xylostella* larval populations; the higher the number of larvae, the higher the percentage of infested plants will increase (Hasnah & Nasril 2009). The percentage of infested plants increased significantly at 42 DAP and peaked at 70 DAP because of the high *C. pavonana* and *P. xylostella* populations. This aligns with Kumarawati et al. (2013), who stated that the highest percentage of plants infested with *C. pavonana* and *P. xylostella* larvae occurred in the tenth week after planting.

Based on the efficacy standard, it was found that the plant-based insecticide formulation of a nanoemulsion mixed with *P. aduncum* extract and *C. nardus* hydrosol was less effective in controlling *C. pavonana* and *P. xylostella* because it had an effectiveness percentage below 70% for five times in eight observations. The ineffectiveness of botanical insecticides is caused by the active ingredients contained in the insecticide, which cannot last long during field application because they are quickly degraded by sunlight and easily washed by rain. According to Badan Meteorologi, Klimatologi dan Geofisika (BMKG) data, the Gumanti Valley experienced rainfall amounts of 513 mm, 344 mm, and 463 mm during the planting period from July to October 2023. This indicates that these months fall into the category of wet months with significant rainfall. Therefore, the application of natural insecticides should be more frequent to increase the effectiveness of the active ingredient (Kardinan & Suriati 2012)

CONCLUSION

The efficacy evaluation of a botanical insecticide nanoemulsion formulation of *P. aduncum* extract and *C. nardus* hydrosol demonstrated comparable activity to *B. thuringiensis* insecticide, although its control of *C. pavonana* and *P. xylostella* on broccoli plant was still lower than that of synthetic insecticides.

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