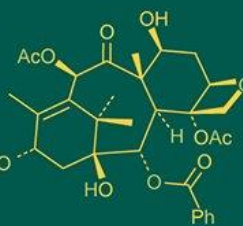
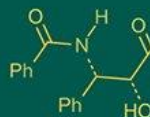


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Comparative efficacy and LT₅₀ analysis of biopesticides and botanical extracts against *Helicoverpa armigera* (Hubner) on chickpea under laboratory conditions

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Abstract

Helicoverpa armigera (Hubner) is a major insect pest of chickpea causing heavy yield loss due to considerable feeding damage. The present study was designed to calculate the median lethal time (LT₅₀) and evaluate the efficacy of biopesticides and botanical extracts against *H. armigera* under laboratory conditions. The experiment was carried out in Completely Randomised Design (CRD) with nine treatments with three replications. The treatments were *Beauveria bassiana* 1.15 WP, Azadirachtin 10000 ppm, Neem Seed Kernel Extract (NSKE) 5%, custard apple leaf extract, Lantana leaf extract, Ipomoea leaf extract, Karanj leaf extract, Datura leaf extract and untreated control. Larval mortality was measured at 24, 48 and 72 hours after treatment by using a leaf dip bioassay. Significant differences among treatments were observed ($P = 0.05$). Maximum larval mortality of 53.33, 86.67 and 100 per cent were recorded at 24, 48 and 72 hours post-treatment respectively with NSKE 5%. Azadirachtin at 10000 ppm resulted in a mortality of 33.33, 66.67 and 86.67 per cent respectively over the same time period. The LT₅₀ data demonstrated that NSKE 5% had a fast insecticidal action with LT₅₀ of 22 hours while LT₅₀ of Azadirachtin at 10000 ppm was 34 hours. Of the three plant extracts tested, Datura leaf extract showed moderate activity with LT₅₀ of 43 hours while Lantana and Ipomoea leaf extracts did not cause 50 per cent mortality of the insects within the period of observation. The results revealed that NSKE 5% and Azadirachtin 10000 ppm were extremely efficient against *H. armigera* and can be used as eco-friendly components in integrated pest management methods for chickpea cultivation.

Keywords: *Helicoverpa armigera*, chickpea, biopesticides, botanical extracts, neem seed kernel extract, azadirachtin, LT₅₀

1. Introduction

Chickpea (*Cicer arietinum* L.) also known as Bengal gram or gram is one of the most important pulse crops grown worldwide and a vital source of dietary protein, carbs, vitamins and minerals especially in developing nations. Chickpea is the third most significant food legume in the world with a production of over 16.5 million tonnes in 2023, mostly driven by India which provides nearly 75% of this amount. India grows chickpeas on 9.9-10 million hectares and produces more than 11 million tonnes, which is vital for the country's food security (FAOSTAT, 2024). Chickpea production in Gujarat has grown significantly, from 6.36 lakh tonnes in 2019-20 to almost 15.63 lakh tonnes in 2024-25, reflecting its growing importance in the state's agricultural sector (Government of Gujarat, 2025). Several biotic and abiotic factors limit the production of chickpea despite its economic importance. Insect pests are considered as one of the key yield limiting elements among the biotic stressors. More than 60 insect species are known to infest chickpea at different crop growth stages. Among these, gram pod borer, *Helicoverpa armigera* is considered as the most devastating insect pest producing substantial economic losses in the chickpea growing regions of the world. *Helicoverpa armigera* is a global, extremely polyphagous insect pest that attacks more than 200 cultivated and wild plant species. The pest infects many economically significant crops which include chickpea, pigeonpea, tomato, chilli, cotton, maize, sunflower, tobacco, okra and many vegetable crops (Dhillon *et al.*, 2012; Ahmad *et al.*, 2014) [6, 3]. In chickpea, larvae feed on fragile leaves, blooms and developing

Pods causing direct damage to the reproductive portions of the plant. During its development stage a single larva can kill a significant number of pods and cause a major drop in production. Yield losses under severe infestation may vary from 30 to 90 per cent depending on pest population, crop stage and environmental circumstances. Thus, the insect is considered to be a major limitation to profitable chickpea production. Synthetic chemical insecticides have been the mainstay for managing *H. armigera* because of their quick action and availability. Chemical pesticides are effective for short-term insect control but their overuse leads to resistance in pests like *H. armigera*, environmental pollution, loss of natural predators, and health risks. This highlights the need for sustainable pest management alternatives. Biopesticides and botanical insecticides, particularly neem products containing bioactive agents such as azadirachtin, have emerged as eco-friendly solutions in integrated pest management (IPM), effectively reducing the feeding and reproduction of pests including *H. armigera*. (Srivastava *et al.*, 2011; Ahmad *et al.*, 2015; Kilani-Morakchi *et al.*, 2021) [13, 2, 9]. Considering the economic importance of chickpea and the growing need for eco-friendly pest management strategies, the current investigation was performed to evaluate the comparative effectiveness of selected biopesticides and botanical extracts against *Helicoverpa armigera* under laboratory conditions and to determine their median lethal time (LT₅₀) values.

2. Materials and Methods

2.1 Experimental Site

The laboratory experiment was carried out in the Department of Entomology, Sardarkrushinagar Dantiwada Agricultural University, Sardarkrushinagar, Gujarat, India during Rabi season of 2025-2026. The present study was conducted to evaluate the efficiency of certain biopesticides and botanical extracts against gram pod borer, *Helicoverpa armigera* under laboratory conditions.

2.2 Experimental Design and Treatments

The experiment was laid out in Completely Randomized Design (CRD) with nine treatments and three replications. The treatments consisted of different biopesticides and botanical extracts along with an untreated control. Details of the treatments are presented in Table 1.

Table 1: Treatments evaluated against *Helicoverpa armigera*

Treatment	Details	Concentration (%)	Dose (ml L ⁻¹ water)
T ₁	Beauveria bassiana 1.15 WP (1 × 10 ⁸ cfu g ⁻¹)	0.0046	3
T ₂	Azadirachtin 10000 ppm	0.002	3
T ₃	Neem Seed Kernel Extract (NSKE)	5.0	3
T ₄	Custard apple leaf extract	10.0	3
T ₅	Lantana leaf extract	10.0	3
T ₆	Ipomoea leaf extract	10.0	3
T ₇	Karanj leaf extract	10.0	3
T ₈	Datura leaf extract	10.0	3
T ₉	Untreated control	-	-

2.3 Preparation of Botanical Extracts

Fresh and healthy plant materials of custard apple (*Annona squamosa*), lantana (*Lantana camara*), *Ipomoea* spp., karanj (*Pongamia pinnata*), and datura (*Datura stramonium*) were collected and washed thoroughly with distilled water. The

leaves were shade-dried and crushed using a sterilized grinder. The extracts were prepared by homogenizing the required quantity of plant material in distilled water and filtering through muslin cloth followed by Whatman No. 1 filter paper to obtain the desired concentration.

2.4 Insect Culture

Helicoverpa armigera larvae were collected from chickpea fields and reared in laboratory conditions (Fig.1). Bioassay studies were conducted with healthy and active larvae. Third instar larvae were maintained at 27 ± 2 °C and 65 ± 5% RH. All treatments were carried out with uniform sized larvae to minimize experimental variation.

2.5 Bioassay Procedure

The efficacy of biopesticides and botanical extracts was evaluated by the leaf-dip method. Fresh chickpea leaves were dipped individually in the respective treatment solutions for approximately 10 seconds and allowed to air dry under room conditions. Similar leaves dipped in distilled water served as untreated control. The treated leaves were placed in sterile Petri dishes lined with moist filter paper. Each treatment consisted of ten Petri dishes per replication, with one larva per Petri dish (Fig. 2). Larval mortality was recorded at 24, 48 and 72 hours after treatment. Mortality was assessed based on the inability of larvae to show coordinated movement upon gentle probing. Leaf-dip bioassays are widely employed for evaluating insecticidal toxicity against *H. armigera* under laboratory conditions.

2.6 Recording of Observations

2.6.1 Larval mortality

Larval mortality was recorded at 24, 48, and 72 hours after treatment and expressed as percentage mortality using the following formula:

$$\text{Per cent mortality (\%)} = (\text{Number of dead larvae} / \text{Total number of larvae}) \times 100$$

2.6.2 LT₅₀ Analysis

The median lethal time (LT₅₀) values were estimated through probit regression analysis based on cumulative mortality data following the procedure described by Finney (1971) [7]. Probit analysis is commonly used for estimating lethal response parameters in insect toxicological studies.

2.7 Statistical Analysis

The mortality data were subjected to analysis of variance (ANOVA) appropriate for Completely Randomized Design (CRD). Percentage mortality values were transformed using angular (arc-sine) transformation before statistical analysis. Treatment means were compared at the 5 per cent level of significance. Standard error of mean [S.E.m. (±)], critical difference (CD) at 5 per cent, and coefficient of variation (CV%) were calculated to assess treatment effects.

3. Results

3.1 Effect of Biopesticides and Botanical Extracts on Larval Mortality of *Helicoverpa armigera*

Significant differences were identified among the investigated biopesticides and botanical extracts against *Helicoverpa armigera* under laboratory conditions (Table 2; Fig. 3). Larval mortality rose progressively with exposure period from 24 to 72 hours in all treatments. At 24 hours

after treatment, Neem Seed Kernel Extract (NSKE) 5% (T₃) recorded the highest larval mortality (53.33%), which was considerably superior to all other treatments. Azadirachtin 10000 ppm (T₂) came second with 33.33 per cent mortality, followed by Karanj leaf extract (T₇) and Datura leaf extract (T₈), each recording 26.67 per cent mortality. *Beauveria bassiana* (T₁) and Custard apple leaf extract (T₄) displayed comparatively reduced mortality of 13.33 per cent, but Lantana leaf extract (T₅), Ipomoea leaf extract (T₆), and untreated control (T₉) showed no mortality within the first 24 hours. At 48 hours following treatment, NSKE 5% continued to display superior efficacy with 86.67 per cent larval mortality, followed by Azadirachtin 10000 ppm (66.67%). Datura leaf extract (53.33%) and both *Beauveria bassiana* and Karanj leaf extract (46.67%) demonstrated moderate effectiveness. Custard apple leaf extract resulted in 40.00 per cent death, while Lantana and Ipomoea extracts showed only 20.00 per cent mortality.

At 72 hours following treatment, total larval death (100%) was seen in NSKE 5% (T₃), making it the most effective treatment among all evaluated biopesticides and botanicals. Azadirachtin 10000 ppm (T₂) recorded 86.67 per cent mortality and remained statistically at par with the best treatment. Datura leaf extract (T₈) yielded 73.33 per cent mortality, followed by *Beauveria bassiana* (T₁) and Karanj

leaf extract (T₇), each registering 66.67 per cent mortality. Custard apple leaf extract resulted in 53.33 per cent death, whilst Lantana and Ipomoea leaf extracts were considerably less effective, recording 40.00 per cent mortality. No mortality was found in the untreated control during the testing period.

3.2 LT₅₀ Analysis

The median lethal time (LT₅₀) results further confirmed the superiority of NSKE and Azadirachtin treatments. Among all treatments, NSKE 5% (T₃) displayed the lowest LT₅₀ value of 22 hours, indicating quick toxic activity against *H. armigera* larvae. Azadirachtin 10000 ppm (T₂) recorded the second lowest LT₅₀ value of 34 hours. Datura leaf extract (T₈) exhibited an LT₅₀ value of 43 hours, whereas Karanj leaf extract (T₇) and *Beauveria bassiana* (T₁) recorded LT₅₀ values of 50 and 51 hours, respectively. Custard apple leaf extract (T₄) required 59 hours to produce 50 per cent mortality. In contrast, Lantana leaf extract (T₅) and Ipomoea leaf extract (T₆) failed to reach 50 per cent mortality during the 72-hour observation period and hence exhibited LT₅₀ values greater than 72 hours. Lower LT₅₀ values suggest faster insecticidal activity and greater efficiency of a treatment (Fig. 4).

Table 2. Effect of biopesticides and botanical extracts on larval mortality and LT₅₀ values of *Helicoverpa armigera* under laboratory conditions

Treatment	24 HAT (%)	48 HAT (%)	72 HAT (%)	LT ₅₀ (h)
T ₁	13.33 (21.42)	46.67 (43.09)	66.67 (54.74)	51
T ₂	33.33 (35.26)	66.67 (54.74)	86.67 (68.58)	34
T ₃	53.33 (46.91)	86.67 (68.58)	100.00 (90.00)	22
T ₄	13.33 (21.42)	40.00 (39.23)	53.33 (46.91)	59
T ₅	0.00 (0.00)	20.00 (26.57)	40.00 (39.23)	>72
T ₆	0.00 (0.00)	20.00 (26.57)	40.00 (39.23)	>72
T ₇	26.67 (31.09)	46.67 (43.09)	66.67 (54.74)	50
T ₈	26.67 (31.09)	53.33 (46.91)	73.33 (58.91)	43
T ₉	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	-
S.E.m. ±	1.50	2.29	1.70	-
CD (P = 0.05)	4.48	6.86	5.09	-
CV (%)	13.87	11.35	6.50	-

*HAT = Hours after treatment. Values in parentheses are angular transformed values used for statistical analysis.



Fig 1: Typical symptom of *Helicoverpa armigera*



Fig 2: Leaf-dip bioassay of *Helicoverpa armigera* larvae on chickpea leaves under laboratory conditions: (A) larvae before feeding on treated leaves and (B) larvae after feeding on treated leaves

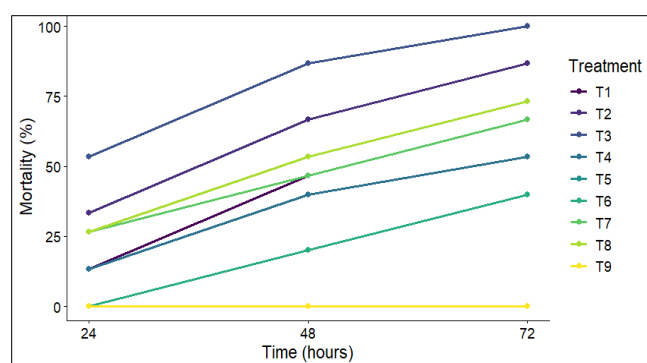


Fig 3: Time mortality response of treatment against

Helicoverpa armigera

The quick action observed in NSKE and Azadirachtin treatments underlines their considerable promise as eco-friendly alternatives to synthetic insecticides for the management of *H. armigera*. The considerably larger LT₅₀ values obtained in other plant extracts indicate slower toxic action and lesser potency under laboratory conditions. Overall, the results revealed that NSKE 5% was the most

effective treatment in terms of both larval mortality and speed of action, followed by Azadirachtin 10000 ppm.

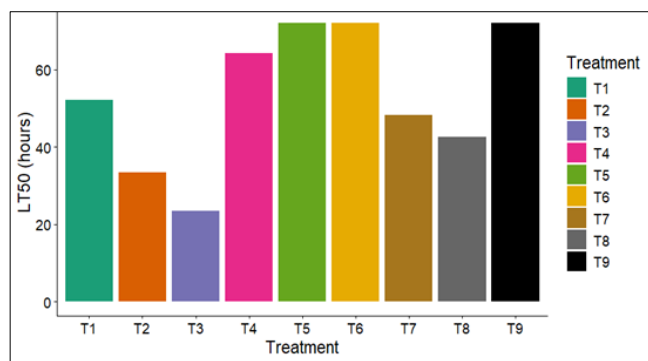


Fig 4: Probit based LT₅₀ comparison of treatments

4. Discussion

In the present study, the biopesticides and botanical extracts produced significant mortality of *Helicoverpa armigera* larvae in the laboratory. Among the treatments studied, Neem Seed Kernel Extract (NSKE) 5% recorded the highest larval mortality and lowest LT₅₀ value followed by Azadirachtin 10000 ppm. The improved success of neem-based treatments documented in the present study could be related to the presence of physiologically active chemicals like azadirachtin, salannin, nimbin and meliantriol which interfere with eating, growing, moulting, digesting and reproduction of insects. The substantially higher mortality recorded in NSKE treated larvae indicates the powerful insecticidal and antifeedant properties of neem derived compounds. Neem-based formulations are known to disrupt the endocrine balance and inhibit normal growth of larvae, causing mortality. Azadirachtin has been described as a potent insect growth regulator and feeding deterrent against many lepidopteran pests including *H. armigera*. Studies have shown that azadirachtin affects several physiological and biochemical pathways of *H. armigera* and reduces survival, development and reproductive potential. The present results are in close agreement with previous reports of substantial mortality of *H. armigera* larvae when exposed to neem-based products (Dindor and Panickar, 2020; Sai *et al.*, 2021; Panickar *et al.*, 2025) [11, 12]. Wakil *et al.* have demonstrated that neem formulations generated considerable larval mortality and suppressed the larval development of *H. armigera* under laboratory conditions. Likewise, Dangi *et al.* (2019) [5] also showed high mortality of third instar *H. armigera* larvae treated with neem seed kernel extract and neem-based products further confirming the efficacy of neem-derived compounds against pod borer populations. Earlier investigations in chickpea and other pulse crops also support the efficacy of NSKE as reported in the present study. Several researchers reported that NSKE significantly reduced larval population, pod damage and feeding activity of *H. armigera* and can be successfully incorporated in integrated pest management techniques. Neem based formulations are regarded safer for the environment than conventional insecticides with their minimal mammalian toxicity and less effect on beneficial species. The second most successful treatment was azadirachtin 10000 ppm. The larvae treated with Azadirachtin showed significant mortality and comparatively low LT₅₀ value due to its effective antifeedant, growth regulating and toxic activities.

Azadirachtin has been found to adversely affect larval growth, pupation, adult emergence, fertility and longevity of *H. armigera*. Other studies have shown that azadirachtin can also disrupt the activity of digestive enzymes and interfere with the hormonal regulation required for the development of the insect. Among other botanical therapies, Datura leaf extract and Karanj leaf extract demonstrated moderate efficacy against *H. armigera* larvae. The insecticidal activity may be attributed to presence of alkaloids, flavonoids, phenolics and other secondary metabolites which may have deleterious influence on feeding behaviour and physiological activities of insects. However, the performance was lesser than the neem-based therapies which shows it is less toxic or slower in its impact on the test insect. The LT₅₀ statistics also demonstrated the efficacy of the drug. NSKE had the lowest LT₅₀ value, which indicated that it was fast in its toxic action and higher in its insecticidal efficiency. Lower LT₅₀ often implies a faster mortality response and better bioefficacy of a drug. The somewhat higher values of LT₅₀ obtained for the other botanical extracts are observed to show a delayed insecticidal activity and worse efficiency under laboratory conditions. Earlier studies have also revealed similar conclusions that neem-based formulations show faster mortality response than other plant derived extracts. The growing issue of pesticide resistance in *H. armigera* population requires the development of safer and sustainable pest management strategies. Many synthetic insecticides are losing their long-term effectiveness due to resistance developed by *H. armigera*. Neem based biopesticides offer good potential in such condition, as neem has many mechanisms of action and so development of resistance is less likely. The present findings in general demonstrated that the NSKE 5% and Azadirachtin 10000 ppm had good insecticidal efficacy against *H. armigera* and can be employed as effective eco-friendly alternatives to the conventional chemical pesticides. Natural enemies can be incorporated into integrated pest management strategies for sustainable chickpea production, reducing environmental problems from the overuse of pesticides. Therefore, NSKE 5% may be recommended as a promising eco-friendly component in integrated management programmes against *H. armigera* in chickpea.

5. Conclusion

The present study proved the potential of biopesticides and botanical extracts for the management of *Helicoverpa armigera* under lab conditions. Among those tested, Neem Seed Kernel Extract (NSKE) 5% was the most effective having the maximum larvae mortality and the lowest LT₅₀ value showing quick and strong insecticidal activity. Azadirachtin 10000 ppm was the second most effective therapy and was comparable in decreasing larval survival. Moderate efficiency was observed in Datura leaf extract, Karanj leaf extract and *Beauveria bassiana* however Lantana and Ipomoea leaf extracts were less effective compared. The results suggested that neem-based solutions are promising as ecofriendly alternatives to traditional chemical pesticides for the control of *H. armigera*. Their use in integrated pest management programmes can aid to achieving sustainable chickpea production by minimising chemical residues, environmental contamination and the risk of insecticide resistance. Therefore, NSKE 5% may be recommended as a promising eco-friendly component in

integrated management programmes against *H. armigera* in chickpea. However, a further study in field circumstances is needed to corroborate the laboratory results and to assess their practical relevance in the large-scale chickpea production.

Declarations

Data Availability

All data generated or analysed during this study are included in this published article.

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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References

1. Abedi Z, Saber M, Vojoudi S, Mahdavi V, Parsaeyan E. Acute, sublethal, and combination effects of azadirachtin and *Bacillus thuringiensis* on *Helicoverpa armigera*. J Insect Sci. 2014;14(30):1-9. doi:10.1093/jisesa/ieu017.
2. Ahmad S, Ansari MS, Muslim M. Toxic effects of neem-based insecticides on the fitness of *Helicoverpa armigera* (Hübner). Crop Prot. 2015;68:72-78. doi:10.1016/j.cropro.2014.11.003.
3. Ahmad S, Ansari MS, Husain M. Effect of insecticides on the management of *Helicoverpa armigera* in chickpea agro-ecosystem. Ann Plant Prot Sci. 2014;22(1):107-111.
4. Chaudhary S, Kanwar RK, Sehgal A, Cahill DM, Barrow CJ, Sehgal R, et al. Progress on *Azadirachta indica*-based biopesticides in replacing synthetic toxic pesticides. Front Plant Sci. 2017;8:610. doi:10.3389/fpls.2017.00610.
5. Dangi UK, Meena BL, Meena BS, Indoria D. Comparative impact study of neem (*Azadirachta indica* A. Juss) products versus chemical insecticides on mortality of *Helicoverpa armigera*. Int J Curr Microbiol Appl Sci. 2017;6(11):465-468.
6. Dhillon MK, Pampapathy G, Wadaaskar RM, Sharma HC. Impact of Bt transgenic cotton and insecticides on target and non-target insect pests, natural enemies and seed cotton yield in India. Indian J Agric Sci. 2012;82(3):248-254.
7. Finney DJ. Probit Analysis. 3rd ed. Cambridge: Cambridge University Press; 1971.
8. Jagtap SR, Patil PD, More DG. Evaluation of microbials and botanicals against *Helicoverpa armigera* (Hübner) infesting chickpea. J Entomol Zool Stud. 2020;8(5):1452-1457.
9. Kilani-Morakchi S, Aribi N, Soltani N. Azadirachtin-based insecticides: overview, risk assessments, and future directions. Front Agron. 2021;3:676208. doi:10.3389/fagro.2021.676208.
10. Kumar A, Singh R, Cheema HK. Novel insecticides for the management of pod borer complex in pigeonpea crop. Indian J Entomol. 2020;82(4):793-798.
11. Panickar BK, Patel CK, Chaudhary M. Eco-friendly management of pod borer (*Helicoverpa armigera*) in organic chickpea. J Food Legumes. 2025;38(1):158-162.
12. Sai KP, Tayde AR, Reddy RD, Srivastava VK. Management of gram pod borer, *Helicoverpa armigera* (Hübner) in chickpea using biopesticides and their combinations. J Entomol Zool Stud. 2021;9(1):561-563.
13. Srivastava CN, Lalit M, Sharma P, Prejwilita M. A review on prospective of synergistic approach in insect pest management. J Entomol Res. 2011;35:255-266.