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Effect of different solvent extracts of botanicals on larval weight gain of diamondback moth, *Plutella xylostella* (L.): A comparative evaluation

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Abstract
The diamondback moth (DBM), *Plutella xylostella* (L.) (Lepidoptera: Plutellidae) is one of the most destructive pests of cruciferous crops worldwide, with management complicated by its rapid development of resistance to synthetic insecticides. Botanical extracts offer an eco-friendly alternative for pest management and the choice of extraction solvent can strongly influence the bioactivity of the resulting extract. In the present study, extracts of six botanicals *viz.*, *Azadirachta indica*, *Nerium oleander*, *Plumeria rubra*, *Solanum grandiflora*, *Swietenia macrophylla* and *Tagetes erecta* were prepared in three solvents of differing polarity (methanol, ethyl acetate and hexane) and evaluated for their effect on larval weight gain of *P. xylostella* over a 5–12 day feeding period, alongside solvent check and untreated check controls. Across all three solvent systems, larvae in the treated groups consistently showed lower weight gain than those in the solvent check and untreated check, with the treated check recording the least weight gain throughout the observation period. Among the botanicals, extracts of *T. erecta*, *S. grandiflora* and *S. macrophylla* were associated with comparatively greater suppression of larval weight gain than *A. indica*, *N. oleander* and *P. rubra*. Solvent polarity also influenced efficacy with methanol and ethyl acetate extracts generally producing stronger growth-suppressive effects than hexane extracts. These findings suggest that polar and semi-polar solvent extracts of selected botanicals hold promise as components of an integrated, eco-friendly management strategy for DBM.

Keywords: *Plutella xylostella*, botanical extracts, larval weight gain, methanol, ethyl acetate, hexane, solvent polarity

Introduction
The diamondback moth, *Plutella xylostella* (L.) (Lepidoptera: Plutellidae), is widely regarded as the most destructive insect pest of cruciferous vegetables worldwide, inflicting annual management costs and yield losses estimated in the billions of US dollars (Sarfraz *et al*. 2005; Zalucki *et al*. 2012) [12, 15]. Its short generation time, high fecundity, and well-documented ability to develop resistance to virtually every class of synthetic insecticide, including *Bacillus thuringiensis* toxins (Zhao *et al*. 2006 [16]; Pu *et al*. 2010), have rendered its management increasingly challenging and have spurred the search for alternative, resistance-breaking control strategies.
Plant-derived bioactive compounds offer a promising alternative to synthetic insecticides owing to their generally lower mammalian toxicity, faster environmental degradation, and multiple modes of action (Bowers 1992 [1]; Mohan *et al*. 2005; Sharma and Srivastava 2005; Pavela 2016) [7, 8], all of which reduce the likelihood of resistance development. Botanicals may act as antifeedants, growth regulators, oviposition deterrents, or direct toxicants, often disrupting larval nutrition and moulting rather than causing acute mortality alone. The polarity of the extraction solvent governs the class of secondary metabolites recovered: hexane selectively isolates non-polar constituents such as terpenoids, fatty acid derivatives, and other lipophilic compounds, whereas ethyl acetate, being of intermediate polarity, additionally recovers moderately polar constituents such as flavonoids and coumarins all of which may contribute to insect growth-regulatory or toxic activity.
The present study was undertaken to comparatively evaluate the effect of methanol, ethyl acetate and hexane extracts of six botanicals *Azadirachta indica*, *Nerium oleander*, *Plumeria rubra*, *Solanum grandiflora*, *Swietenia macrophylla* and *Tagetes erecta* on the larval weight gain of *P. xylostella*, with the objective of identifying the most effective plant solvent combination(s) for further development as a botanical-based larvicidal or growth-inhibitory agent.

Materials and Methods
1. Insect culture
A laboratory culture of *P. xylostella* was maintained on fresh mustard seedlings under controlled laboratory conditions. Healthy second-instar larvae were used for all bioassays.

2. Plant material and extraction
Six botanicals, namely neem (*Azadirachta indica*), oleander (*Nerium oleander*), frangipani (*Plumeria rubra*), hummingbird tree (*Sesbania grandiflora*), Honduran mahogany (*Swietenia macrophylla*) and marigold (*Tagetes erecta*), were selected based on the results of preliminary screening. Unripe fruits of each plant were collected separately (1 kg), shade-dried for 15–20 days until completely brittle and ground using a mechanical blender.

The ground material was passed through a 40-mesh sieve to obtain a uniform fine powder. The powdered samples were stored in amber-coloured bottles under dry conditions and protected from direct light until further use.

The powdered plant materials were extracted separately using methanol, ethyl acetate and hexane. 10 g of powdered plant material was placed in a cellulose thimble and subjected to continuous hot extraction using a Soxhlet apparatus, maintaining the extraction temperature below the boiling point of the solvent, until the solvent in the extraction chamber became visibly clear. The extracts obtained with each solvent were separately filtered through a Büchner funnel using Whatman No. 1 filter paper. The respective solvents were removed under reduced pressure using a rotary evaporator at 40–45 °C. The resulting crude extracts were collected separately, weighed and stored in amber-coloured vials under appropriate conditions until further bioassay and chemical analysis. The percentage recovery of each extract was calculated based on the initial weight of the powdered plant material.

3. Preparation of test concentrations and bioassay method

Test solutions of the three solvent extracts were prepared at 5 per cent concentration using 0.1% Triton X-100 as a surfactant. Fresh, untreated cabbage leaves were washed with distilled water and air-dried for 30 minutes. Leaf discs of 5 cm diameter were cut and dipped in the respective 5 per cent test solution for approximately 30 seconds to ensure uniform deposition of the active ingredient (Ingle *et al*., 2017) [3]. Treated discs were held in a slanted position for about two minutes in a tray to drain excess solution, and then air-dried for approximately 30 minutes at room temperature before use.

Second-instar larvae of *P. xylostella*, pre-starved for 2 hours, were released on to each treated leaf disc at 10 larvae per replication, within a plastic container lined with moist filter paper to maintain humidity. The treated check consisted of azadirachtin 1% EC at 2 ml/l, applied by the same leaf-dip procedure; solvent check discs were dipped in the respective carrier solution (0.1% Triton X-100 in distilled water) without extract, and untreated check discs received no treatment. The solvent extracts were evaluated in a Completely Randomized Design (CRD) replicated three times. Larval weight gain was recorded till pupation using an electronic balance.

4. Statistical analysis

Data on larval weight gain were subjected to square root transformation prior to analysis of variance (ANOVA) to normalize variances and satisfy homogeneity assumptions. Treatment means within each trial were compared using Duncan's Multiple Range Test (DMRT) at $p \leq 0.05$.

Results

1. Methanol

Larval weight gain in the methanol extract bioassay increased progressively from Day 5 to Day 12 across all treatments, but the rate and magnitude of increase differed markedly between the botanical extracts and the controls. The solvent check and untreated check showed the steepest and highest overall weight gain, exceeding 1.4 g by Day 10–11, indicating unimpeded larval development in the absence of bioactive extract. Among the botanical treatments, *P. rubra* and *N. oleander* permitted comparatively higher larval weight gain than the other extracts, while *S. grandiflora*, *S. macrophylla*, and *T. erecta* extracts were associated with the lowest weight gain, remaining below 0.6 g throughout the observation period. The treated check (standard insecticide) consistently recorded the lowest larval weight gain of all treatments, confirming effective suppression of larval growth (Fig. 1).

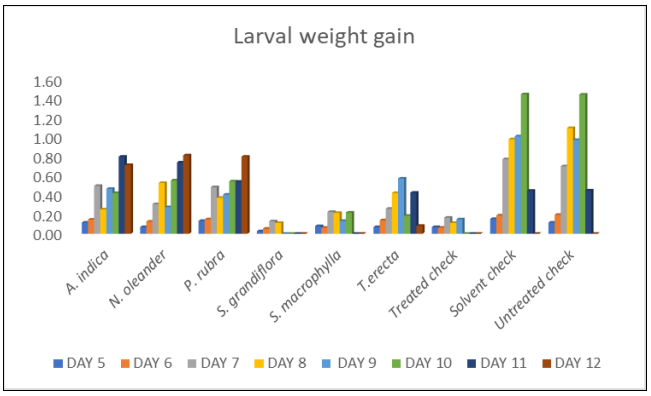

Fig 1: Effect of methanol extract on larval weight gain of *P. xylostella*

2. Ethyl acetate

A broadly similar pattern was observed with ethyl acetate extracts, although overall weight gain values were lower than in the methanol series, with the solvent check and untreated check peaking around 1.0 g by Day 7–8 before gradually declining toward Day 12. Among the botanicals, *A. indica* and *P. rubra* extracts allowed relatively higher larval weight gain, particularly by Day 12, whereas *S. grandiflora* and *S. macrophylla* extracts strongly suppressed larval weight gain, keeping values close to or below 0.4 g across most of the assessment period. The treated check again showed the lowest and most stable (near-flat) weight gain trend across all days (Fig. 2).

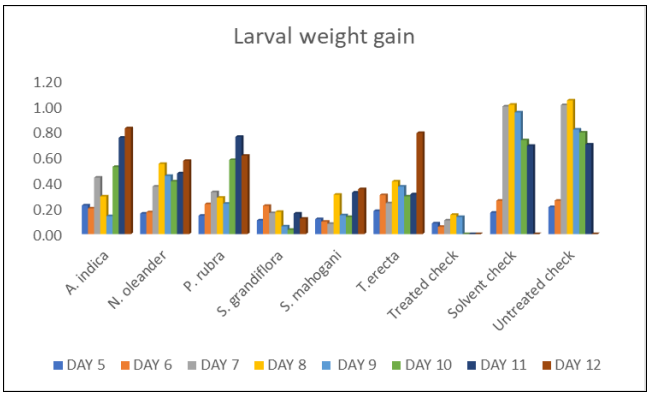

Fig 2: Effect of ethyl acetate extract on larval weight gain of *P. xylostella*

3. Hexane

In the hexane extract bioassay, the solvent check and untreated check exhibited the highest larval weight gain among all three solvent systems, reaching approximately 1.3 g by Day 7–8. Botanical extracts in hexane generally produced an intermediate suppressive effect: *P. rubra* and *A. indica* extracts permitted moderate larval weight gain (peaking around 0.6–0.9 g), while *S. grandiflora* and *S. macrophylla* extracts again showed the strongest

suppression, with weight gain remaining below 0.4 g for most of the study period. As in the other two solvent systems, the treated check maintained the lowest larval weight gain throughout.

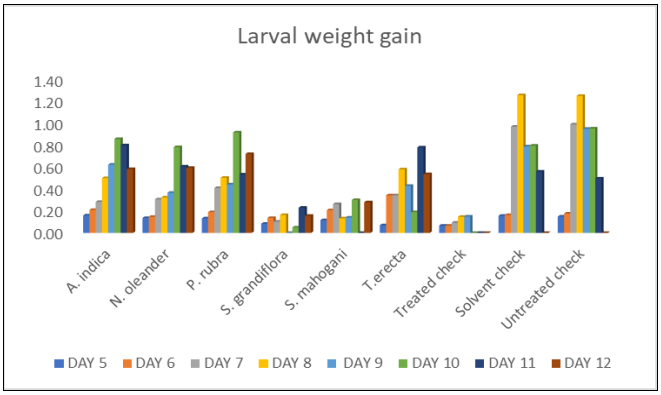

Fig 3: Effect of hexane extract on larval weight gain of *P. xylostella*

4. Cross-solvent comparison

Across all three solvents, a consistent ranking pattern emerged: Treated check < Botanical extracts < Solvent check ≈ Untreated check. Among the botanicals, *S. grandiflora*, *S. macrophylla* and *T. erecta* extracts were the most consistently effective in suppressing larval weight gain, regardless of solvent, whereas *P. rubra*, *N. oleander*, and *A. indica* extracts were comparatively less effective. With respect to solvent polarity, methanol and ethyl acetate extracts generally exerted a stronger growth-suppressive effect than hexane extracts, suggesting that the bioactive constituents responsible for growth inhibition in these plants are predominantly polar to semi-polar in nature.

Discussion

The present findings, wherein *S. grandiflora* and *S. macrophylla* extracts caused significant reduction in larval weight gain, agree with earlier reports establishing that botanical pesticides adversely affect insect growth and development. Talukder (2006) [14] reported that botanical pesticides diminish the weight of larvae, pupae, and adults while prolonging developmental phases, and Koul *et al*. (2008) and Shaalan *et al*. (2005) [4, 13] similarly documented reduced larval/pupal survival and disrupted development following exposure to plant-derived extracts. The efficacy of *S. grandiflora* corroborates Sangavi and Edward (2017) [11], who reported that ethyl acetate leaf extract of *S. grandiflora* at 0.6% reduced adult emergence to 45% against 100% in the control, and Meenambigai (2019) [5], who recorded similar insecticidal activity of this species. Likewise, the growth-inhibitory effects of *S. macrophylla* align with Dinesh-Kumar *et al*. (2018) [2], who observed reduced adult longevity with methanolic leaf extract, and Safraz *et al*. (2005), who reported an insect-static effect of *S. macrophylla* twig extracts on *Spodoptera frugiperda* metamorphosis. These effects have been linked to fatty acid derivatives preferentially recovered by non-polar solvents such as hexane, which may explain the pronounced hexane-fraction activity of both species in the present study.

Comparable growth-suppressive trends for the remaining botanicals are supported by independent literature. For *A. indica*, Hassan *et al*. (2018) reported 56–68% reductions in larval weight with neem extracts, and Charleston *et al*. (2006) confirmed adverse effects of neem-based extracts on *P. xylostella* development. For *N. oleander*, earlier workers recorded 26–63% reductions in larval weight gain in *Crocidolomia binotalis*, attributed to oleandrin-driven antifeedant activity, consistent with Sivakumar *et al*. (2022) on *Helicoverpa armigera*. For *T. erecta*, a 50% reduction in larval weight of *S. frugiperda* was reported at 500 ppm acetone extract, consistent with the thiophene and polyacetylene content typical of *Tagetes* species. *P. rubra*, though less studied against lepidopteran larvae specifically, has documented larvicidal activity against mosquito vectors, suggesting a toxicity-driven rather than antifeedant mode of action — consistent with its comparatively weaker suppression of larval weight gain here.

Overall, the stronger and more consistent activity of *S. grandiflora*, *S. macrophylla*, and *T. erecta* relative to *A. indica*, *N. oleander*, and *P. rubra* mirrors the broader literature pattern in which antifeedant- and growth-regulator-rich extracts suppress larval biomass more effectively than those acting mainly through direct toxicity. The generally greater activity of methanol and ethyl acetate extracts over hexane further points to polar-to-semi-polar constituents as the principal drivers of growth inhibition, with *S. grandiflora* and *S. macrophylla* as notable exceptions. Since the treated check recorded the lowest larval weight gain throughout, these botanicals while less potent than the synthetic standard show clear potential as components of an integrated management strategy for *P. xylostella*.

Conclusion

This comparative evaluation demonstrated that methanol, ethyl acetate, and hexane extract of selected botanicals differentially affect the larval weight gain of *Plutella xylostella*. Extracts of *Solanum grandiflora*, *Swietenia macrophylla*, and *Tagetes erecta* particularly in methanol and ethyl acetate were the most effective in suppressing larval weight gain, indicating their potential as candidates for botanical pesticide development. Hexane extracts were generally less effective, pointing to polar and semi-polar compounds as the principal bioactive constituents. Further studies involving phytochemical characterization, dose-response bioassays, and field-level evaluation are recommended to validate these findings and support the practical deployment of these botanicals in sustainable DBM management.

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Conflict of Interest: The authors declare that they have no conflict of interest.

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