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Evaluation of the larvicidal potential of *Solanum xanthocarpum* Fruit Extract Against *Aedes albopictus*

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Abstract
Aedes albopictus is a highly invasive mosquito vector that transmits dengue fever, chikungunya, and Zika virus disease worldwide. Resistance and environmental concerns have made synthetic insecticides increasingly ineffective, necessitating plant-derived alternatives. The present study used an aqueous extract of the fruit of *Solanum xanthocarpum* Schrad. & Wendl. Third-instar larvae of *Ae. albopictus* were used to assess larvicidal activity under laboratory conditions. Preliminary phytochemical screening identified alkaloids, saponins, tannins, flavonoids, and phenolic compounds. Larvicidal bioassays conducted at five concentrations (25, 50, 100, 150, and 200 µg/mL) according to the WHO protocol (2005) showed concentration- and time-dependent larval mortality. The LC_{50} was 274.5 µg/mL at 24 h and 83.7 µg/mL at 48 h. The effect of concentration on larval mortality was significant (One-way ANOVA, $F(4, 120) = 32.33$; $p < 0.0001$). Based on these findings, *S. xanthocarpum* fruit extract exhibits considerable larvicidal activity, warranting exploration toward the development of eco-friendly mosquito control agents.

Keywords: Aedes albopictus, *Solanum xanthocarpum*, Phytochemicals, Mortality, Dengue fever

Introduction

The Asian tiger mosquito *Aedes albopictus* (Skuse, 1894) is one of the most widespread invasive arthropod species in the world. It was originally found in Southeast Asia and has since spread to more than 70 countries across Europe, the Americas, Africa, and the Indian subcontinent (Bonizzoni *et al*., 2013; Gratz *et al*., 2004; Dass *et al*., 2026) [6, 8, 12]. It is an exceptionally ecologically plastic species that tolerates a wide range of breeding habitats and can endure temperate winters, so it has been spreading rapidly worldwide and is a major public health threat in both urban and rural settings (Hawley *et al*., 1988) [14].

Ae. albopictus is a competent vector of at least 22 arboviruses and is responsible for the spread of dengue fever, chikungunya, and Zika virus. Outbreaks are reported in new non-endemic areas (Nawarathne *et al*., 2024; Bhatt *et al*., 2013) [4, 21]. Dengue incidence has increased over the past few decades and is estimated to cause 390 million infections, along with significant morbidity, mortality, and economic burden each year (Bhatt *et al*., 2013) [4]. Recent outbreaks of chikungunya and Zika virus further underscore the urgent need for sustainable and efficient vector control measures (Nawarathne *et al*., 2024) [21].

Traditional vector control is heavily dependent on synthetic insecticides, especially organophosphates and pyrethroids; however, the widespread occurrence of insecticide resistance in Ae. albopictus populations has rendered the approach ineffective (Rizvi *et al*., 2020; Dhiman *et al*., 2022) [11, 22]. Additionally, the use of indiscriminate insecticides is associated with detrimental environmental impacts, including toxicity to beneficial insects, pollution of aquatic systems, and effects on pollinator communities (Baskar *et al*., 2018) [3]. This has led to a paradigm shift toward bio-rational and eco-friendly alternatives, particularly bio-larvicides extracted from Medicinal Plants (Baskar *et al*., 2018; Dass *et al*., 2026) [3, 8].

Plant extracts are a treasure trove of bioactive compounds that have been shown to have insecticidal and larvicidal effects, and their potential has only begun to be tapped (Singh *et al*., 2023; Yadav *et al*., 2015) [24, 26]. *Solanum xanthocarpum* Schrad. & Wendl., yellow-berried nightshade or wild brinjal, is a thorny undershrub found throughout the Indian subcontinent and is commonly used in Ayurvedic medicine as a remedy for asthma, cough, and fever (Changbunjong *et al*., 2010; Mohan *et al*., 2005) [7, 18]. It is known to contain a variety of phytochemicals, including steroidal alkaloids (solanine, solasonine, solasodine), saponins, tannins, flavonoids, and phenolic acids, some of which have been shown to possess cytotoxic, anti-inflammatory, and antimicrobial properties (Yadav *et al*., 2014; Maheswaran *et al*., 2012) [16, 27]. Extracts of *S. xanthocarpum* have demonstrated larvicidal activity against *Culex quinquefasciatus* and *Anopheles stephensi*, and leaf and fruit extracts have been found to be promising at sub-lethal concentrations by several investigators (Mohan *et al*., 2007; Mahesh Kumar *et al*., 2012) [15, 17]. Although these results are encouraging, the larvicidal efficacy of *S. xanthocarpum* fruit extract against *Aedes albopictus*, a major vector of dengue and chikungunya viruses, has not been fully studied (Simeon-Oke *et al*., 2015) [23].

For this purpose, the following three objectives were set: (i) to prepare an aqueous extract of *S. xanthocarpum* fruit by cold maceration and determine its yield; (ii) to conduct phytochemical screening of the extract using the Harborne

(1998) [13] procedure; and (iii) to test the larvicidal activity of the extract against third-instar larvae of *Ae. albopictus* under standard WHO (2005) laboratory bioassay conditions to provide a scientific basis for further research and development of *S. xanthocarpum* fruit extract as an eco-friendly larvicidal agent.

Materials and Methods

Plant material collection and authentication

The fruits of *Solanum xanthocarpum* Schrad. & Wendl. were collected from the Thanjavur region, Tamil Nadu, India, on 02/03/2026. The plant was identified by a qualified taxonomist, and a voucher specimen (Accession No. SX004) was deposited in the institutional herbarium at Bon Secours College for Women, Thanjavur. The freshly collected fruits were washed with distilled water, sliced into thin pieces, and shade-dried for 14 days at 28 ± 2 °C to a constant weight. The dried material was coarsely ground in a mechanical grinder, sieved through a 40-mesh sieve to obtain a uniform powder, and stored in an airtight container at 4 °C until further use.

Preparation of fruit extract

An aqueous extract was prepared by cold maceration: 100 g of fruit powder was steeped in 1000 mL of distilled water at room temperature for 72 h, with occasional stirring. The macerate was filtered through Whatman No. 1 filter paper, and the filtrate was concentrated under reduced pressure using a rotary evaporator at 45 °C to obtain a semi-solid residue. The extract was reconstituted in 0.1% dimethyl sulphoxide (DMSO) to prepare a stock solution of 10,000 µg/mL. Serial dilutions of the stock solution were prepared to obtain working concentrations of 25, 50, 100, 150, and 200 µg/mL for bioassay purposes.

Preliminary phytochemical screening

Preliminary phytochemical screening was performed according to standard methods to detect alkaloids (Dragendorff's and Mayer's reagents), saponins (foam test), tannins and phenolic compounds (ferric chloride test), steroids (Liebermann-Burchard test), flavonoids (alkaline reagent test), and anthraquinones (Borntrager's test).

Mosquito larvae

Laboratory-reared *Aedes albopictus* larvae were maintained in standard insectary conditions at 27 ± 2 °C, 75 ± 5% relative humidity, and a 12:12 h light: dark photoperiod. Larvae were fed a diet of powdered yeast and dog biscuits (3:1 ratio) throughout the rearing cycle. Healthy, late third-instar larvae obtained from the third consecutive laboratory generation were used for all bioassays to minimize genetic variability arising from initial field-collected individuals.

Larvicidal bioassay

Larvicidal bioassays were conducted in accordance with the standard WHO guidelines for laboratory testing of mosquito larvicides (WHO, 2005). Twenty-five third-instar larvae were introduced into 250 mL glass beakers containing 249 mL of distilled water and 1 mL of the respective test solution to achieve the desired final concentration (25, 50, 100, 150, or 200 µg/mL). Each concentration was tested in triplicate, with a solvent control (0.1% DMSO in distilled water) included in every batch. Larval mortality was recorded at 24 h and 48 h post-treatment. Larvae were considered dead if they failed to show any movement when gently probed with a fine brush. Dead larvae were counted and removed at each observation interval to prevent decomposition from confounding subsequent counts (Murugesan *et al*., 2025) [20].

Statistical analysis

Percentage larval mortality was calculated for each concentration and time point, and data were expressed as mean ± standard deviation (SD) of three replicates. A one-way analysis of variance (ANOVA) followed by Tukey's honestly significant difference (HSD) post-hoc test was used to compare larval mortality across concentration groups ($\alpha = 0.05$). Lethal concentration values (LC50 and LC90) along with their 95% confidence intervals were calculated using probit regression analysis. All statistical computations were performed using GraphPad Prism (version 9.0, GraphPad Software, San Diego, CA, USA).

Results

Qualitative phytochemical analysis

Qualitative phytochemical screening of the aqueous fruit extract of *Solanum xanthocarpum* revealed the presence of alkaloids, saponins, tannins, flavonoids, and phenolic compounds. The extract contained neither steroids nor anthraquinones under the test conditions. These secondary metabolites, especially alkaloids, saponins, flavonoids, tannins, and phenols, could contribute to the biological activity of the fruit extract and might be linked to larvicidal activity against *Aedes albopictus*. However, more detailed phytochemical analysis is needed to identify the specific bioactive compounds responsible for these effects.

Table 1: Qualitative phytochemical analysis of Solanum xanthocarpum fruit extract.

S. No	Phytochemicals	Aqueous Extract
	Alkaloid	+
	Saponin	+
	Tanins	+
	Steroids	-
	Flavonids	+
	Phenol	+
	Anthraquinone	-

Larvicidal activity of *Solanum xanthocarpum*

The fourth-instar larvae of *Aedes albopictus* were exposed to the aqueous fruit extract of *Solanum xanthocarpum*, which exhibited concentration- and exposure-dependent larvicidal activity. At 24 h, larval mortality increased from 13.05 ± 2.459% at 25 µg/mL to 47.04 ± 2.449% at 200 µg/mL. After 48 h of exposure, mortality increased from 21.00 ± 3.162% at 25 µg/mL to 84.00 ± 2.449% at 200 µg/mL. Therefore, mortality was consistently higher at 48 h than at 24 h across all concentrations tested. Larvicidal efficacy at 24 and 48 h increased with exposure duration, with LC_{50} values of 274.5 µg/mL and 83.7 µg/mL, respectively. Statistical analysis showed a significant difference among treatment concentrations ($F(4, 120) = 32.33$, $p < 0.0001$).

Table 2: Larvicidal activity of the Solanum xanthocarpum fruit extract against 4th instar larvae of *Aedes albopictus*.

Dosage µg/mL	24 h mortality	48 h mortality	LC_{50} µg/mL 24 h	LC_{50} µg/mL 48 h	df	MS	F (DFn, DFd)	p value
25	13.05 ± 2.459	21.00 ± 3.162	274.5	83.7	120	71.3	F (4,120) = 32.33	p < 0.0001
50	19.07 ± 3.741	32.03 ± 3.741						
100	27.54 ± 2.186	46.05 ± 2.449						
150	38.10 ± 1.435	62.07 ± 3.741						
200	47.04 ± 2.449	84.00 ± 2.449						

Discussion

The present study findings show that the aqueous fruit extract of *Solanum xanthocarpum* exhibits significant, concentration- and time-dependent larvicidal activity against third-instar larvae of *Aedes albopictus*. Bioaccumulation or cumulative physiological disruption of the bioactive constituents in the larvae resulted in a progressive increase in larvicidal potency at 24 h (274.5 µg/mL) and 48 h (83.7 µg/mL). The one-way ANOVA result ($F(4, 120) = 32.33$; $p < 0.0001$) confirmed that concentration was a very significant factor in larval mortality across the various treatments and supports the dose-response trend observed in the bioassay data.

The larvicidal activity observed in this study, when compared with the existing literature, is comparable to that of extracts from other Solanum species tested against other mosquito species. The 24 h LC_{50} of the ethanolic leaf extract of S. xanthocarpum was reported as 166.2 µg/mL against *Culex quinquefasciatus* by Mahesh Kumar *et al.* (2012), which is more potent than the 24 h LC_{50} observed in this study. The relatively low LC_{50} of the fruits obtained in the present study might be explained by the use of various parts of the plant (fruit vs. leaf), different solvents used to extract the bioactive compounds (aqueous vs. ethanolic), and the differential distribution of non-polar bioactive compounds between the organic solvent and the aqueous phase, as well as by different insect species tested (Ae. albopictus vs. *Cx. quinquefasciatus*).

In comparison, the present study reports larvicidal activity against Ae. albopictus. The 48 h LC_{50} of 83.7 µg/mL obtained for *Ae. albopictus* in the present study is comparable to those of some botanical extracts. Similar LC_{50} values were reported by Bilal *et al.* (2012) [5] for various plant extracts, ranging from 120 to 450 µg/mL for *Ae. albopictus*, and by Ahbirami *et al.* (2014) [1] for a plant-derived extract, which reported a 48 h LC_{50} of 92.3 µg/mL for Ae. albopictus, similar to the values obtained in the current investigation. However, Dhayalini *et al.* (2016) [10] reported a higher 48 h LC_{50} of 158.4 µg/mL for an alternative botanical formulation, suggesting that *S. xanthocarpum* fruit extract is equally effective or more effective when tested under the same conditions.

The observed larvicidal activity is likely due to the complex mixture of phytochemical constituents in the fruit extract, including alkaloids, saponins, tannins, flavonoids, and phenolic compounds. Steroidal alkaloids such as solasonine and solanine have been reported to inhibit acetylcholinesterase (AChE) activity, which disrupts normal neurotransmission and causes paralysis in insect larvae (Yadav *et al.*, 2014; Maheswaran *et al.*, 2012) [16, 27]. Saponins are well documented to act as detergent-like amphiphilic agents, altering the integrity of midgut epithelial cell membranes in larvae by increasing permeability and causing ion leakage (Mohan *et al.*, 2010) [19]. The larvicidal activity of tannins results from protein precipitation and disruption of digestive enzyme activity (Bansal *et al.*, 2009) [2], and flavonoids and phenols induce oxidative stress by generating reactive oxygen species (Singh *et al.*, 2023) [24].

What is of special practical significance in the present study is the use of aqueous extraction. Water is a universal, non-toxic solvent that eliminates the occupational and environmental risks associated with organic solvents and enables large-scale preparation in resource-limited environments. In addition, the aqueous extract is likely to be less damaging to non-target aquatic organisms than solvent-based formulations, but non-target bioassays need to be conducted carefully to confirm this assumption before application in the field. Based on the above results, it can be concluded that the aqueous extract of *S. xanthocarpum* fruit material is effective at concentrations lower than 100 µg/mL for 48 h, indicating its potential for developing a water-based larvicidal formulation.

There are some limitations to the present study that should be noted. Only laboratory conditions were used for the bioassay, and these conditions may not fully represent the often-complex physicochemical and biological interactions found in natural larval habitats. The identity and contribution of each larvicidal compound were not determined, and bioactivity-guided fractionation will be required to isolate and characterize the major active constituents. In addition, no attempt was made to assess sublethal effects on larval development or adult emergence, nor was there an evaluation of potential toxicity to non-target aquatic organisms. Before it can be recommended for use in an IVM program, these aspects need to be considered in future studies.

Conclusion

The results of the present study provide compelling evidence that the aqueous fruit extract of *Solanum xanthocarpum* possesses significant larvicidal activity against *Aedes albopictus*, with LC50 values of 274.5 µg/mL at 24 h and 83.7 µg/mL at 48 h. Preliminary phytochemical screening identified alkaloids, saponins, tannins, flavonoids, and phenolic compounds as the likely bioactive constituents responsible for the observed larvicidal effects. The highly significant ANOVA result ($F(4, 120) = 32.33$; $p < 0.0001$) confirms a robust concentration-mortality relationship, supporting the potential of this plant extract as a bio-rational tool for mosquito larval control. Future investigations should focus on bioactivity-guided fractionation and isolation of the principal larvicidal compounds, evaluation of sublethal effects on larval development and adult emergence, assessment of non-target toxicity to beneficial aquatic organisms, and field efficacy trials under natural breeding conditions. Such studies will be essential to establish the safety profile, environmental compatibility, and operational suitability of *S. xanthocarpum* fruit extract for integration into sustainable vector management strategies.

Data availability
Open for all

Declaration
We declare that this manuscript is original, has not been published before, and is not currently being considered for publication elsewhere.

Conflict of interest
The authors declare no conflicts of interest

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