

# Comparative Study of Larvicidal Efficacy of Different Plant Extracts on *Culex quinquefasciatus*

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## **Abstract**

*The experiment intends to analyze and assess the bioactive competence of some plants such as *Chromolaena odorata*, *Azadirachata indica*, *Ocimum tenuiflorum*, *Vitex negundo*, *Citrus sinensis*, *Citrus limon*, *Eucalyptus globulus*, *Calendula officinalis*, and *Aloe barbadensis*. All these extracts were evaluated for larvicidal activity. The highest larvicidal activity was recorded as 100% for cold water crude extract of *Citrus limon*, 95% respectively for hot and cold-water crude extracts of *Vitex negundo*, cold water extract of *Citrus sinensis*, and cold-water extract of *Calendula officinalis*. Therefore, plants may be explored widely as an alternative to modern medicine.*

**Keywords:** *Bioactive efficacy, natural compounds, larvicidal activity*

## 1. Introduction

Mosquitoes are significant vectors of human diseases, including yellow fever, dengue, malaria, and encephalitis which pose a global health threat.<sup>[1]</sup> Mosquito-borne diseases are a global health crisis, with India facing particular challenges due to its complex socio-ecological landscape exacerbating its vulnerability to these infections.<sup>[2]</sup> The *Culex* genus, prevalent in warm tropical temperate regions, is a particular concern due to its association with various viral diseases.<sup>[3]</sup> Among *Culex* species, *Culex quinquefasciatus* is a dominant anthropophilic species found in subtropical regions of America, Asia, Africa, Australia, and New Zealand. This species is a primary vector for arbovirus causing Rift Valley fever and Saint Louis encephalitis.<sup>[4]</sup>

Larvicides are essential for preventing and controlling mosquito populations, but their use is associated with various environmental and ecological risks.<sup>[5]</sup> The traditional mosquito control methods focus on eliminating breeding sites, and the containment of mosquito larvae and adults by introducing larvicides (methoprene, larvicidal oils, monomolecular surface films) and insecticides (organochlorines, pyrethroids, carbamates, organophosphates, phenylpyrazoles).<sup>[6]</sup> While they are effective, these chemicals pose significant hazards to both human health and the environment. To mitigate these risks, researchers are exploring the potential of natural products as an eco-friendly alternative.<sup>[7]</sup> By leveraging the medicinal properties of plants, it is hoped to develop effective and sustainable strategies for mosquito control. India boasts a rich tradition of utilizing medicinal plants for healthcare, with a vast majority of the population incorporating them into daily life. According to the National Institutes of Health (NIH), over 200 different plant varieties form the basis of numerous medicinal preparations.<sup>[8]</sup> The World Health Organization (WHO) further emphasizes the reliance of 80% of the population on traditional medicine derived from plants and their active compounds.<sup>[9]</sup> These plants have been employed for centuries to treat a wide array of ailments with various parts of the plants.<sup>[9,10]</sup>

Given the growing interest in environmentally friendly pest control, this study investigates the larvicidal potential of several plant extracts namely *Chromolaena odorata*, *Azadirachata indica*, *Ocimum tenuiflorum*, *Vitex negundo*, *Citrus sinensis*, *Citrus limon*, *Eucalyptus globulus*, *Calendula officinalis*, and *Aloe barbadensis* against the 3<sup>rd</sup> instar larvae of *Culex quinquefasciatus*. These plant extracts have demonstrated efficacy as mosquito repellents. The primary goal of this research is to develop a sustainable, natural larvicide as a safer alternative to synthetic chemicals, which pose risks to both human health and the environment.

## 2. Materials and Methods

### 2.1. Collection of Plant material

Leaves, peels, and gel samples from commonly available plants were collected from various locations in Mettupalayam and Nilgiris district, Tamil Nadu. *Table 1* and *Figure 1* provide a detailed list of the plant species used in this study.

**Table 1. List of plants used in the study**

S. No	Plant Name	Family	Common Name	Vernacular name (Tamil)	Parts Used
1	<i>Chromolaena odorata</i>	Asteraceae	Siam weed, camphor grass	Oonni chedi	L
2	<i>Azadirachata indica</i>	Meliaceae	Neem	Vembu	L
3	<i>Ocimum tenuiflorum</i>	Lamiaceae	Holy basil or Tulsi	Tulasi	L
4	<i>Vitex negundo</i>	Lamiaceae	Chaste tree	Nocchi	L
5	<i>Citrus sinensis</i>	Rutaceae	Orange	Thoodaipalam	P
6	<i>Aloe barbadensis</i>	Asphodelaceae	Aloe vera	Katraalai	G & L
7	<i>Eucalyptus globulus</i>	Myrtaceae	Eucalyptus	Thaila ilia	L
8	<i>Calendula officinalis</i>	Asteraceae	Marigold	Sendigaipoo	L
9	<i>Citrus limon</i>	Rutaceae	Lemon	Elumichai	P

Note: L – Leaves, P – Peel and G – Gel

**Figure 1. Different plant parts used in the study**

## 2.2. Preparation of the extract

### 2.2.1. Hot water extract

About 10g of each plant sample was weighed and cleaned by rinsing thrice in tap water and sterile distilled water to eliminate contaminants. Subsequently, the samples were boiled in 100 mL of sterile distilled water for 30 mins at 45 °C. The resulting mixture was filtered through muslin cloth and centrifuged at 3000 rpm for 10 mins. The supernatant was collected using Whatman No. 1 filter paper and stored for further analysis.<sup>[11]</sup>

### 2.2.2. Cold water extract

About 10g of each plant sample was weighed and thoroughly cleaned with tap water thrice, followed by sterile distilled water to remove impurities. Under aseptic conditions, the samples were crushed using a motor and pestle with 100 mL of sterile distilled water. The resulting mixture was filtered with muslin cloth and centrifuged for 10 mins at 3000 rpm. The supernatant was collected using Whatman No: 1 filter paper for subsequent analysis.<sup>[11]</sup>

## 2.3. Mosquito larvae collection

The mosquito larvae were obtained from the National Centre for Disease Control (NCDC) at Mettupalayam, Nilgiris district, during March and April and housed in a plastic container. To ensure safety, the larvae were maintained in a mosquito chamber under a standard laboratory which survived in normal environmental conditions.

## 2.4. Mosquito larvicidal bioassay

The larvicidal activity of plant extracts against *Culex quinquefasciatus* mosquito larvae was assessed by measuring mortality rates. A negative control group of 10 larvae was placed in a 100 mL beaker with 50 mL of drinking water. A positive control group of 10 third-instar larvae were exposed to 45 mL of drinking water containing 5 mL of acetone. The experimental groups consisted of 10 larvae exposed to 50 mL solutions containing various concentrations of plant leaves and peels with 5 mL, 10 mL, and 15 mL of cold and hot water plant extracts. Each treatment was conducted in two replicates.<sup>[11,12]</sup> After 24 hours, larval mortality was recorded and calculated as a percentage.

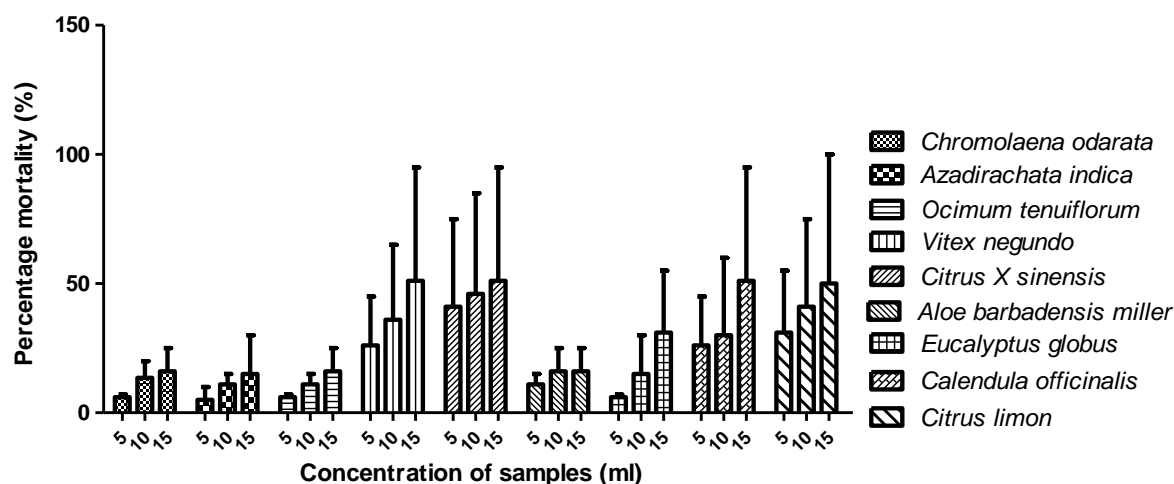
$$\text{Percentage Mortality} = \text{Number of dead larvae} / \text{Number of larvae introduced} \times 100$$

## 3. Results and discussion

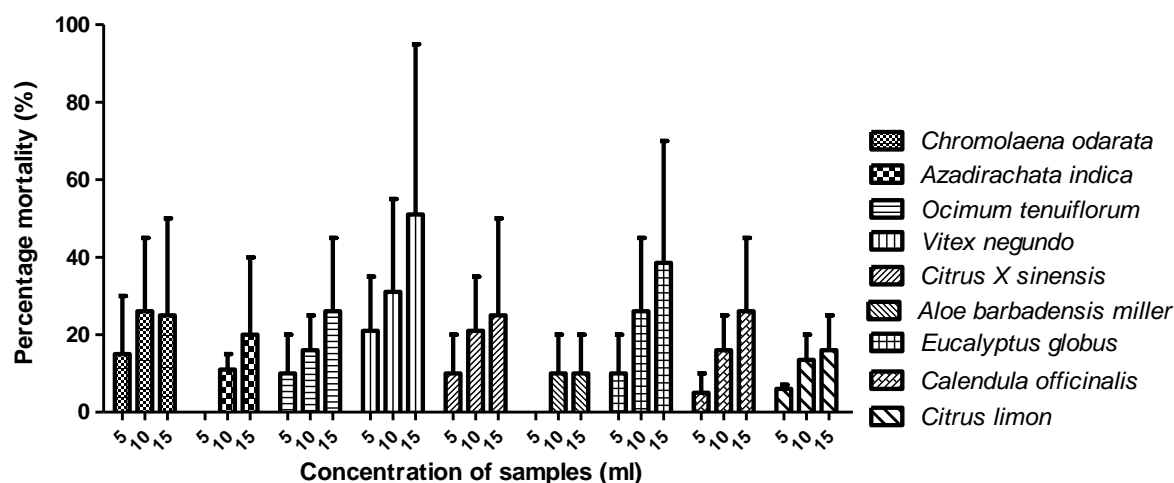
### 3.1. Mosquito larvicidal bioassay of plant extracts

The larvicidal potential of various plant extracts against the third instar larvae of *C. quinquefasciatus* was evaluated under sterile conditions. The cold and hot water extracts of *C. odorata*, *A. indica*, *O. tenuiflorum*, *V. negundo*, *C. sinensis*, *A. barbadensis*, *E. globulus*, *C. officinalis*, and *C. limon* were tested for their ability to act as larvicides. All the plant extracts showed moderate larvicidal effects after 24 h; notably, the highest larvicidal activity was recorded as 100 % for cold water crude extract of *Citrus limon*, 95 % respectively for hot and cold-water crude extracts of *Vitex negundo*, cold water extract of *Citrus sinensis* and cold-water extract of *Calendula officinalis* are shown in *Figure 2* and *Figure 3*.

**Figure 2. Mosquito larvicidal bioassay activity of cold-water extracts**



**Figure 3. Mosquito larvicidal bioassay activity of hot-water extracts**



Previous research showed that methanolic leaf extracts of *Tinospora rumphii* exhibited the highest larvicidal activity against *Aedes aegypti* mosquitoes, achieving up to 90 % mortality. In contrast, *Jatropha curcas* leaf extracts demonstrated the lowest efficacy among the tested plant samples. Our mosquito larval bioassay results aligned with these findings. <sup>[11]</sup>

#### 4. Conclusion

The bioactive molecules present in the plants are used as therapeutic drugs in order to overcome the limitations that are rendered by modern medicine. Based on the above ideas the present experiment was intended to screen the larvicidal efficacy of the plant extracts. our research shows that all the different plant extracts are used as good larvicides. Among these, *Citrus limon* leaf, *Calendula officinalis* leaf, and *Citrus sinensis* peel can be potentially used as eco-friendly larvicides. These results reveal that the plants possess numerous bioactive efficacies, which could be used more in the future as an alternative to larvicides.

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## Conflict of interest

The authors declare that they have no conflict of interests.

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