

## Larvicidal activity of *Acalypha indica* L. (Euphorbiaceae) solvent extracts against mosquito vectors

P Saranraj<sup>1\*</sup>, K Inbavalli<sup>2</sup>, T Prabu<sup>3</sup>, B Lokeshwari<sup>1</sup>, R Nisha<sup>1</sup>

<sup>1</sup> Department of Microbiology, Sacred Heart College (Autonomous), Tirupattur, Tamil Nadu, India

<sup>2</sup> Department of Biochemistry, Acharya Institute of Allied Health Sciences, Soladevanahalli, Bangalore, Karnataka, India

<sup>3</sup> Department of Zoology, H. H. The Rajah's College (A), Pudukkottai, Tamil Nadu, India

### Abstract

*Acalypha indica* L. is a medicinal plant, referred in English as "Indian Acalypha". It is widespread in India as well as numerous other countries. A multitude of nations have documented its extensive utilisation in traditional medicine. It is employed in the treatment of bronchial asthma, skin conditions, and constipation, among others. Worldwide, mosquitoes are a significant vector of numerous human maladies. The present research was aimed to assess the Larvicidal efficacy of the medicinal herb *Acalypha indica* L. against three distinct species of Mosquitoes. At concentrations of 50, 100, 150, 200, and 250 ppm, ethanol, acetone, and chloroform extracts of the *Acalypha indica* plant were applied to Third-instar larvae. After observing the mortality after a 24 hours exposure, the LC<sub>50</sub> and LC<sub>90</sub> values were calculated. The results clearly indicate that the effectiveness of larvicidal agents was dependent on the dosage. The ethanol extract of *Acalypha indica* exhibited the highest larvicidal efficacy when tested against *Culex quinquefasciatus*.

**Keywords:** *Acalypha indica* L., *Culex quinquefasciatus*, *Anopheles stephensi*, *Aedes aegypti*, Larvicidal activity and Ovicidal activity

### Introduction

In earlier days, the primary source of medication was plants and its products. Native Americans have used a range of herbal remedies to effectively treat a wide range of illnesses. The botanicals utilized, as well as how medications are prepared and given, differ from place to place. Even while traditional healers and elderly tribal people are increasingly losing their expertise of herbal medicine, some of them are still using plants as a kind of treatment. A great deal of traditional natural products are currently gaining popularity. The diversity and abundance of therapeutic plants found in India are well-known (Renganathan *et al.*, 2009) [1]. Traditional healers in India employ over 2,500 plant species for medicinal purposes (Bussmann and Glenn, 2010) [2]. The perennial erect herb *Acalypha indica* L. is also known as "Kuppai meni". It is a member of the Euphorbiaceae Family. This shrub is widely found in Indian gardens, backyards, and waste areas across the country's plains. Herbal activity is present in *Acalypha indica*'s root, stem, and leaf (Saranraj *et al.*, 2010) [3].

Since they can spread infections that cause diseases that affect millions of people globally, mosquitoes pose a serious threat to human health (WHO, 2010) [4]. *Aedes*, *Anopheles*, and *Culex* genera contain a number of species that act as vectors for diseases such as Filariasis, Japanese encephalitis, Malaria, Dengue fever, and Dengue hemorrhagic fever (Borah *et al.*, 2010) [5]. Dengue and Yellow fever are known to be carried by *Aedes aegypti*, Malaria by *Anopheles stephensi*, and Filarial illness by *Culex quinquefasciatus*. Counting between 300 and 500 million cases every year, Malaria remains one of the most significant infectious diseases. Approximately, 40 % of people on the planet currently reside in regions where Malaria is endemic (Werndorfer, 2003) [6]. Lymphatic filariasis is mostly transmitted by the *Culex quinquefasciatus* vector, which is found in tropical regions (Bernhard *et al.*, 2003) [7]. In addition to having more breeding sites within urban areas,

these diseases are currently spreading because mosquitoes are becoming more resistant to commercial insecticides like carbamates, organochlorides, and organophosphates as well as biological insecticides (Yadav *et al.*, 1997) [8]. The present research was designed to evaluate the Larvicidal activity of the Medicinal plant *Acalypha indica* L. (belongs to the Euphorbiaceae family) Solvent extracts against the disease transmitting Mosquito vectors.

### Material and Methods

#### 1. Collection and Identification of Plant material

A commercial Stainless steel blender was used to grind 100 g of dried *Acalypha indica* L. leaves into a fine powder. Then, in a Soxhlet device, the leaves were extracted with 500 ml of Chloroform, Acetone, and Ethanol until all of the liquid was gone. Rotavapor concentrated the extract at 45 °C with a lowered pressure of 22 – 26 mm Mercury. The residue that was left over was stored in a Sterile container at 4 °C, sealed in silver foil, and brought to the laboratory.

#### 2. Extraction method

Hundred grams of *Acalypha indica* shade dried leaves were dried and ground into a powder using a Stainless steel blender. The powdered leaves were then extracted in a Soxhlet apparatus using ethanol, acetone, and chloroform (500 ml, Ranchem) in order of exhaustion. The extract was concentrated by "Rotavapour" at 45 °C under reduced pressure of 22 – 26 mm Mercury, and the obtained residue was kept at 4 °C in an amber vial (Saranraj *et al.*, 2022) [9]. Following that, the vials were given names, wrapped in silver foil, and brought to the lab. The vials were stored at 4 °C in a dark, cold place until they were used.

#### 3. Larvicidal activity

The larvicidal efficacy of *Acalypha indica* solvent extracts was assessed using the previously published WHO procedure. Six distinct test concentrations *viz.*, 50 mg/L, 100

mg/L, 150 mg/L, 200 mg/L, and 250 mg/L) were made from the stock solution and tested against recently moulted (0 – 6 hrs) Third instar Mosquito larvae of *Anopheles stephensi*, *Culex quinquefasciatus*, and *Aedes aegypti*. In order to produce the test medium (500 ml plastic cups), 1 ml of the appropriate test concentration dilution was added (Salomi *et al.*, 2023) <sup>[10]</sup>. This was combined with 249 ml of Dechlorinated water to make as 250 ml test solution. An amount of 50 mg/L of dry Yeast powder was supplied to the larvae at the water's surface. Parallel to each replicated experiment were the control trials, which lacked plant extracts. Four replicates were kept at a time for every experiment (Vedha *et al.*, 2023) <sup>[11]</sup>. For every experiment, a minimum of 25 larvae per concentration were employed. After 24 hours, Mosquito larvae mortality was studied by using the formula which was introduced by Abbott (1925) <sup>[12]</sup>, the Percentage mortality was adjusted for control mortality.

**Results and Discussion**

The findings of *Acalypha indica* L. Larvicidal activity against *Anopheles stephensi*, *Aedes aegypti*, and *Culex quinquefasciatus* using crude ethanol, acetone, and chloroform solvent extracts was furnished in the Table - 1. The ethanol extract of *Acalypha indica* revealed the plant's larvicidal qualities, indicating its potential application in the management of Mosquito larvae population. Larval mortality was seen in the *Acalypha indica* ethanol extracts (Tables - 2 to Table - 4). The most vulnerable species were *Anopheles stephensi*, *Aedes aegypti*, and *Culex quinquefasciatus*. The highest larvicidal activity against *Culex quinquefasciatus* larvae was demonstrated by the *Acalypha indica* ethanol extract, with LC<sub>50</sub> and LC<sub>90</sub> values of 94.21 and 177.21 mg/L, respectively. In relation to *Anopheles stephensi*, the LC<sub>50</sub> and LC<sub>90</sub> values of *Acalypha*

*indica* ethanol, acetone, and chloroform extract were 110.21, 117.86, and 139.07 mg/L; 204.14, 207.26, and 244.30 mg/L respectively. The *Acalypha indica* Chloroform Extract's LC<sub>50</sub> and LC<sub>90</sub> values against *Aedes aegypti* were 129.62, 148.28, and 170.00 mg/L; 232.86, 260.56, and 285.00 mg/L respectively.

According to the findings of Baluselvakumar *et al.* (2012) <sup>[13]</sup>, the ethanol leaf extract of *Acalypha indica* exhibited ovicidal and repellent properties against *Aedes aegypti*. At concentrations of 120, 160, 200, and 240 ppm, the ethanol extract of *Acalypha indica* caused 100 % egg mortality for *Aedes aegypti*, while at higher concentrations of 3.0 mg/cm<sup>2</sup>, it offered 100 % protection for 80, 100, 120, and 140 minutes. Baranitharan and Dhanasekaran (2014) <sup>[14]</sup> studied the larvicidal activity of *Acalypha indica* extracts in chloroform, benzene, and acetone extracts they exhibited the mortality at 73.49, 85.93, 76.03, and 80.56 mg/L, respectively. Elumalai *et al.*, (2012) <sup>[15]</sup> reported that, 100 % mortality was detected from the acetone and ethanol extracts of 100 ppm for *Acalypha indica* with LC<sub>50</sub> values of 121.65 and 139.86 ppm. When four different extracts of *Acalypha indica* were tested for their repellent properties against *Anopheles stephensi*, the results of the skin repellent test at concentrations of 1.0, 2.5, and 5.0 mg per cm<sup>2</sup> ranged in mean complete protection time from 119.17 to 387.83 minutes. Saranraj *et al.* (2023) <sup>[16]</sup> found that *Acalypha indica* LC<sub>50</sub> and LC<sub>90</sub> values for Acetone, Chloroform, and Acetone against *Culex quinquefasciatus* larvae First instar in a 24 hours period were 136.75, 145.69, 139.49, and 143.64 mg/L; 149.07, 158.24, 151.95, and 156.14 mg/L respectively. These findings may stimulate the hunt for novel, potent natural substances that can replace manufactured pesticides derived from other therapeutic plants.

**Table 1:** Probit analysis of larvicidal activity of *Acalypha indica* L. extracts against *Aedes aegypti*, *Anopheles stephensi* and *Culex quinquefasciatus*

Species	<i>Acalypha indica</i> Extract	LC <sub>50</sub> (mg/L)	95% Confidence Limits		LC <sub>90</sub> (mg/L)	95% Confidence Limits		x <sup>2</sup>
			LCL	UCL		LCL	UCL	
			<i>Aedes aegypti</i>	Ethanol		129.62	119.12	
	Acetone	148.28	137.42	159.26	260.56	241.36	286.33	0.489
	Chloroform	170.00	159.00	182.01	285.00	263.30	314.55	0.087
<i>Anopheles stephensi</i>	Ethanol	110.21	99.80	119.81	204.14	190.00	222.52	5.761
	Acetone	117.86	108.00	127.03	207.26	193.55	225.00	6.233
	Chloroform	139.07	128.61	149.36	244.30	227.16	267.00	1.283
<i>Culex quinquefasciatus</i>	Ethanol	96.21	86.46	105.10	177.21	164.87	193.13	4.317
	Acetone	107.07	97.74	115.81	188.85	176.33	204.86	3.660
	Chloroform	115.76	106.71	124.40	197.50	185.00	213.55	3.631

LC<sub>50</sub>= Lethal concentration that kills 50 % of the exposed parasite, LC<sub>90</sub>= Lethal concentration that kills 90 % of the exposed parasite. LCL – Lower Confident Limit, UCL - Upper Confident Limit.

**Table 2:** Larvicidal activity of *Acalypha indica* Ethanol extract against Larvae of *Aedes aegypti*, *Anopheles stephensi* and *Culex quinquefasciatus*

Mosquito larvae	Concentration of <i>Acalypha indica</i> Ethanol extract (ppm)	Mortality (%)
<i>Aedes aegypti</i>	25	38.0
	50	50.0
	75	64.0
	100	81.0
	125	89.0
	Control	0

<i>Anopheles stephensi</i>	25	33.0
	50	45.0
	75	61.0
	100	71.0
	125	83.0
	Control	0
<i>Culex quinquefasciatus</i>	25	45.0
	50	61.0
	75	68.0
	100	80.0
	125	91.0
	Control	0

**Table 3:** Larvicidal activity of *Acalypha indica* Acetone extract against Larvae of *Aedes aegypti*, *Anopheles stephensi* and *Culex quinquefasciatus*

Mosquito larvae	Concentration of <i>Acalypha indica</i> Acetone extract (ppm)	Mortality (%)
<i>Aedes aegypti</i>	25	35.0
	50	47.0
	75	61.0
	100	78.0
	125	86.0
	Control	0
<i>Anopheles stephensi</i>	25	30.0
	50	42.0
	75	58.0
	100	68.0
	125	80.0
	Control	0
<i>Culex quinquefasciatus</i>	25	42.0
	50	58.0
	75	65.0
	100	77.0
	125	88.0
	Control	0

**Table 4:** Larvicidal activity of *Acalypha indica* Chloroform extract against Larvae of *Aedes aegypti*, *Anopheles stephensi* and *Culex quinquefasciatus*

Mosquito larvae	Concentration of <i>Acalypha indica</i> Chloroform extract (ppm)	Mortality (%)
<i>Aedes aegypti</i>	25	30.0
	50	42.0
	75	56.0
	100	73.0
	125	81.0
	Control	0
<i>Anopheles stephensi</i>	25	25.0
	50	37.0
	75	53.0
	100	63.0
	125	75.0
	Control	0
<i>Culex quinquefasciatus</i>	25	37.0
	50	53.0
	75	60.0
	100	72.0
	125	83.0
	Control	0

## Conclusion

In this present research, we concluded that the Ethanol leaf extract of *Acalypha indica* used possess an excellent Larvicidal activity against the disease transmitting Mosquito vectors, *Anopheles stephensi*, *Aedes aegypti*, and *Culex quinquefasciatus*. The plentiful availability of *Acalypha indica* in this universe may make them economical for field use in Mosquito vector control programs for controlling various Vector borne deadly microbial diseases.

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