# LARVICIDAL EFFECT OF *ARISTOLOCHIA BRACTEATA* RETZ. AGAINST *AEDES AEGYPTI* L.

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**Abstract**

*Aedes aegypti* is one of the most important disease transmitting and nuisance causing mosquitos which transmits Dengue, Chikungunya. Development of resistance in vector mosquitoes against synthetic insecticides coupled with their persistence and toxicity to non-target organisms has been widely recognized and has promoted the search for new means of control strategies. Biologically active plant extracts have been well documented for evolving an economically sound and environmentally acceptable mosquito control programmes. In the present findings, the larvicidal activity of *Aristolochia bracteata* against *Aedes aegypti* L., hexane root, ethyl acetate seed extracts have 100% mortality at 250 ppm and 500 ppm respectively. The study reveals that the root extracts of *Aristolochia bracteata* at 250 ppm is better for mosquito control program. This investigation clearly observes that botanical insecticides act as good toxicants and will reduce the chemical burden on the environment.

**Keywords**: *Aristolochia bracteata*, *Aedes aegypti*, biopesticide, larvicidal properties,

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## I. INTRODUCTION

Mosquitoes (Diptera: Culicidae) are an ancient group of insects which has persisted for millions of years ago. Mosquitoes are one of the most successful groups in the animal kingdom that have exploited an ecological niche in and around human dwellings and settlements. Mosquitoes are pestiferous insects, which are responsible for the transmission of various viral diseases [17, 18, 19]. WHO has declared the mosquito as "**Public Enemy Number One**" because mosquitoes are responsible for the transmission of various dreadful diseases [18]. More than 3100 species of mosquitoes which belong to 34 genera, have been recorded and arranged under three sub-families, namely *Anophelinae, Culicinae* and *Toxorhynchitinae* [9]. This group assumes a greater importance since it acts as major vector of a number of diseases like Malaria, Filariasis, Dengue, Japanese Encephalitis and Chikungunya [13] [2]. *Aedes aegypti* is predominantly found in the tropics and the warmer temperate regions. *Aedes aegypti* mosquito, one of the most domesticated mosquitoes, breeds only in and around human habitations in tropical countries [7]. *Aedes aegypti* mosquito develops in rain water collected in artificial containers, tyres and water barrels and this mosquito transmits diseases like Dengue, Dengue Haemorrahgic Fever (DHF) and Chikungunya [8].

### *A. Mosquito and Herbal products*

Plant products have been used traditionally by human communities in many parts of the world against the vectors and pest species of insects [12]. Plants offers great promise as a rich source of phytochemicals for the control of vector mosquitoes, as they exhibit different degrees of toxicity to different stages of mosquito as potent larvicide, pupicide, adulticide, growth and reproductive inhibitor, repellent and oviposition deterrents. Traditional repellents not only provide protection against mosquito bites, but also curtail malarial transmission. The leaf extract of *Aristolochia indica*, *Leucas aspera*, *Ocimum sanctum* and pulp extracts of *A. sativum* and rhizome extract of *Curcuma longa* act as potent repellents against *Anopheles stephensi*. However there is no report on larvicidal properties of *Aristolochia bracteata* root, seed, leaf extracts on *Aedes aegypti*. Hence the present work has been carried out.

## II. MATERIALS AND METHODS

### A. The experimental species

The vector mosquito selected for the present study as experimental species is *Aedes aegypti* L. The seed, root and leaf extract of *Aristolochia bracteata* was taken for the present study. *Aristolochia bracteata* Retz. commonly known as 'bracteated birthwort' or 'worm killer' is a perennial herb, belongs to the family Aristlochiaceae. In Tamil it is known as *Adutinnappalai*. The plant exhibits purgative, emmenagogue, alterative, antiperiodic, antihelminthic properties and is used to treat syphilis, gonorrhoea and skin diseases. The plant is oxytocic [5].

### B. Preparation of plant extracts

*Aristolochia bracteata* (Aristolochiaceae) seeds, roots and leaves are collected Dindugal district, Tamil Nadu, India, were brought to the laboratory, shade dried at room temperature and powdered coarsely. The dried seeds, roots and leaves were extracted with hexane, ethyl acetate and methanol for a period of 72h each and filtered. The extracts were concentrated at reduced temperature on a rotary vacuum evaporator. The crude extracts of hexane, ethyl acetate and methanol of seeds, roots and leaves were then air dried and stored at a temperature of 40C. One percent stock solution was prepared by dissolving 1.5g of plant extract in 150ml of distilled water. From the stock solution 1000, 500, 250 125 and 62.5ppm concentration were prepared [17].

### C. Larvae rearing technique

The larvae of *A. aegypti* mosquitoes were collected from stagnant rain water at various places within Chennai. They were colonized and maintained continuously for generations since 2008 in the laboratory free to exposure to pathogens, insecticides or repellents. They were maintained at 27 ± 2ºC, 75-85%RH in the insectory of vector control laboratory. Larvae were fed with finely ground mixture of dog biscuits and yeast in the ratio 3:1. The first instar larvae developed into pupae through four stages in about 8-10 days. The pupae were transferred in to mosquito cage for emergence. Blood meal from mice was given to adult mosquitoes after three days of emerging out. After 3-4 days blood feeding for adult mosquitoes, the Petri dishes filled with tap water and lined with filter paper were placed in the cage for oviposition. The eggs were separated and placed in glass troughs for hatching.

### D. Larvicidal activity test

Each crude extract was dissolved in water with 0.1% emulsifier (Tween 80) to get the experimental concentration Twenty numbers of late third instar larvae of *A.aegypti* were kept in 500ml plastic bowls, containing 249 ml of dechlorinated tap water and 1ml of desired plant extract concentration. Three replicates for each

concentration were run at the same time. Tween 80 was used as a negative control. After 24 hours the percentage mortality [17] was calculated by using the formula (1) and corrections for mortality when necessary were done by using [1]Abbott's (1925) formula (2). Dead larvae were identified when they failed to move after probing with a needle in the siphon.

**E. Percentage mortality**:
Number of dead larvae/ Number of larvae introduced X 100 …….. (1)

**F. Corrected Percent mortality**:
(1-**n** in **T** After treatment) / **n** in **C** After treatment X 100 …….(2)

Where **n** is the number of larvae, **T** the treated and **C** is the control.

**Statistical analysis:** Profit analysis Finny [4], was used for determination of $LC_{50.}$

## III. RESULT AND DISCUSSION

As the concentration of the plant extract increases the total larval morbidity of the mosquitoes was also found to be increased. In the present study, clearly indicated that the hexane root extract showed 86.66% of mortality at 62.5ppm and 100% mortality were observed in 250ppm (Figure 1). The $LC_{50}$value of hexane root extract was 29.00ppm (Table.1). The ethyl acetate root extract showed 86.66% mortality at 62.5pm and 100% mortality at 1000ppm. The LC50values of ethyl acetate root extract were 50.75ppm. Seed extract of the ethyl acetate shows 100% mortality at 250ppm. The methanol seed extract showed 26.66% mortality at 62.5ppm and 81.66% mortality at 1000ppm. The $LC_{50}$ value of methanol seed extract were 2172.83ppm. Botanical derivatives in mosquito control, especially for mosquito larvae, as an alternative to synthetic insecticides offer a more environment friendly method of control [6]. Various plant species have been exploited to control the mosquito population throught the world [20, 16, 15, 10, 11]. Larvae from the three medically important mosquito genera *Aedes Anopheles* and *Culex* are all susceptible to a greater or lesser extents to some photochemicals [14]. The screening of local medicinal plants for mosquito larvicidal activity may eventually lead to their use in natural product based mosquito abatement practices [3]. The bioactive organic chemical contents may serve as insecticides, antifeedants, repellents, etc. They are less toxic, easily degradable, and do not have any adverse effects on non target organisms. Public health care is a vital component for the development of a disease free society. Promotion of health is essential to national progress. Health is the basis of a country's progress and it can be measured [8]. The present study confirms earlier investigations and establishes the fact that *Aristolochia bracteata* was the potent larvicide against *Aedes aegypti*. Therefore, further in-depth investigations on crude extract /phytotoxic compound of *Aristolochia bracteata* are needed to elucidate the larvicidal activity against a wide range of all stages of mosquito species and also the active ingredients of the extract responsible for larvicidal activity in *Aedes aegypti* should be identified, and small scale field trials are needed for usage of this plant as a mosquitocidal agent.

**Table 1**. Larvicidal activity of three solvent **extract of *Aristolochia bracteata*** on *Aedes aegypti*at different concentrations
(Mean ± S.D) (n=20)

| Solvent | Concentration (ppm) | 1000 ppm | 500ppm | 250ppm | 125ppm | 62.5ppm | LC 50 value (ppm) |
|---|---|---|---|---|---|---|---|
| **Hexane** | **Seed extract** | 12.66±1.15 | 10.33±0.57 | 08.00±0.00 | **20.00±0.00** | 06.66±0.57 | **50.69** |
| | **Root extract** | **20.00±0.00** | **20.00±0.00** | 06.66±0.57 | 18.66±0.57 | 06.66±1.15 | **29.00** |
| | **Leaf extract** | 07.66±0.57 | 07.33±0.57 | 05.66±0.57 | 17.33±0.57 | 03.66±1.15 | **-117.89** |
| **Ethyl acetate** | **Seed extract** | **20.00±0.00** | **20.00±0.00** | 19.66±0.57 | 18.66±0.57 | 17.33±0.57 | **1674.49** |
| | **Root extract** | **20.00±0.00** | 18.66±0.57 | 18.33±0.57 | 18.0±0.00 | 17.33±0.57 | **50.75** |
| | **Leaf extract** | 18.66±0.57 | 17.33±0.57 | 16.0±1.00 | 14.66±0.57 | 2.33±0.57 | **3958.88** |
| **Methanol** | **Seed extract** | 10.33±0.57 | 08.66±0.57 | 07.33±0.57 | 06.33±0.57 | 05.33±0.57 | **2172.83** |
| | **Root extract** | 08.33 ±0.57 | 06.66±0.57 | 06.33±0.57 | 05.66±0.57 | 04.33±0.57 | **3239.90** |
| | **Leaf extract** | 10.66±1.15 | 08.33±0.57 | 07.00±1.00 | 06.00±0.00 | 04.33±0.57 | **2298.09** |

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