



**The effect of Milkweeds (*Calotropis procera*),
Lemon grass (*Cymbopogon citratus*), Ageratum
(*Ageratum houstonianum*), Neem (*Azadirachta indica*)
leaf extracts as a larvicide against *Aedes aegypti***

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Abstract

Mosquito are the carrier of several diseases that have notable influence on the worldwide public health. Synthetic insecticides are most accepted method of mosquito control, but their overuse has led to the evolution of resistance in mosquitoes and environmental concerns. So, for that reason we need to explore the substitute methods of mosquito larva control including natural plant-based repellent. The present study work on mosquito repellent plants and their potency in controlling mosquito larva populations. We confer the methods used to identify and evaluate mosquito repellent plants, the criteria used to choose plants for evaluation, and the experimental design used to test the potency of the selected plants. The results of these studies are analysed to determine the most effective plant species and the factors that influence their potency. Ultimately, main focus on the advantages and limitations of using mosquito repellent plants compared to other methods of mosquito control and provide recommendations for further research and practical application. Our review highlights the potential of plant-based repellent as a non-toxic and eco-friendly alternative to chemical insecticides for mosquito control.

Keywords: Potency, repellent, insecticides, *Aedesa egypti*, plant extract.

Introduction

Mosquitoes are considered as vectors of several diseases such as malaria, dengue fever and Zika virus. Various ways have been done either naturally or chemically to reduce the mosquito population, with the intention of preventing and eradication mosquito borne diseases, or disturbances cause by these mosquitoes (Malavige et.al, 2004), (Guzma et.al, 2015). However, it should be endeavoured in mosquito control activities not to cause harm to humans and the environment. Chemical cope with mosquitoes among others by using mosquito repellent spray or mosquitos' repellent lotion that has been circulating in the market (West et. al, 2014; Majeed S., 2013). The overuse of synthetics has led to the evolution of resistance in mosquitoes as chemical/synthetics insecticides are the most usual method to control mosquito. Therefore, there is growing interest in exploring substitute methods for mosquitoes control counting the use of natural plant-based repellents. These natural plant-based mosquito repellents are innocuous and environmentally –safe, which takes them an interesting substitute of chemical/synthetic insecticides (Dewi, 2007). Over the span of years, there has been a growing body of research focused on identifying and evaluating the effectiveness of different plant species in repelling mosquitoes. Among these plant species are lemongrass, neem *calotropis*, and *ageratum* which have been found to be effective in repelling mosquitoes in various studies.

Plants (Plate 1)

A. Lemongrass (*Cymbopogon citratus*) is a tropical plant commonly used in herbal medicines. The extracts of lemon grass contain citral, which has been shown to repel mosquitoes effectively. A study has been conducted in Thailand found that a lotion containing 15%

lemongrass oil provided complete protection (Plate-1).





B. Neem (*Azadirachta indica*) the extracts of neem have been found to have insecticidal properties against mosquito larvae. There are certain active compounds present in neem azadirachtin, and limonoids disrupts the physiological processes of the larvae, leading to their death (Plate-1).

C. Ageratum is also known as *Ageratum houstonianum*, is a flowering plant that belongs to the Asteraceae family. It has distinctive blue, purple, flowers that grow in dense cluster. It's relatively low maintenance plant that can thrive in full moon as well as partial shade. Their fragrance contains coumarin, which is also known for its mosquito repellent properties and is commonly used in commercial insect repellents. The scent of ageratum acts as a natural deterrent for mosquitoes (Plate-1).

D. Calotropis is known as milkweed, the plants contain various bioactive compounds that possess insecticidal properties. The extracts pf *Calotropis* have been shown to be effective in repelling mosquito larvae. The active compound present in the plant extract disrupts the larval growth and development (Plate-1)

The utilization of natural mosquito larvae repellents such as *Calotropis*, Neem, Lemon grass, *Ageratum* leaf extracts can be safe and eco-friendly replacement to synthetic / chemical insecticides. These natural compounds have insecticidal belongings that can successfully control mosquito population, thus preventing mosquito –borne illnesses. Further studies are needed to optimize the use of these natural compounds in controlling mosquito larvae in different environmental conditions.

Plate 1- Plants name whose extracts are used for the experiment

S.No.	Common Name	Botanical Name
1	Milkweed	 <i>Calotropis procera</i>
2	Lemongrass	 <i>Cymbopogon citratus</i>
3	Neem	 <i>Azadirachta indica</i>
4	Ageratum	 <i>Ageratum houstonianum</i>

Materials and Methods

Collection of leaves of Lemongrass (*Cymbopogon citrates*), Neem, *Calotropis*, *Ageratum* were collected from the Department of Zoology & Department of Nutrition Isabella Thoburn College India.

The present study conducted all experiments on mosquito larvae of *Aedes aegypti* obtained from drainage water supply and stagnant water places from Department of Zoology, Isabella Thoburn College India.

200 grams of Lemongrass, Neem, *Calotropis*, *Ageratum* each leaf was taken and air dried for 1 day and the leaves were mashed and soaked into the water for 1 day, and the liquid was filtered using filter paper, the juice obtained is 100% natural extract with no added chemical/synthetics.

The 20 *Aedes* larvae were transferred into the container and three replicas were made for each leaf extract. 15 minutes were given to the larvae to get stable and adjust into the environment. Then in each container 3, 6, 9ml of each leaf extract were transferred with the help of dropper and the deaths of the larvae were counted and effectiveness of the leaf extract was noted. Control was taken in each.

Results

Statistical analysis

Mean mortality of *Aedes* larvae after exposure were expressed in terms of number in Table -1 After that values were allowed to make a comparison with control to find out the correct mortality of the *Aedes* larvae. In Table-2, result for %mean mortality is expressed which is highest in *Calotropis*. All the tables were done using excel spreadsheet. Data were submitted to one way analysis of variance using SPSS software. When the ANOVA statistics were significant ($p < .001$), the mean separation was performed by Dunnett multiple range test.

Calotropis extract mortality rate of *Aedes* larvae exposed to *Calotropis* at a concentration of 9ml was observed to be 100%. This indicates that *Calotropis* has a potent larvicidal effect on *Aedes* larvae, achieving complete eradication (Table-2). Extract of Lemongrass exposure on *Aedes* larvae at a concentration of 9ml resulted in mortality rate 28% (Table-2). Although the mortality rate was significantly lower compared to *Calotropis*, lemongrass still exhibit a notable larvicidal effect. Extract of Neem, A concentration on 9ml of neem led to mortality rate of 23.3% (Table-2) among *Aedes* larvae. Neem demonstrated moderate larvicidal activity against the larvae. *Ageratum* at a concentration of 9ml exhibited a mortality rate of 21.65% on *Aedes* larvae (Table-2). Although the efficiency was slightly lower as compared to neem *ageratum* still displayed substantial larvicidal properties.

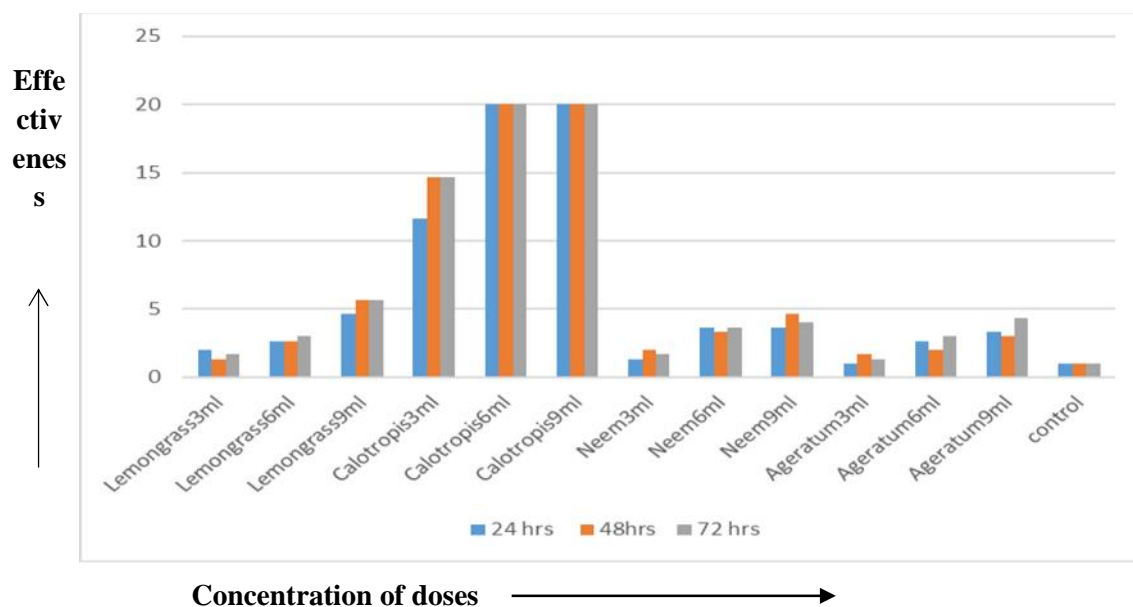
Table: 1-*Aedes* larva mortality after exposure to leaf extract

Mortality (Mean± S.E) after exposure to plant extract												
	Lemongrass			Calotropis			Neem			Ageratum		
Conc.	Day1	Day2	Day3	Day1	Day2	Day3	Day1	Day2	Day3	Day1	Day2	Day3
3ml	2.00±.00	1.33±.33	1.66±.33	11.66±.88	14.66±1.77	14.66±.88	1.33±.33	2.00±.57	1.66±.33	1.00±.00	1.66±.33	1.33±.33
6ml	2.66±.66	2.66±.33	3.00±.57	20.00±.00	20.00±.00	20.00±.00	3.66±.33	3.33±.66	3.66±.33	2.66±.33	2.00±.57	3.00±.00
9ml	4.66±.88	5.66±.33	5.66±.57	20.00±.00	20.00±.00	20.00±.00	3.66±.57	4.66±.88	4.00±.57	3.33±.33	3.00±.57	4.33±.33
Control	1.00±.00	1.00±.00	1.00±.00	1.00±.00	1.00±.00	1.00±.00	1.00±.00	1.00±.00	1.00±.00	1.00±.00	1.00±.00	1.00±.00

Table: 2 %Mean mortality of Aedes larvae after exposure to plant extracts

%Mean mortality of Aedes larvae after exposure to plant extract												
	Lemongrass			Calotropis			Neem			Ageratum		
Conc.	Day1	Day2	Day3	Day1	Day2	Day3	Day1	Day2	Day3	Day1	Day2	Day3
3ml	10%	6.65%	8.3%	58%	73.3%	73.3%	6.65%	48%	8.3%	5%	8.3%	6.65%
6ml	13.3%	13.3%	15%	100%	100%	100%	18.3%	16.65%	18.3%	13.3%	10%	15%
9ml	23%	28%	28%	100%	100%	100%	18.3%	23.3%	20%	16.65%	15%	21.65%
control	5%	5%	5%	5%	5%	5%	5%	5%	5%	5%	5%	5%

Graph: 1 –The above graph shows the mortality of *Aedes* larvae after exposure to the leaf extracts



Discussion

The average number of dead mosquito larvae was highest in *Calotropis*. One of the ways to control mosquito larvae is through the use of natural products like *Calotropis* (milkweed) as it holds bitter milky sap. The larvicidal capability of leaf extract of *Calotropis procera* has been inspected in case of larvae of the three mosquito species viz. *Anopheles stephensi*, *Culex quinquefasciatus*, and *Aedes aegypti* (Singh et.al, 2005). Various studies have been carried out with the latex of the plant. For the first time (Giridhar et al.1984) has described compounds with larvicidal pursuit in the latex of *Calotropis procera*. *Calotropis* is effective in controlling mosquito larvae because of its latex; the latex of *Calotropis* plant is toxic to mosquito larvae and can kill them.

Lemongrass is another natural plant which is effective in controlling mosquito larvae it contains citronella oil, which is natural insecticidal. The lemongrass extract can be extracted from leaf. The dried lemongrass extract demonstrates to be more agile such that within two hours of utilization, all the larvae were dead. This outcome is in agreement with earlier work done by (Sosan et al. 2013) which showed that

Cymbopogon citratus and *Ageratum conyzoides* used against *Aedes aegypti* did achieved 100% mortality at 120, 200, and 300 mg/L concentrations respectively. The biological activity of these plants' extracts might be due to various compounds, including phenolic, flavonoids.

Neem is another well liked genuine product which contains insecticidal characteristics like *Azadirachtin* which is noxious to mosquito. *Azadirachtin indica* has been reported to be eco-friendly and non-toxic to vertebrates. The plant crude or botanical extracts are less expensive and highly effective for the control of mosquitoes that contributes too many serious vectors borne diseases rather than purified compounds or extracts of the plant (Alouani et al, 2009; Khalafalla et al., 2007; Bhagavan and Rahuman, 2011)

Ageratum are small, bushy plant contain compound like coumarin, which possesses strong insect repelling properties. Numerous plants have been comprehensively studied for their insecticidal effects.

Phytochemicals such as phenolic acid, flavonoids are known for their mosquito repellent and insecticidal effects. *Ageratum houstonianum* essential oil and extracts have been declared to have bioactive molecules (El Hadidy et.al, 2019) with repellence and adulticidal steps in case of adult mosquitoes (Tennyson et.al, 2012)

Conclusion

The results of this study indicate that *Calotropis* possesses remarkable larvicidal activity against *Aedes* larvae, with a mortality rate of 100%. It suggests that *Calotropis* could be a valuable natural agent for controlling *Aedes* larvae. Lemongrass, Neem, *Ageratum* also exhibited significant larvicidal effects, albeit with lower mortality rates compared to *Calotropis*. These findings highlight the potential of these substances as alternative larvicidal agents in larvae control strategies.

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