



Calotropis Procera As Bio-Larvicide For The Control of Mosquito Vectors in Clean and Polluted Water Breeding Habitats

KEYWORDS

Calotropis procera, larvicidal, mosquito vectors, clean & polluted water breeding habitats

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ABSTRACT The larvicidal potential of crude and extracted latex of *Calotropis procera* was tested against mosquito vectors in clean and polluted water breeding habitats. Insecticide susceptibility tests were carried out using WHO standard. Analysis of Variance showed that extracted latex has good larvicidal effect as compared to aqueous latex at 100 ppm concentration. Statistically significant result ($P < .01$) was observed when we compared methanol extract versus aqueous latex ($t=5.2782, df=8$) and acetonitrile extract with aqueous latex ($t=5.9758, df=8$), similarly when efficacy of both the extracts was compared it showed non-significant difference ($P > .01$) and these two being comparable ($t=0.2558, df=8$). This showed that both the extracts are effective to control mosquito breeding in domestic & peri-domestic breeding containers as compared to aqueous latex. The finding of the study revealed that extracted latex of *C. procera* can be used as an alternative approach for the control of mosquito vectors in all breeding sites. The study would be of great importance in planning vector control strategy based on alternative plant derived insecticides.

Introduction

Mosquito control is essential to prevent transmission of vector borne diseases and to improve quality of public health and environment. Mosquito borne diseases are prevalent in more than 100 countries, infecting 300-500 million people and causing about 1 million deaths every year¹. Targeting the breeding sites of mosquitoes is an efficient and effective approach for the control of mosquitoes. During larval stages mosquitoes are less mobile and concentrated within smaller areas as compared to much larger areas required for targeting adult stages². Use of synthetic insecticides is one of the methods available for controlling the mosquitoes. Temephos and *Bacillus thuringiensis israelensis* (Bti) are commonly used for clean water habitats while Fenthion and Pirimiphos methyl are used for polluted water breeding habitats. Owing to development of genetic resistance to synthetic insecticides³ and to bio-pesticides such as *Bacillus thuringiensis* and its adverse effect on environment have been reported. Recently attention has been given to search plant based herbal compositions for managing mosquito population by human communities. Bio-compounds are known to possess several advantages over the synthetic chemical insecticides such as cost effective, highly potent, less toxic and biodegradable^{4&5}. *Calotropis procera* R. Br. (Asclepiadaceae) is one of the important weed plants used for the preparation of traditional medicines and its milky sap is used by folk healers to cure many diseases. Latex of the plant was reported to possess analgesic activity⁶, anti-inflammatory activity⁷, antimicrobial activity⁸, anti-diarrhoeal activity^{9&10}, antinociceptive activity and anti-malarial activity¹¹. The latex in the green parts of the plant is produced and accumulated as a defense strategy against organisms such as virus, fungi, and insects. *C. procera* is known to contain several phyto-chemical compounds such as calotropin, calotropagenin, calotoxin, galactin, uscharin, amyridin, amyridin esters, uscharidin, corologalactin, frugoside, corotoxinigenin, calotropagenin and voruscharin used in many therapeutic applications¹². The present study was carried out to demonstrate the utility of *Calotropis procera* extracted

latex for the control of mosquito population in clean as well as polluted water breeding habitats.

Material and Methods

Naturally grown plants were used for the collection of the latex (Fig-1, A) and were identified by Botanical Survey of India and deposited at National Bureau of Plant Genetic Resources with accession no IC-0597244 seeds, Pusa Campus, New Delhi. The latex was manually collected directly from the plant (Fig.1B) and extracted by organic solvents (AR grade) as per protocol developed¹³. The latex volume was measured and extraction solvent was added to it in 1:1 ratio (v/v). After 1 hour the mixture was filtered through Whatman filter paper no.1. The clear filtrate is collected in Petri dishes and left for air drying in a cool and dry place at room temperature till a dried layer of extract is left on the plates (Fig.1C). The dried extract was made into powder and stored at 4°C in refrigerator. Polluted water has been collected from different sources such as industrial waste, biological water from ponds and textile water.



Larvicidal bio-assay

The larvicidal efficacy of crude and extracted latex of *C. procera* was assayed by the WHO protocol¹⁴. 100 ppm dose has been tested and found effective in field breeding containers¹⁵. Larvicidal bioassays were undertaken at optimized dose of 100 ppm of aqueous, methanol and acetonitrile extract against mosquito vectors in clean,

textile, biological and industrial water. Batches of 20 larvae (5 replicates), a total of 100 larvae and control group were taken for experiments. Mortality was observed after 24 hours exposures and dead larvae were counted and the percentage mortality was recorded from the average of five replicates.

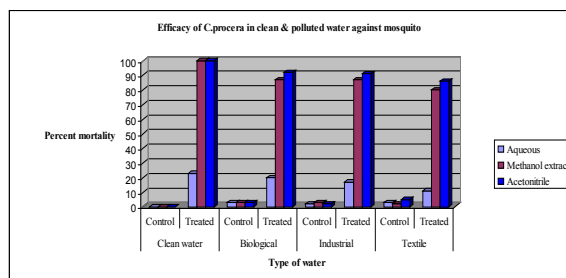
Statistical analysis- Analysis of variance (ANOVA) was applied to study efficacy of aqueous and extracted latex against mosquito vectors in clean and polluted water.

Results and Discussion

Methanol and acetonitrile extracted latex has shown good larvicidal effect as compared to aqueous latex at optimized dose of 100 ppm concentration in clean as well as polluted water against mosquitoes. The highest mortality was observed against vectors in clean water (100%), followed by 88% in biological water, 87% in industrial water and 80% in textile water; whereas in crude latex percentage of mortality was in order 23% in clean water, 20% in biological water, 17% in industrial water and 11% in textile water as shown (Fig.2). ANOVA analysis of extracts and aqueous latex in clean and polluted water against mosquito vectors showed that extracted latex is more effective in all types of water as compared to crude latex. Statistically significant difference ($P < .01$) was observed when we compared methanol extract versus aqueous latex ($t = 5.2782, df = 8$) and acetonitrile extract with aqueous latex ($t = 5.9758, df = 8$). Efficacy of methanol & acetonitrile extracted latex in clean and polluted water showed non significant differences $P > .01$ and these two being comparable ($t = 0.2558, df = 8$). It showed that both the extracts are effective to control mosquito breeding in both clean as well as polluted water habitats. Effectiveness of extracted latex at 100 ppm concentration against mosquitoes i.e. *Aedes*, *Anopheles* and *Culex* vectors encourages its application in domestic & peri-domestic breeding containers of clean water as well as dirty water bodies containing polluted water to control mosquito breeding. The extraction of bio-chemical compound depends on the polarity of solvent used and thus effect chemo-profile of plant. *C. procera* is a common weed plant growing in arid and semi arid part of Rajasthan throughout the year. The latex can easily be collected, extracted and applied to breeding places which do not require any special skill, instrument for its application. The result of our study has been supported by other workers as Girdhar et al¹⁶ has shown the larvicidal potential of whole latex of *C. procera* for mosquito control at 10,000 ppm. Singhi et al¹⁷⁻¹⁹ has shown larvicidal activity against three important vector causing dengue malaria and lymphatic filariasis and reported very important observations on refractoriness towards ovipositioning behavior of female mosquito up to 3 gonotrophic cycle. This behavioural change towards latex solution may help for the selection of bio-larvicide for mosquito control. Laboratory studies on methanol extract of leaf, seed and flower of *C. procera* as larvicide against *An. stephensi*, a vector of malaria has been studied. To improve quality of public health and environment, it becomes essential to search for alternatives to prevent transmission of vector borne diseases. Bioactive extracted chemical compounds from the plants could be used as alternatives to the currently used insecticides. The screening of locally available plants for mosquito control would reduce the dependence on expensive mosquito control devices and also stimulate local effort to enhance public health system. From the results, it can be concluded that *C. procera* extracts possess good larvicidal activity against mosquitoes in clean as well as polluted

water breeding sites and more studies are indicated to extract the active compounds for future studies and use in mosquito control.

Fig.2



Conclusion

C. procera shown good larvicidal potential even at low concentration (100 ppm) against mosquitoes in clean as well as polluted water breeding habitats. The plant grows in nature across road sides without any care & cost throughout the year especially when the mosquito population is higher. Due to high cost of synthetic chemical pesticides, natural chemistry of plants can be used for sustainable pest management with minimum environmental and health effect in future. In this regards latex of *Calotropis procera* act as promising bio larvicide.

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