

## Journal of Comprehensive Pharmacy

**Research Article**

Available Online at: [www.jcponline.in](http://www.jcponline.in)

ISSN NO: 2349-5669

### Larvicidal efficacy of plant extracts against the filarial vector mosquito *Culex quinquefasciatus*

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#### ARTICLE INFO

##### Article history:

Received 15 May 2014

Accepted 25 June 2014

Available online 21 August 2014

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#### Abstract

**Aim:** The View of this work was to study the larvicidal activity of dried root part of *Sida acuta* (Burn.) against the larvae of *Culex quinquefasciatus*. These larvae are the most significant vectors. Mosquitoes are the known major vector for the transmission of several diseases like filariasis, malaria, dengue fever, yellow fever, and Japanese encephalitis. Which are considered harmful towards the population in tropic and subtropical regions. The millions of people are suffering from filariasis around the world.

**Methods:** The preclusive laboratory was revealed that to determine the efficacy of Chloroform extract of dried root of *Sida acuta* (Burn.) belonging to the family Malvaceae at various concentrations against the late third instar larvae of *Culex quinquefasciatus* by following the WHO guidelines.

**Results:** The results suggest that 100% mortality effect of Chloroform extract of *Sida acuta* (Burn.) was practically observed at 50 to 300 ppm (parts per million). The results obviously showed use of plants in larvae control as an alternative method for minimizing the noxious effect of some pesticide compounds on the environment. Thus the extract of *Sida acuta* (Burn.) is claimed as more selective and biodegradable agent.

**Conclusion:** This study observed that plant of *Sida acuta* (Burn.) has a realistic mortality result for larvae of filarial vector. This is safe to individual and communities against mosquitoes. It is a natural arms for mosquito control.

**KEYWORDS:** *Culex quinquefasciatus*, *Sida acuta* (Burn.), larvicidal activity, filarial vector.

#### INTRODUCTION

Mosquito is the most indisputable medicinal significant arthropod vector of diseases. The vector-borne diseases caused by mosquito are one of the major health plights in most of the countries. It is affecting the socio economical status of many nations and it is a critical

pest against human causing allergy too. That includes a local skin reaction too [1]. They transmit parasites and pathogens which continue to have disadvantageous impact on human beings [2]. The dash diseases like filariasis, dengue, yellow fever, malaria, Japanese encephalitis and chikungunya are the deadly diseases spread by mosquitoes. *Culex quinquefasciatus* is an

important vector of *Bancroftian filariasis* in tropical and subtropical regions. According to WHO (1984) about 90 million people worldwide were once infected with *Wuchereria bancrofti*, the lymphatic dwelling parasite, and ten time more people are at the risk of being infected. In India alone is 25 million people harboring microfilaria and 19 million people suffer from filarial diseases [3]. Mosquito is frequently found due to inferior drainage system especially during rainy seasons, fish pond, and irrigation ditches and rice fields. This provides a better breeding place for mosquitoes [4]. There is provocative interest in research for larvicidal compound from natural sources. Even though chemical vector program has been carried on for long time, these mosquito vectors remain because of repeated use of synthetic products, house hold spray, and insecticides for mosquito control. As a result, the mosquito develops their resistance [5]. Hence, there is a need for developing biologically active natural chemical constituents which act as a larvicidal and promising to reduce the risk to humans and harmful accumulated residues [2, 6]. This has necessitated the need for research and development of environmentally safe, biodegradable, and low cost indigenous method for vector control, which can be used with minimum care by individuals and communities in specific situation [7]. The plant *Sida acuta* (Burn.) is described in Ayurveda and Siddha as a potent drug. It is traditionally used for the health disorders like cough, stomachic, tonic, diuretic, antipyretic, nervous & urinary disorder, testicular swelling as well as elephantiasis. [8–10]. The petroleum ether extract was found to possess estrogenic activity and anti-implantation activity [11]. The leaf extract of *Sida acuta* has proven antibacterial and anthelmintic activity [12]. The plant of *Sida acuta* (Burn.) is used for various medicinal purposes also such as liver disorder, abortifacient, asthma, fever, headache (migraine), could ulcer, snake bite urinary diseases and anti-fertility activity [12]. The leaf has reported as pharmacologically significant on Antimicrobial activity [13]. The root has properties like aphrodisiac, anti-rheumatic, stomachic and wound healing [14]. Besides the claim of larvicidal property of *Sida acuta* there is no scientific evidence to prove this claim. Hence the present study has been attempted to make a survey of root of the plant extract of *Sida acuta* (Burn.) in a view that the root extract may found most of its biological value when compared to other parts, in the context of integrated vector control management.

The results of the present study would be useful in promoting research. The goal to development of new agent's for mosquito control based on bioactive from of indigenous plant source.

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## MATERIALS AND METHODS

### Collection of a Plant Material:

The plant was collected during flowering stage in the month of July-August from Nilgris. Then, their identification was established with the aid of an experienced botanist Dr. S. Rajan and compared with herbarium sheets of the authentic sample. Many of defensive components are biodegradable with non residual effect on the biological environment; hence, an attempt has been made in present investigation to identify plant with potential to control vector mosquitoes.

### Preparation of Extraction:

The dried root part of the plant of *Sida acuta* (Burn) was powdered and extracted by Soxhlet with chloroform. The extracts were concentrated under the rotary vacuum at evaporator until the complete solvents evaporated at (>45°C) to get semisolid mass of crude extracts [15]. And then use freeze dried (-80°C) to obtain solid residue [16]. These extracts were used for determining the larvicidal activity against mosquito larvae of bancroftian filariasis vector mosquito *Culex quinquefasciatus* [17, 18].

### Larvicidal Bioassay:

Larvicidal activity was evaluated in concurrence with WHO standard with slight modification being done for study [19]. The larvae of *Culex quinquefasciatus* were obtained and reared from the neonatal mosquito in the National Institute of Communicable Disease, Southern India branch field station. This located at Mettupalayam, Coimbatore District, Tamil Nadu. They were maintained at the temperature of  $28 \pm 2^\circ\text{C}$  and  $80 \pm 10\%$  RH (relative humidity) under the 12-hour light and dark photoperiod cycle. The larvae were fed dog biscuit and a brewer's yeast powder mixture 3: 1 ratio is used in the laboratory. After five days, adult male mosquitoes were fed a 10% of sucrose solution. The emerging female mosquitoes obtain blood meal from white albino rat for 2-3 h for eggs production [20]. A stock solution was prepared and stored in a refrigerator at  $15^\circ\text{C}$ . The 25 healthy late third instar larvae were collected and to compose of chloroform extracts in the 500 mL beaker (sterilized glass and plastic beaker)

each of which was conducted in triplicate. 25 larvae were collected with a pasture pipette, placed on a filter paper for removal of excess of water and placed in 250mL de-chlorinated tap water containing various concentrations of crude extracts. Three controls in triplicate were set up; with distilled water (250 mL). The beakers were covered with muslin cloth to avoid entry of any foreign material. After exposure the larvae in the extract were observed and outright recorded. In between the experiment, no food was ceded to larvae. At the end of 24h, the observed mortality (crude mortality) was recorded. There is no sign of any movement even after mild touch with a glass rod [16] and dead larvae are to be counted as described in the WHO technique report series. From this crude mortality, percentage of crude mortality was obtained. Subsequently, control mortality, if any, was recorded and percentage of crude mortality was obtained. The percentage of crude mortality was corrected by Abbot's formula. The corrected probit mortality and expected mortality were also obtained.

#### Statistical Analysis:

The average larval mortality data were subjected to probit analysis for calculating LC50 and LC90 (lethal concentration) values and their 95% confidence limits were estimated by fitting a probit regression model to the observed relationship between percentage of mortality of larvae and logarithmic concentration of the substance. Separate probit models were fitted for each extract [21]. All of the analysis was carried out using the SPSS (Statistical Package Social Science) software version 13.0.

## RESULTS

The chloroform extracts of *Sida acuta* (Burn.) were screened for larvicidal bioassay using water as a vehicle, respectively. The procedure followed for determining larvicidal is the same as described by WHO. The experiments were conducted by using 25 of late third instar larvae for each test solution, keeping a minimum of three replicates and sufficient controls. A total number of 75 larvae were exposed for each concentration for each extract. Six different concentrations of test solution ranging from 50 to 300 ppm were kept for chloroform extract. The observed and expected mortalities of larvae based on probit regression analysis for different concentrations of *Sida acuta* (Burn.) are shown in Table 1. The estimated LC<sub>50</sub> and LC<sub>90</sub> values (95% confidence intervals) were 149.8 (139.9- 159.6) and 231.4 (217.6-249.0) respectively. The potency of *Sida acuta* (Burn.) is efficacious with

chloroform extract. These plants have a few important chemical constituents like carbohydrates, alkaloids, glycosides, proteins, saponins, anthocyanins, phenolic compounds, flavones, steroids [11] and tannins [12]. This component has potential larvicidal activity due to individual or synergistic effects on the larvae. This needs to be a further deep research to identify the effective constituents for controlling mosquitoes. The results were considered to be statistically significant as given in Table 1.

## DISCUSSION

Globally, there is a prompt awareness going on and always desired to use natural, eco-friendly compounds for larvicidal activity [22]. Mosquito risk has become more acute in recent time and the death of millions of people every year due to mosquito-borne diseases has resulted in the loss of socioeconomic wealth in many countries. The control of mosquito by chemical substance is not safe at present because of insecticide resistance by vectors and environmental imbalance. Application of chemical or synthetic insecticides leads to deleterious effects in the long term; hence it does not provide absolute results. That is why alternative mosquito control method is needed [23]. The extract which is obtained from plant parts like leaves, root, flower, bark, seed, and fruits in their crude extracts has been used as conventional larvicidal [22]. The secondary compounds of plants are vast repository of compounds with a wide range of biological activities. Tennyson [15] studied the larvicidal activity of twenty-five plant extracts against the larvae of *Culex quinquefasciatus*. Likewise, Pavela [24] studied the larvicidal activity of thirty one Euro-Asiatic methanolic extracts against the larvae *Culex quinquefasciatus*. The leaf extract of *Mesua ferra* [25], fruit extract of *Croton caudatus*, flower extract of *Tiliacora acuminata* [26], leaf extract of *Typhonium trilobatum* [27] and flower extract of *Tagetes erecta* [28] were found to cause larval mortality to *Culex quinquefasciatus*. In this study, the petroleum ether and N-butanol extracts of *Cassia occidentalis* (Linn.) whole plant exhibited a dose dependent activity which is similar to other studies which have also reported dose dependency of plant against mosquito larvae [29-31]. The mechanism of action exhibited by *Sida acuta* (Burn.) root part of the plant may possibly to its toxic effects on the larvae. This is supported by a previous study which reported that an ethanol extract of *P. Pinnata* exhibited toxic effects in the Swiss albino mice [32]. Other studies also showed clearly that the chloroform and methanolic

**Table 1: Observed and expected mortality of *Culex quinquefasciatus* larvae exposed to *Sida acuta* with chloroform extract. Expected mortality is based on probit regression analysis.**

Conc. (µg/ml)	No. of larvae		Mortality (%)		Expected Mortality			Probit (mortality)	$\chi^2$	LC <sub>50</sub>	LC <sub>90</sub>
	Exposed	Dead	Crude	Corrected	Probit	Dead	%	= a + b x conc	D.F	(95 % CI)	(95 % CI)
<i>Sida acuta</i> with chloroform extract											
50	75	6	8.0	8.0	-1.57	4.4	5.8	-2.3532+ 0.0157 x conc	$\chi^2 = 3.07$	149.8	231.4
100	75	15	20.0	20.0	-0.78	16.3	21.7		D.F.=4	(139.9-159.6)	(217.6-249.0)
150	75	38	50.7	50.7	0.00	37.6	50.1		P=0.5		
200	75	55	73.4	73.4	0.79	58.8	78.4				
250	75	72	96.0	96.0	1.57	70.7	94.2				
300	75	75	100.0	100.0	2.36	74.3	99.1				

D.F. = Degrees of freedom

extracts of *Tagetes minuta* L. have a pronounced larvicidal effect on mosquito *Anopheles stephensi* which is due to the presence of Phytochemicals such as flavonoids, phenols, terpenoids, and saponins. Thus, the above statement may potentiate that the shown larvicidal activity of *Sida acuta* (Burn.) root part of the plant may also be due to presence of alkaloid, flavonoids, phenols, glycosides, and tannins. Hence the present study, the authors concluded that the chloroform extracts of *Sida acuta* (Burn.) root part of plant extract have larvicidal activities.

## CONCLUSION

On the basis of the above results, we can conclude that *Sida acuta* (Burn.) has a paramount larvicidal importance. The synthetic chemical can be obtained easily at a very low cost. But the use of the plants for larvae control offers a safer alternative also. Besides, these results could be useful in the search for newer compounds. These extracts are inexpensive, easy to handle, and safer products for the control of mosquito larvae.

## ACKNOWLEDGMENTS

The authors are thankful to the J.S.S. College of Pharmacy, Ooty, Tamil Nadu, and the National Institute of Communicable Diseases, Coonoor, Southern India branch field station located at Mettupalayam (District Coimbatore of Tamil Nadu State), for providing all necessary facilities which were required for completing this research work.

## Conflict of Interests

The authors declare that they have no conflict of interests regarding the publication of this paper.

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**Cite this Article as:** Kumar D, Chawla R, Perumalsamireddy D. Larvicidal efficacy of plant extracts against the filarial vector mosquito *Culex quinquefasciatus*. J Compr Phar 2014;1(2):31-36.