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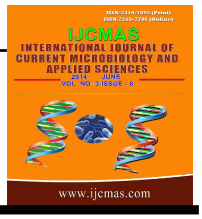

Original Research Article

Mosquito larvicidal properties of commiphora caudate (Wight & Arn.) (Bursaceae) against *Aedes aegypti* (Linn.), *Anopheles stephensi* (Liston), *Culex quinquefasciatus* (Say)

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Keywords

Commiphora caudata, *Aedes aegypti, Anopheles stephensi* and *Culex quinquefasciatus,* Larvicidal activities.

A B S T R A C T

Mosquitoes transmit serious human diseases, causing millions of deaths every year. Natural products of plant origin with insecticidal properties have been used in recent year for control of a variety of pest insects and vectors. The present study was carried out on the larvicidal activity of three selective insidious medicinal plants - *Commiphora caudata* (Wight & Arn.), against third instar larvae of *Aedes aegypti, Anopheles stephensi* and *Culex quinquefaciatus*. Larvicidal activities were conducted at different concentration (50-250 ppm) of ethyl acetate, hexane, chloroform and acetone leaf extracts of the plants. The mortality was recorded after 24 hrs exposure and LC_{50} and LC_{90} were determined. The present investigation revealed that the LC_{50} and LC_{90} value of ethyl acetate, hexane, chloroform and acetone extract of *C. caudate* against *Aedes aegypti* larvae in 24 hrs were 97.19, 112.85, 99.17 and 109.67, 105.59, 138.36, 108.89 and 126.33mg/L respectively. The LC_{50} and LC_{90} value of *Anopheles stephensi* were 96.04, 104.16, 97.13 and 106.53, 104.44, 118.37, 105.50 and 121.81 mg/L for ethyl acetate, chloroform, hexane and acetone, respectively. For *Culex quinquefaciatus*, the calculated LC_{50} and LC_{90} values were 94.76, 102.95, 95.98 and 105.09, 103.10, 116.12, 104.75 and 119.08 mg/L, for ethyl acetate, chloroform, hexane and acetone, respectively. It is concluded that the highest larvicidal activity against *Cx. quinquefasciatus* was obtained with ethyl acetate extract of *C. caudate* followed by *A. squarrosa* and *C. alata*.

Introduction

Mosquitoes represent a significant threat to human health because of their ability to vector pathogens that cause diseases that afflict millions of people worldwide (WHO 2010). Several species belonging to genera Aedes, Anopheles and Culex are vectors for the pathogen of various diseases like dengue fever, dengue hemorrhagic fever, malaria, Japanese encephalitis and filariasis (Borah et al., 2010; Rahuman 2009; Samuel 2010). *Aedes aegypti* is known to carry dengue and yellow fever; malaria is carried by *Anopheles stephensi*; and filarial disease by Culex. The dengue fever incidence has increased fourfold since 1970 and nearly half the world's population is now at risk. In 1990, almost 30% of the world population, 1.5 billion people, lived in regions where the estimated risk of dengue transmission was greater than 50% (Hales et al., 2002). An outbreak of Chikungunya virus disease emerged in the southwest Indian Ocean islands in 2005, spread out

to India, and resulted in an ongoing outbreak that has involved 1. 5 million patients, including travelers who have visited these areas (Taubitz et al., 2007).

Anopheles stephensi are major malaria vectors in India. With an annual incidence of 300-500 million, malaria is still one of the most important communicable diseases. Currently, about 40% of the world's population live in areas where malaria is endemic (Wernsdorfer and Wernsdorfer, 2003). Culex quinquefasciatus, a vector of lymphatic filariasis, is widely distributed in tropical zones with around 120 million people infected worldwide and 44 million people having common chronic manifestation (Bernhard et al., 2003).The present proliferation of these diseases is not only due to higher number of breeding places in urban agglomeration, but also due to increasing resistance of mosquitoes to current commercial insecticides such asorganochlorides, organophosphates, carbamates and also to biological insecticides (Goettel et al., 1992; Das and Amalraj, 1997; Yadav et al., 1997).

Many studies on plant extract against mosquito larvae have been conducted in the region of the World. Plants may be a source of alternative agents for control of mosquitoes because they are rich in bioactive chemicals, are active against a limited number of species including specific target insects, and are biodegradable. They are potentially suitable for use in integrated pest management programs (Alkofahi et al., 1989, Dharmshaktu et al., 1987, Green et al., 1991). Two of such plants C. caudata and C. var pubescens (CP) were selected to explore the potent bioactive compounds for its antioxidant activity to discover the actual value of folkloric remedies. C. caudate (Arn) Engl (Bursaceae) commonly known as "hill mango" is a moderate sized tree/shrub occurring in dry forest and commonly cultivated as an avenue tree (Nayar 1956) found throughout South India. C. var pubescens (Wight & Arn) belongs to the same family a variety of Commiphora species, a middle sized tree/shrub (Gamble 1997) which also grows in the dry forest of South India. Its gum resin from the bark is used for treating stomach troubles (Latha et al., 2005). In this context, the purpose of the present investigation is to explore the larvicidal properties of C. caudate leaf extract and against Chikungunya vector, *A. aegypti*, *An. stephensi* and *Cx. quinquefaciatus* under the laboratory conditions.

Materials and Methods

Collection and Identification of Plant material

The medicinal plant, *C. caudate* was collected from Muthupet in Tamilnadu, India. Bulk samples were air-dried in the shade. After drying, these were ground to fine powder. At the time of collection, voucher herbarium specimens were prepared and identified with the help of Plant Taxonomist, Department of Botany, Annamalai University, and Chidambaram.

Extraction method

The dried leaves (100g) were powdered mechanically using commercial stainless steel Blender and extracted sequentially with ethyl acetate, hexane, chloroform and acetone (500 ml, Ranchem), in a Soxhlet apparatus separately until exhaustion. The extract was concentrated under reduced pressure of 22-26 mm Hg at 45^{o}C by 'Rotavapour' and the residue obtained was

stored at 4°C in an amber vial. Then the vials were named and covered with silver foil and transported to the laboratory. Until use those vials were kept in cool and dark place at 4°C.

Larvicidal activity

The larvicidal activity of crude extract was evaluated as per the protocol previously described by WHO (2005). From the stock solution, six different test concentrations (50, 100, 150, 200, and 250 mg/l) were prepared and tested against the freshly molted (0–6 h) III instar larvae of *A. aegypti*, *An. stephensi* and *Cx. quinquefaciatus*.

The test medium (500 ml plastic cups) was prepared by adding 1 ml of appropriate dilution of test concentrations and mixed with 249 ml of dechlorinated water to make up 250 ml of test solution. The larvae were fed with dry yeast powder on the water surface (50 mg/l). The control (without plant extracts) experiments were also run parallel with each replicate. For each experiment, four replicates were maintained at a time. A minimum of 25 larvae per concentration was used for all the experiments. The larval mortality was observed and recorded after 24 h post-treatment. Percent mortality was corrected for control mortality using Abbott's formula (1925).

Statistical analysis

The larval mortality data were subjected to probit analysis for calculating LC_{50}, LC_{90} (Finney 1971) and other statistics at 95% confidence limits of upper confidence limit and lower confidence limit values were calculated using the SPSS software package 12.0 (2007). Results with $p \leq 0.05$ were considered to be statistically significant.

Results and Discussion

Larvicidal activity of ethyl acetate, hexane, chloroform and acetone crude leaf extracts of *C. caudate* are shown in Table 1 & 2. As evidenced from the table, generally increased larval mortality was observed with increased concentration of the extracts tested against *A. aegypti, An. stephensi* and *Cx. quinquefaciatus*. The present investigation revealed that the LC_{50} value of ethyl acetate, chloroform, hexane and acetone extract of *C. caudata* against *A. aegypti* larvae in 24 hrs were 94.76, 102.95, 95.98 and 105.09 mg/L; LC_{90} value of 103.10, 116.12, 104.75 and 119.18 mg/L, respectively. The LC_{50} value of *An. stephensi* were 94.04, 104.16, 97.13 and 106.53 mg/L; LC_{90} value of 104.44, 118.37, 105.50 and 121.81 mg/L for ethyl acetate, chloroform, hexane and acetone, respectively.

For *Cx. quinquefaciatus*, the calculated LC_{50} values of 97.19, 112.85, 99.17 and 109.67 mg/L; LC_{90} values of 105.59, 138.36, 108.89 and 126.33 mg/L for ethyl acetate, chloroform, hexane and acetone, respectively (Table 1), Reported in the plant suggesting their use in larvicidal population control (Table 2). It is concluded that the highest larvicidal activity against *Cx. quinquefasciatus* was obtained with ethyl acetate extract of *C. caudate*. This result is also comparable to earlier reports of Dhanasekaran *et al.*, (2013) have that the LC_{50} of ethanol crude extracts of selected indigenous medicinal plants were 82.86ppm, 89.45ppm, 109.37ppm 109.87ppm and 172.31 respectively, against the malarial vector, *An. stephensi*, dengue vector *Ae. aegypti*, Japanese Encephalitis vector, *Culex tritaeniorhynchus*. Gokulakrishnan *et al.*, (2012) reported that the larvicidal and Ovicidal efficacy of different solvent leaf extract of *Ariitolochia indica* against

Anopheles stephensi. The hatch rates were assessed 48 h after treatment. The LC_{50} and LC_{90} values of Acetone, Benzene, Chloroform, Hexane and Methanol extracts of *A .indica*against *An. Stephensi* larvae in 24 h were 76.29, 58.82, 53.59, 65.84, 51.78 and 205.85, 193.23, 185.16, 196.72 and 181.00 ppm, respectively. Kaliyamoorthy krishnappa *et al.,* (2012) has ethanol extracts of *Gliricidia sepium* showed larvicidal showed LC_{50} and LC_{90} value of 121.79 ppm and 231.98 respectively, against the malarial vector, *An. Stephens*.

Elangovan *et al.,* (a) have leaf extracts of *Corchorus capsularis* the against common malarial vector, *Anopheles stephensi* (LC_{50} and LC_{90}values of 197.34, 205.48, 176.19 and 358.59, 363.42, 334.56 ppm), against dengue vector *Ae. aegypti* (LC_{50} and LC_{90} values of 222.45, 190.52, 182.06 and 383.06, 354.84, 306.81 ppm), respectively. Mathivanan *et al.*, (2010) reported that the LC50 and LC90 values of crude methanol extract of leaves of *E.coronaria* on *Cx. quinquefasciatus, Ae. aegypti* and *An. stephensi* larvae in 24 h were 72.41, 65.67, 62.08 and 136.55, 127.24 and 120.86 mg/l, respectively. Significance level was set at $p < 0.05$. The larvicidal and ovicidal, activities of crude benzene and ethyl acetate extracts of leaf of *Ervatamia coronaria* and *Caesalpinia pulcherrima* were assayed for their toxicity against three important vector mosquitoes, *viz., Anopheles stephensi, Aedes aegypti,* and *Culex quinquefasciatus* (Diptera: Culicidae). Elumalai *et al*., (2012) have that mosquitocidal activity of *Abrus precatorius* against chickungunya vector, *Ae. aegypti* (LC_{50} and LC_{90} values of 264.57ppm and 500.76ppm), against the Japanese Encephalitis vector, *Culex tritaeniorhynchus* (LC_{50} and LC_{90} values of 257.73ppm and 496.94ppm), respectively. Baluselvakumar *et al.,* (2012) have plant extracts of *Oxystelma esculentum* against *Anopheles stephensi* (LC_{50} and LC_{90} values of 75.46, 68.55, 98.47, 88.24, 63.84 and 140.66, 130.65, 184.10, 169.36 and 122.48 ppm,) respectively.

Elangovan *et al*., (2012) have that the larvicidal and ovicidal activity of *Exacum pedunculatum* against the common malarial vector, *Anopheles stephensi* (LC_{50} and LC_{90} values of 127.45, 121.39, 151.96 and 121.24). Mullai and Jabanesan (2007) have leaf extracts of *C. colocynthis* and *Cucurbita maxima* showed LC_{50} values of 47.58, 66.92 and 118.74 ppm and 75.91, 117.73 and 171.64 ppm, respectively, against *Cx. quinquefasciatus* larvae. Cavalcanti *et al.,* (2004) reported that the larvicidal activity of essential oils of Brazilian plants against *Ae. aegypti* and observed the LC50 to range from 60 to 533 ppm. Karunamoorthi and ilango (2010) have that the LC_{50} and LC_{90} of a methanol leaf extract of *Croton macrostachyus* (*C. macrostachyus*) were 89.25 and 224.98 ppm, respectively against late third instar larvae of the malaria vector, *Anopheles arabiensis* (*A. arabiensis).* Rajkumar and Jebanesan, (2002) who observed that increase in the concentration of leaf extract of *S. aerianthum* induced the oviposition attractant activity in *Cx. quinquefasciatus*. it may concluded that natural products as extracts from parts of plants insecticidal and medicinal values have higher efficiency in reducing mosquito menace due to their larvicidal toxicity. Further studies on the screening, isolated and purification of bioactive phytochemical constituents/compounds followed by in-depth laboratory and field bioassays are needed as the present study shows that there is scope to use *C. caudate* leaf extracts to control the immature stages of vector mosquitoes.

Table.1 Probit analysis of larvicidal activity of *C. caudate* extracts against *A. aegypti*, *An. stephensi* and *Cx. quinquefaciatus*

Species	Extract	LC_{50} (mg/L)	95%Confidence limits		LC_{90} (mg/L)	95%Confidence limits		Regression
			LCL	UCL		LCL	UCL	
A. aegypti	Ethyl acetate	97.19	95.16	99.27	105.59	103.56	107.66	Y=35.61×-65.78
	Hexane	112.85	107.90	118.02	138.36	133.03	143.91	Y=14.47×-24.70
	Chloroform	99.17	96.88	101.51	108.89	106.60	111.22	Y=31.56×-58.02
	Acetone	109.67	106.23	113.23	126.33	122.79	129.97	Y=20.87×-37.58
An. stephensi	Ethyl acetate	96.04	93.98	98.15	104.44	102.39	106.53	Y=35.21×-64.80
	Hexane	104.16	101.05	107.38	118.37	115.22	121.60	Y=23.08×-41.57
	Chloroform	97.13	95.09	99.20	105.50	103.47	107.56	Y=35.69×-65.93
	Acetone	106.53	103.27	109.90	121.81	118.49	125.22	Y=22.01×-39.66
Cx. quinquefaciatus	Ethyl acetate	94.76	92.68	96.88	103.10	101.03	105.61	Y=34.97×-64.13
	Hexane	102.95	100.02	105.97	116.12	113.16	119.15	Y=24.52×-44.34
	Chloroform	95.98	93.92	98.08	104.75	102.70	106.84	Y=35.29×-64.95
	Acetone	105.09	102.03	108.25	119.18	116.08	122.36	Y=23.45×-42.42

LC_{50}= Lethal concentration that kills 50% of the exposed parasite, LC_{90}= Lethal concentration that kills 90% of the exposed parasite. LCL- Lower Confident Limit, UCL- Upper Confident Limit.

Table.2 Larvicidal activity of *C. caudate* extracts against *A. aegypti*, *An. stephensi* and *Cx. quinquefaciatus*

Mosquitoes	Solvents	Mortality (%) ± SD				
		50ppm	100ppm	150ppm	200ppm	250ppm
A. aegypti	Ethyl acetate	21.75±1.63	42.25±1.41	59.5±2.82	80.75±2.87	100.00±1.15
	Hexane	7.75±1.25	15.25±2.06	24.75±2.51	33.25±1.63	41.25±2.87
	Chloroform	15.75±1.73	32.25±2.44	53.25±2.51	66.5±1.73	88.25±2.21
	Acetone	4.5±1.25	11.75±1.73	20.5±1.41	36.25±2.87	55.75±3.31
An. stephensi	Ethyl acetate	23.5±1.73	44.5±2.64	64.75±1.25	85.5±2.06	100.00±1.89
	Hexane	11.75±1.63	20.5±2.44	32.25±1.41	47.25±2.51	65.00±2.06
	Chloroform	21.5±1.63	40.25±2.87	60.25±1.5	83.5±1.41	100.00±0.95
	Acetone	8.25±1.25	15.5±0.81	27.25±2.06	43.75±0.81	60.25±1.63
Cx. quinquefaciatus	Ethyl acetate	28.25±1.25	47.25±2.64	67.5±1.25	96.25±2.06	100.00±1.41
	Hexane	12.25±1.63	23.75±1.25	36.5±1.63	51.25±2.44	67.5±2.06
	Chloroform	23.75±1.73	43.25±1.73	65.25±1.25	88.00±1.5	100.0±1.41
	Acetone	8.5±0.95	19.5±2.06	31.75±2.44	47.25±1.73	64.5±2.51

Significant at P>0.05 level, SD- Standard Deviation, PPM- Parts Per Million.

During the present study, the Ethyl acetate and chloroform extract of leaves of *C.caudata* showed significantly higher larvicidal activity against *Ae.aegypti, An. stephensi* and *Cx.quinquefasciatus*. The hexane and acetone extracts of other parts of this plant were weak larvicidal.

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References

Abbott WS (1925). A method of computing the effectiveness of an insecticide. J Eco Entomol; 18: 265-267.

Alkofahi A, Rupprecht JK, Anderson JE, Mclaughlin JL, Mikolajczak KL, Scott BA (1989). Search for new pesticides from higher plants. In: Arnason JT, Philogene BJR, Morand P (Eds) Insecticides of Plant Origin. In: ACS Sym. Ser, 387. Am Chem Soc, Washington, DC; 25–43.

Balaselvakumar, Gokulakrishnan J, Elumalai K, Dhanasekaran S, Anandan A, Krishnappa K (2012). Mosquito larvicidal activity of Oxystelma esculentum plant extracts against Anopheles stephensi (Diptera: Culicidae). Int. J. Recent. Sci. Res; 3 (5): 321-324.

Bernhard L, Bernhard P and Magnussen P (2003) Management of patient with lymphoedema caused by filariasis in northeastern Tanzania: alternative approaches. Physiothe. 89(12): 743-749.

Borah R, Kalita MC, Kar A, Talukdar AK (2010). Larvicidal efficacy of Toddalia asiatica (Linn.) Lam against two mosquito vectors Aedes aegypti and Culex quinquefasciatus. African J Biotechnol; 9 (16): 2527-2530.

Cavalcanti ESP, Morais SM, Lima MA and Santana EW (2004) Larvicidal activity of essential oils from Brazilian plants against Aedes aegypti L. Mem. Inst. Oswal. Cruz. 99: 541-544.

Das, P.K., Amalraj, D., (1997). Biological control of malaria vectors. Indian Journal of Medical Research 106, 174–197.

Dhanasekaran S, Krishnappa K, Anandan A and Elumalai K. J (2013). Agri. Technology; 9 (1): 29-47.

Dharmshaktu NS, Prabhakaran PK, Menon PK (1987). Laboratory study of the mosquito larvicidal properties of leaf and seed extract of plant Agava Americana. J Trop Med Hyg; 90: 79–82.

Elangovan A, Dhanasekaran S, Anandan A, Krishnappa K, Gokulakrishnan J and Elumalai K (2012) Larvicidal and ovicidal activities of Exacum pedunculatum (Linn.) (Gentinaceae) against a common malarial vector, Anopheles stephensi Liston (Diptera: Culicidae). Int. J. Rec. Sci. Res. 3(6): 559 – 563.

Elumalai K, Dhanasekaran S, Anandan A, Krishnappa K, Gokulakrishnan J and Elangovan A (2012). Int. J. curr. agricul. SCI; 2 (6): 31-38.

Finney DJ (1971) A ststistical treatment of the sigmoid response curve. In: Probit analysis. Cambridge University Press, London.633.

Gamble JS (1997). Flora of the Presidency of Madras vol-II, Bishen Singh, Mahendra Pal Singh, Dehradun, India.

Goettel, M.S., Toohey, M.K., Ram, R.C., Pillae, J.S., (1992). Field evaluation of Bti against Aedes vigilax and Culex sitiens in Fiji. Mosquito News 42, 277–278.

Gokulakrishnan J, Balu Selvakumar, Elumalai K and Krishnappa K (2012) Mosquito larvicidal and ovicidal efficacy of Ariitolochia indica Linn (Aristolochiaceae) leaf extracts against malarial vector mosquito Anopheles stephensiListon (Diptera: Culicidae). Int. J. Curr. Lif. Sci.2 (10): 48-52.

Green MM, Singer JM, Sutherland DJ, Hibbon CR (1991). Larvicidal activity of Tagetes minuta (Marigold) towards Aedes aegypti. J Am Mosq Control Assoc; 7: 282–286.

Guidelines for laboratory and field testing of mosquito larvi-cides. WHO/CDS/WHOPES/GCDPP/ 2005.13.

Hales S, Wet ND, Maindonald J, Woodward A (2002). Potential effect of population and climate changes on global distribution of dengue fever: an empirical model. Lancet; 360: 830-4.

Kaliyamoorthy Krishnappa (2012), Shanmugam Dhanasekaran, Kuppusamy Elumalai. Asian J. trop. med; 598-604.

.Karunamoorthi K, Ilango K (2010). Larvicidal activity of Cymbopogon citratus (DC) Stapf, and Croton macrostachyus Del. against Anopheles arabiensis Patton, a potent malaria vector. Eur Rev Med Pharmacol Sci; 14(1):57-62.

Latha S, Selvamani P, Sen DJ, Gupta JK, Pal TK, Gosh AK. (2005) .Antibacterial activity of Commiphora caudata and Commiphora berryi leaves. Ind. Drugs 42: 696-698.

Mathivanan T, Govindarajan M, Elumalai K, Krishnappa K and Ananthan A (2010) Mosquito larvicidal and phytochemical properties of Ervatamia coronaria Stapf. (Family: Apocynaceae). J. Vec. Bor. Dis. 47: 178–180.

Nayar SL (1956) .Glossary of Indian Medicinal Plants II, CSIR, New Delhi, p. 330.

Mullai K, Jebanesan A (2007). Larvicidal, ovicidal and repellent activities of the leaf extract of two Cucurbitacious plants against filarial vector Culex quinquefasciatus (Say) (Diptera: Culicidae). Trop Biomed; 24 (1): 1-6.

Rahuman AA, Bagavan A, Kamaraj C, Saravanan E, Zahir AA, et al (2009) Efficacy of larvicidal botanical extracts against culex quinquefasciatus say (Diptera: Culicidae). Parasitol Res 104: 1365-1372.

Rajkumar S and Jebanesan A (2002) Oviposition attractancy of Solanum aerianthum leaf extract for Culex quinquefasciatus. J. Expe. Zoo. India. 5(2): 221–224.

Samuel T (2010). Studies on the mosquitocidal activity of Ageratum houstonianum Mill. (Asteraceae) against vector mosquitoes [PhD Thesis]. Tamilnadu, India: University of Madras.

SPSS. SPSS for windows (2007), version 11.5. Chirago, IL: SPSS.

Werndorfer G, Wernsdorfer WH (2003). Malaria at the turn of the 2nd to the 3rd millennium. Wien Klin Wochenschr; 115: 2-9.

WHO (2010). Malaria fact sheets No. 94. WHO Report. Geneva: WHO media center.

Yadav, R.S., Sharma, V.P., Upadhyay, A.K., (1997). Field trial of Bacillus sphaericus B-101 (serotype H5a, 5b) against filariasis and Japanese Encephalitis vectors in India. Journal of American Mosquito Control Association 13, 158–163.