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Adulticidal efficacy of *Delonix elata* against filariasis vector mosquito, *Culex quinquefasciatus* (Diptera: Culicidae)

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PEER REVIEW

Peer reviewer

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Comments

The research work is very much important for society to control morbidity and other defects caused by mosquitoes and other insect vectors. This work reports a novel approach for the control of filariasis vector mosquitoes.

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ABSTRACT

Objective: To evaluate the adulticidal activity and toxicity of different solvent crude extracts of *Delonix elata* (*D. elata*) against filariasis vector mosquito *Culex quinquefasciatus* (*Cx. quinquefasciatus*).

Methods: The adulticidal activities of crude hexane, benzene, ethyl acetate, chloroform and methanol leaf and seed extracts of *D. elata* were assayed for their toxicity against vector mosquito *Cx. quinquefasciatus*. Bioassay was carried out by WHO method for determination of adulticidal activity against mosquitoes. The adult mortality was observed after 24 h of exposure.

Results: All extracts showed moderate adulticidal effects; however, the highest adult mortality was found in methanol extract of *D. elata* leaf against the adults of *Cx. quinquefasciatus* with the LC_{50} and LC_{90} values at 197.28 and 347.45mg/L, respectively.

Conclusions: These results suggest that the leaf solvent plant extracts have the potential to be used as an ideal eco-friendly approach for the control of mosquitoes. This study provides first report on the mosquito adulticidal activity of *D. elata* plant extracts against filariasis vector mosquito, *Cx. quinquefasciatus*.

KEYWORDS

Adult mosquitoes, *Delonix elata*, *Culex quinquefasciatus*, Leaf, Seed

1. Introduction

Mosquitoes are the principal vectors of many vector-borne diseases affecting human beings and animals, in addition to its being a nuisance. Vector-borne diseases in India, *e.g.*, malaria, dengue, chikungunya, filariasis, Japanese encephalitis and leishmaniasis, cause thousands of deaths per year. India reported 1.48 million malarial cases and about 1 173 deaths; 1.4 million suspected and 1 985 confirmed chikungunya cases; 5 000 Japanese encephalitis cases and approximately 1 000 deaths; 383 dengue cases and six deaths during 2006 and 2007[1]. *Culex quinquefasciatus* (Say.) (*Cx. quinquefasciatus*) acts as a vector for filariasis in India. Human filariasis is a major public health hazard and remains a challenging socioeconomic problem in most of the tropical countries. Lymphatic filariasis caused by *Wuchereria bancrofti* and transmitted by mosquito *Cx. quinquefasciatus* is found to be more endemic in the

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Indian subcontinent. It is reported that *Cx. quinquefasciatus* infects more than 100 million individuals worldwide annually[2]. Mosquito control relies heavily on synthetic insecticide application. However, over and injudicious application of synthetic insecticides results in resistance to these insecticides and unwarranted toxic or lethal effects on nontarget organisms as well as environmental/health problem. As an alternate, biological control of mosquitoes could be very promising eco-friendly as well as cost effective. Hence, there is a constant need for developing biologically active plant materials as insecticides, which are expected to reduce the hazards to humans and other organisms by minimizing the accumulation of harmful residues in the environment. Natural products of plant origin are generally preferred because of the less harmful nature to nontarget organisms and the innate biodegradability[3].

Botanicals can be used as alternative synthetic insecticides or along with other insecticides under integrated vector control programmes. The plant product of phytochemical is used as insecticides for killing larvae or adult mosquitoes or as repellents for protection against mosquito bites[4]. The adulticidal and repellent activities of crude hexane, chloroform, benzene, acetone and methanol extracts of the leaf of *Cassia tora* were assayed for the toxicity against three important vector mosquitoes, *viz.*, *Cx. quinquefasciatus*, *Aedes aegypti* (*Ae. aegypti*) and *Anopheles stephensi* (*An. stephensi*)[5]. The efficacy of few Malaysian essential oils such as *Laternula elliptica*, *Polygonum minus*, and *Piper aduncum* are used as potential mosquito adulticides[6]. The adulticidal, repellent, and ovicidal potential of the crude hexane, ethyl acetate, benzene, aqueous and methanol solvent extracts from the medicinal plants *Andrographis paniculata*, *Cassia occidentalis* and *Euphorbia hirta* were studied against the medically important mosquito vector, *An. stephensi*[7]. The larvicidal and adulticidal activities of hexane, ethyl acetate and methanol extract from *Momordica charantia, Moringa oleifera, Ocimum gratissimum, Ocimum tenuiflorum, Punica granatum* and *Tribulus terrestris* are against *Culex gelidus* and *Cx. quinquefasciatus*[8].

Delonix elata (*D. elata*) is a deciduous tree about 2.5-15 m tall with a spreading and rounded crown, crooked poor stem form and drooping branches. Bark is smooth and shining, sometimes flaking. Leaves 3-6 or more, bipinnate; pinnae usually 4-6 pairs; leaflets 10-14 pairs, oblong or oblanceolate-oblong, 0.6-1.2 cm long. Pods red-brown or purple-brown, up to 20 cm long and smooth, compressed elliptic-oblong. The genera comprises of 3 tropical species. *D. elata* is a varied species; two variants are recognized in east Africa. *Delonix* is from the Greek word "*delos*", meaning evident and "*onux*", a claw in allusion to the shape of the petals; the epithet "*elata*" means lofty or tall. *D. elata* is easily grown from poles. Direct seeding is a favoured propagation method. Seed is commonly found in animal droppings, some of these germinate to produce seedlings. *D. elata* is a fast growing tree raised easily from seed. Because of its high light requirements. it should be planted in full sunlight. Young seedlings need protection from browsers. Pollarding, lopping and trimming are recommended management practices[9]. As every literature survey could ascertain, no information was available on the adulticidal activity of the experimental plants species given against filariasis vector *Cx. quinquefasciatus*. Therefore, the aim of the present study on investigating the mosquito adulticidal activity of different solvent crude extracts of *D. elata* was assayed for their toxicity against filariasis vector mosquito *Cx. quinquefasciatus*.

2. Materials and methods

2.1. Collection of plants

The mature leaves and seeds of the *D. elata* were collected from Thanjavur District (between 9° 50′ and 11° 25′ of the north latitude and 78° 45′ and 70° 25′ of the east longitude), Tamil Nadu, India. It was authenticated by a plant taxonomist from the Department of Botany, Annamalai University. A voucher specimen was deposited at the Herbarium of Plant Phytochemistry Division, Department of Zoology, Annamalai University.

2.2. Extraction

The fully developed fresh leaves and seeds were washed with tap water, shade-dried and finely ground. The finely ground plant leaf and seed powder (1.0 kg/solvent) was loaded in Soxhlet apparatus and was extracted with five different solvents, *viz.*, hexane, benzene, chloroform, ethyl acetate and methanol, individually. The solvents from the extracts were removed using a rotary vacuum evaporator to collect the crude extract. Standard stock solutions were prepared at 1% by dissolving the residues in ethanol. From this stock solution, different concentrations were prepared and these solutions were used for adulticidal bioassays.

2.3. Test organisms

Cx. quinquefasciatus were reared in the vector control laboratory, Department of Zoology, Annamalai University. The larvae were fed on dog biscuits and yeast powder in the 3:1 ratio. Adults were provided with 10% sucrose solution and 1-week-old chick for blood meal. Mosquitoes were held at (28±2) °C, 70%-85% relative humidity, with a photoperiod of 12 h light and 12 h dark.

2.4. Adulticidal activity

Sugar-fed adult female mosquitoes (5 to 6 days old) were used. The different concentration of plant extracts were impregnated on filter papers (140 mm×120 mm). A blank paper consisting of only ethanol was used as control. The papers were left to dry at room temperature to evaporate off the ethanol overnight. Impregnated papers were prepared freshly prior to testing. The bioassay was conducted in an experimental kit consisting of two cylindrical plastic tubes both measuring 125 mm×44 mm following the WHO method[10]. One tube served to expose the mosquitoes to the plant extracts and another tube was used to hold the mosquitoes before and after the exposure periods. The impregnated papers were rolled and placed in the exposure tube. Each tube was closed at one end with a 16 mesh size wire screen. Sucrose-fed and blood starved mosquitoes (20) were released into the tube, and the mortality effects of the extracts were observed every 10 min for 3 h exposure period. At the end of 1, 2, and 3 h exposure periods, the mosquitoes were placed in the holding tube. Cotton pads soaked in 10% sugar solution with vitamin B complex was placed in the tube during the holding period of 24 h. Mortality of the mosquitoes was recorded after 24 h. The above procedure was carried out in triplicate for each solvent plant crude extracts concentration.

2.5. Statistical analysis

The average adult mortality data were subjected to probit analysis for calculating LC_{50}, LC_{90} and other statistics at 95% confidence limits of upper confidence limit (UCL), lower confidence limit (LCL) and *Chi*-square values were calculated using the SPSS12.0 (Statistical Package of Social Sciences) software. Results with $P<0.05$ were considered to be statistically significant.

3. Results

The adulticidal activity of different solvent leaf and seed extracts of *D. elata* against the filariasis vector mosquitoes, *viz.*, *Cx. quinquefasciatus* are presented in Tables 1 and 2. At higher concentrations, the adult showed restless movement for some times with abnormal wagging and then died. The rates of mortality were directly proportional to concentration. The *Chi*-square values are significant at $P<0.05$ level. The 95% confidence limits LC_{50} and LC_{90} (LCL-UCL) were also calculated. No mortality was recorded in the control. The results revealed that the *D. elata* leaf methanol extracts had the significant adulticidal activity with the LC_{50} and LC_{90} values at 197.28 mg/L and 347.45 mg/L, respectively. Seed extracts have moderate activity, the higher adulticidal activity was observed in methanol extract with the LC_{50} and LC_{90} values at 253.61 mg/L and 471.53 mg/L, respectively.

Table 1

Adulticidal activity of different solvent leaf extracts of *D. elata* against *Cx. quinquefasciatus*.

Extract	Concentration (mg/L)	% of mortality±SD	LC_{50} (mg/L) (LCL-UCL)	LC_{90} (mg/L) (LCL-UCL)	X^2
Hexane	Control	0.0±0.0	265.97	487.19	13.216*
	90	20.4±1.6	(213.25-325.16)	(407.39-649.10)	
	180	37.9±1.4			
	270	52.6±0.8			
	360	63.8±2.0			
	450	86.3±1.4			
Benzene	Control	0.0±0.0	241.33	433.80	10.853*
	90	21.4±1.4	(197.86-285.89)	(373.21-540.06)	
	180	40.6±0.8			
	270	54.1±1.2			
	360	76.2±1.6			
	450	91.9±2.0			
Chloroform	Control	0.0±0.0	225.43	416.97	13.805*
	90	25.2±1.4	(174.91-275.27)	(352.04-539.14)	
	180	45.1±1.8			
	270	59.3±1.2			
	360	77.5±1.0			
	450	93.9±0.8			
Ethyl acetate	Control	0.0±0.0	209.08	385.10	12.986*
	90	26.3±1.8	(162.29-253.74)	(327.83-487.19)	
	180	48.2±1.2			
	270	64.7±1.0			
	360	83.1±2.2			
	450	96.3±1.8			
Methanol	Control	0.0±0.0	197.28	347.45	10.782*
	80	21.6±1.2	(161.56-232.54)	(301.26-425.01)	
	160	43.2±0.8			
	240	60.5±1.6			
	320	81.7±1.4			
	400	97.2±1.2			

*: $P<0.05$.

Table 2

Adulticidal activity of different solvent seed extracts of *D. elata* against *Cx. quinquefasciatus*.

Extract	Concentration (mg/L)	% of mortality±SD	LC_{50} (mg/L) (LCL-UCL)	LC_{90} (mg/L) (LCL-UCL)	X^2
Hexane	Control	0.0±0.0	330.98	597.20	9.612*
	115	20.8±1.4	(276.55-389.29)	(514.56-742.12)	
	230	35.3±1.6			
	345	52.4±2.0			
	460	71.9±1.8			
	575	86.6±1.0			
Benzene	Control	0.0±0.0	310.56	565.87	10.932*
	115	23.7±1.4	(253.30-369.56)	(484.97-709.76)	
	230	37.1±0.8			
	345	56.4±1.6			
	460	75.8±1.8			
	575	89.8±2.0			
Chloroform	Control	0.0±0.0	289.95	532.77	12.928*
	115	26.7±1.2	(228.36-351.11)	(452.44-679.79)	
	230	40.2±1.6			
	345	58.4±1.4			
	460	81.3±1.8			
	575	92.6±1.0			
Ethyl acetate	Control	0.0±0.0	270.76	500.00	15.504*
	115	29.8±0.8	(204.01-334.96)	(419.44-653.86)	
	230	43.1±1.8			
	345	61.6±2.0			
	460	84.2±1.6			
	575	96.1±1.4			
Methanol	Control	0.0±0.0	253.61	471.53	17.843*
	115	32.4±1.4	(182.25-320.25)	(390.91-631.77)	
	230	46.7±1.8			
	345	64.9±1.4			
	460	87.2±1.6			
	575	98.1±2.0			

*: $P<0.05$.

4. Discussion

Crude extracts from plants have been used as insecticides in many countries for centuries. Crude plant extracts often consist of complex mixtures of active compounds. Advances of using complete mixture may act synergistically and show greater overall bioactivity compared to the individual constituents. The results showed that crude extract of *D. elata* have significant adulticidal and repellent activity against *Cx. quinquefasciatus* mosquitoes. This result is also comparable to earlier reports of Govindarajan *et al.*[11] that the adulticidal activity of hexane, ethyl acetate, benzene, chloroform and methanol leaf and seed extract of *Pithecellobium dulce* against *Cx. quinquefasciatus*. The LC_{50} and LC_{90} values of leaf and seed methanol extracts of *Pithecellobium dulce* against *Cx. quiquefasciatus* were 234.97 mg/L, 309.24 mg/L and 464.86 mg/L, 570.80 mg/L, respectively. The insecticidal activity of *Zingiber officinale* against the larval maturation and adult emergency of *Anopheles pharoensis* third stage was evaluated the concentrations of 100%, 70%, 50%, 25%, 5%, 2%, 1%, 0.9%, 0.7%, 0.5%, and 0.3% showed 100% larval mortality rate and at 0.2% and 0.1% caused mortality of 66.7%, respectively[12]. The adulticidal activity of the essential oil of *Lantana camara* was evaluated against different mosquitoes species on 0.208 mg/cm^2 impregnated papers, the knockdown times (KDT_{50} and KDT_{90}) values of the essential oil were 20, 18, 15, 12, and 14 min and 35, 28, 25, 18, and 23 min against *Ae. aegypti*, *Cx. quinquefasciatus*, *Anopheles culicifacies*, *Anopheles fluvialitis*, and *An. stephensi* with their percent mortality of 93.3%, 95.2%, 100%, 100%, and 100% respectively[13]. The aqueous extracts from leaves of *Ricinus communis* showed that 50% of adult EI_{50} were 374.97 mg/L and 1 180.32 mg/L against third-instar larvae of *Anopheles arabiensis* and *Cx. quinquefasciatus* and the extract showed oviposition deterrent effect against both species[14].

The larvicidal activity of crude extract of *Sida acuta* against three important mosquitoes with LC_{50} values ranging between 38 to 48 mg/L[15]. The adulticidal and repellent activities of crude hexane, ethyl acetate, benzene, chloroform, and methanol extracts of leaf of *E. alba* and *Andrographis paniculata* were assayed for their toxicity against two important vector mosquitoes, *viz.*, *Cx. quinquefasciatus* and *Ae. aegypti*. The highest adult mortality was found in methanol extract of *A. paniculata* against the adults of *Cx. quinquefasciatus* and *Ae. aegypti* with the LC_{50} and LC_{90} values were 149.81, 172.37 mg/L and 288.12, 321.01 mg/L, respectively[16]. The larvicidal and adulticidal activities of ethanolic and water mixture (50:50) of plant extracts *Eucalyptus globulus*, *Cymbopogan citratus*, *Artemisia annua*, *Justicia gendarussa*, *Myristica fragrans*, *Annona squamosa* and *Centella asiatica* were tested against *An. stephensi*, and the most effective between 80% and 100% was observed in all extracts[17]. In conclusion, an attempt has been made to evaluate the role of medicinal plant extracts for the adulticidal bioassay against *Cx. quinquefasciatus*. The results reported in this study opened the possibility for further investigations of the efficacy of adulticidal properties of natural product extracts.

Conflict of interest statement

We declare that we have no conflict of interest.

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Comments

Background

Mosquitoes are the principal vectors of many vector-borne diseases affecting human beings and animals, in addition to its being a nuisance. Vector-borne diseases in India, *e.g.*, malaria, dengue, chikungunya, filariasis, and Japanese encephalitis, cause thousands of deaths per year. This study was aimed to investigate the efficacy of *D. elata* against the adult of *Cx. quinquefasciatus* in the laboratory.

Research frontiers

The aim of this study is to determine the control of vector mosquito *Cx. quinquefasciatus* by the plant *D. elata*.

Related reports

The present paper reported the adulticidal activity of the plant extracted of *D. elata* against vector mosquito. The earlier author reported that the Senthilkumar *et al*. (2009) and Kovendan *et al*. (2013).

Innovations and breakthroughs

Control of vector is an important aspect. Using plan products as a natural enemy without causing any percentage of destruction to environment and very much important to the society. So far, there is no previous record of literature available about the mosquito adulticidal activity of selected plant.

Applications

Product development for mosquito control. Plants are always considered as vast repository of natural compounds, the exploration of research leading to their possible utilization certainly pave the way for the search of new phytochemical compounds and their proper role in the near future as eco-friendly natural pesticides.

Peer review

The research work is very much important for society to control morbidity and other defects caused by mosquitoes and other insect vectors. This work reports a novel approach for the control of filariasis vector mosquitoes.

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