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Mosquito adulticidal properties of *Delonix elata* (Family: Fabaceae) against dengue vector, *Aedes aegypti* (Diptera: Culicidae)

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PEER REVIEW

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Comments

The research work is very much important to the society to control morbidity and other defects caused by mosquitoes and also other insect vectors. This work reports a novel approach for the control of vector mosquitoes.
Details on Page 392

ABSTRACT

Objective: To determine the adulticidal activity of hexane, benzene, chloroform, ethyl acetate and methanol leaf and seed extracts of *Delonix elata* (*D. elata*) against *Aedes aegypti* (*Ae. aegypti*).
Methods: The bioassay was conducted in an experimental kit consisting of two cylindrical plastic tubes both measuring 125 mm×44 mm following the WHO method; mortality of the mosquitoes was recorded after 24 h.
Results: The adulticidal activity of plant leaf and seed extracts showed moderate toxic effect on the adult mosquitoes after 24 h of exposure period. However, the highest adulticidal activity was observed in the leaf methanol extract of *D. elata* against *Ae. aegypti* with the LC_{50} and LC_{90} values 162.87 and 309.32 mg/L, respectively.
Conclusions: From this result, it can be concluded the crude extract of *D. elata* was an excellent potential for controlling *Ae. aegypti* mosquitoes.

KEYWORDS
Delonix elata, Adulticidal activity, *Aedes aegypti*

1. Introduction

Mosquito-borne diseases, such as malaria, yellow and dengue fevers, are a major threat to over two billion people in the tropics. Mosquito bites may also cause allergic responses including local skin reactions and systemic reactions such as urticaria and angioedema[1]. *Aedes aegypti* (*Ae. aegypti*), is generally known as a vector for an arbovirus responsible for dengue fever, which is endemic to Southeast Asia, the Pacific island area, Africa, and the Americas. This mosquito is also the vector of yellow fever in Central and South America and West Africa. Dengue fever has become an important public health problem as the number of reported cases continues to increase, especially with more severe forms of the disease, dengue hemorrhagic fever and dengue

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shock syndrome, or with unusual manifestations such as central nervous system involvement. Dengue is prevalent in more than 100 countries and threatens the health of approximately 2.5 billion people. Around 80 million people are infected annually at an attack rate of 4% worldwide[2]. In recent years, phytochemicals have received much attention due to its target specific, eco-friendly, readily biodegradable, cost-effective, no ill effect on non-target organisms and induce multiple effects against vector mosquitoes. Several phytochemicals extracted from various botanical sources have been reported to have detrimental effects on mosquitoes[3,4]. The adult emergence inhibition (EI) and adulticidal activities of hexane, chloroform, ethyl acetate, and acetone leaves extracts of *Anisomeles malabarica*, *Euphorbia hirta*, *Ocimum basilicum*, *Ricinus communis*, *Solanum trilobatum*, *Tridax procumbens* and seeds of *Gloriosa superba* were tested against *Anopheles stephensi* (*An. stephensi*)[5]. The larvicidal and ovicidal potential of the crude hexane, ethyl acetate, benzene, chloroform and methanol solvent extracts from the medicinal plants *Andrographis paniculata* (*A. paniculata*) were evaluated against the medically important mosquito vectors, *Culex quinquefasciatus* (*Cx. quinquefasciatus*) and *Ae. aegypti*[6].

The adult EI and adulticidal activity of the leaf hexane, chloroform, ethyl acetate, acetone, and methanol extracts of *Aegle marmelos*, *Andrographis lineate*, *A. paniculata*, *Cocculus hirsutus*, *Eclipta prostrate* and *Tagetes erecta* were tested against Japanese encephalitis vector, *Culex tritaeniorhynchus*[7]. The adulticidal and repellent activities of crude hexane, ethyl acetate, benzene, chloroform, and methanol extracts of leaf of *Eclipta alba* and *A. paniculata* were assayed for their toxicity against two important vector mosquitoes *viz.*, *Cx. quinquefasciatus* and *Ae. aegypti*[8]. The acetone, chloroform, ethyl acetate, hexane and methanol leaf extracts of *Acalypha indica*, *Achyranthes aspera*, *Leucas aspera*, *Morinda tinctoria* and *Ocimum sanctum* were studied against the early fourth-instar larvae of *Ae. aegypti* and *Cx. quinquefasciatus*[9].

Delonix elata (*D. elata*) commonly known as white gold mohur (Fabaceae) is used by folklore for joint pains and in flatulence. In Indochina, the bark is considered as febrifuge and antiperiodic. The leaf and bark in the form of paste are used by local people to reduce inflammation and pain. It has been used in traditional Indian medicine for the treatment of rheumatism and stomach disorders, and its leaves are used in the treatment of bronchitis and pneumonia in infants[10]. As far as our literature survey could ascertain, no information was available on the adulticidal activity of the experimental plant species given here against *Ae. aegypti*. Therefore, the aim of this study was to investigate the mosquito adulticidal activity of the different solvent extracts of *D. elata*. This is the first report on the mosquito adulticidal activity of the solvent extracts of selected plant.

2. Materials and methods

2.1. Collection of plants

Fully developed 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 is deposited at the Herbarium of Plant Phytochemistry Division, Department of Zoology, Annamalai University.

2.2. Extraction

The 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, namely 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 bioassay.

2.3. Test organisms

Ae. aegypti 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

Five to six-day-old sugar-fed adult female mosquitoes were used. The different concentrations 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[11]. 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 were 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) and lower confidence limit (LCL), and *Chi*-square values were calculated using the SPSS 12.0 software. Results with $P<0.05$ were considered to be statistically significant.

3. Results

The results of the adulticidal activity of hexane, benzene, chloroform, ethyl acetate, and methanol extracts of *D. elata* against the adult of dengue vector mosquitoes, *viz.*, *Ae. aegypti* are presented in Tables 1 and 2. Among extracts tested, the highest adulticidal activity was observed in the leaf methanol extract of *D. elata* against *Ae. aegypti*. 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 LC_{50} and LC_{90} values of *D. elata* leaf and seed extracts against adulticidal activity of (hexane, benzene, chloroform, ethyl acetate, and methanol) *Ae. aegypti* were the following: *Ae. aegypti* (leaf) LC_{50} values were 223.58, 203.03, 190.41, 178.87 and 162.87 mg/L; and LC_{90} values were 401.45, 357.33, 345.70, 316.27 and 309.32 mg/L; seed LC_{50} values were 277.47, 255.46, 237.34, 220.35 and 199.86 mg/L; and LC_{90} values were 507.53, 480.01, 448.95, 415.98 and 387.70 mg/L, respectively. 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.

Table 1
Adulticidal activity of different solvent leaf extracts of *D. elata* against *Ae. aegypti*.

Name of the extract	Concentration (mg/L)	Mortality (%)	LC_{50} (mg/L) (LCL-UCL)	LC_{90} (mg/L) (LCL-UCL)	χ^2
Hexane	Control	0.0±0.0	223.58 (186.64-262.23)	401.45 (347.26-494.72)	9.604*
	80	19.8±1.2			
	160	37.4±1.4			
	240	56.2±0.8			
	320	71.6±2.0			
	400	89.3±1.8			
Benzene	Control	0.0±0.0	203.03 (164.19-241.88)	357.33 (306.95-446.10)	12.274*
	80	21.8±1.4			
	160	40.5±0.8			
	240	59.3±1.6			
	320	78.2±1.0			
	400	97.2±1.8			
Chloroform	Control	0.0±0.0	190.41 (142.20-236.93)	345.70 (288.62-458.57)	17.129*
	80	25.3±1.4			
	160	47.6±1.8			
	240	61.2±1.2			
	320	80.1±0.8			
	400	98.3±2.0			
Ethyl acetate	Control	0.0±0.0	178.87 (147.39-209.60)	316.27 (275.90-381.79)	9.801*
	75	22.1±1.4			
	150	45.3±1.2			
	225	63.2±1.8			
	300	84.6±1.6			
	375	97.2±0.8			
Methanol	Control	0.0±0.0	162.87 (116.94-205.07)	309.32 (257.37-409.84)	16.530*
	75	32.7±0.8			
	150	49.1±1.4			
	225	68.8±1.6			
	300	85.4±2.0			
	375	97.3±1.4			

Values of mortality are expressed as mean±SD. *Significant at $P<0.05$. χ^2: *Chi*-square.

Table 2
Adulticidal activity of different solvent seed extracts of *D. elata* against *Ae. aegypti*.

Name of the extract	Concentration (mg/L)	Mortality (%)	LC_{50} (mg/L) (LCL-UCL)	LC_{90} (mg/L) (LCL-UCL)	χ^2
Hexane	Control	0.0±0.0	277.47 (225.85-331.63)	507.53 (432.81-644.15)	11.223*
	100	21.4±1.4			
	200	40.2±1.8			
	300	54.1±0.8			
	400	72.5±1.2			
	500	88.3±1.6			
Benzene	Control	0.0±0.0	255.46 (199.16-312.03)	480.01 (405.621.44)	13.263*
	100	25.3±1.2			
	200	44.4±1.4			
	300	59.6±0.8			
	400	77.4±2.0			
	500	90.2±1.6			
Chloroform	Control	0.0±0.0	237.34 (179.14-292.73)	448.95 (317.52-583.54)	14.285*
	100	27.6±1.4			
	200	48.1±1.2			
	300	63.7±2.0			
	400	81.1±1.8			
	500	93.4±1.0			
Ethyl acetate	Control	0.0±0.0	220.35 (160.74-275.52)	415.98 (347.56-546.52)	15.894*
	100	31.2±1.0			
	200	48.9±1.4			
	300	68.8±1.6			
	400	84.1±1.2			
	500	97.2±0.8			
Methanol	Control	0.0±0.0	199.86 (129.25-261.55)	387.70 (315.32-539.87)	20.602*
	100	36.2±0.8			
	200	55.6±1.4			
	300	72.4±1.2			
	400	87.3±1.6			
	500	98.8±1.2			

Values of mortality are expressed as mean±SD. *Significant at $P<0.05$. χ^2: *Chi*-square.

4. Discussion

The growing resistance of *Ae. aegypti* populations to the current commercial pesticides has hampered the efforts to control dengue vector effectively. In addition, other serious problems such as high environmental and human toxicity and low biodegradability have been created by the continuous use of synthetic pesticides. Hence, there has been an increasing interest in the development of alternative methods of mosquito control which are less hazardous to humans and other living organisms. In this regard, plant derived compounds have emerged as good candidates, not only as new effective tools in vector management but also as environmentally safer agents[3]. The results are also comparable to earlier reports of Senthilkumar *et al.*[12], reporting that 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 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% showing 100% larval mortality rate and at 0.2% and 0.1% caused mortality of 66.7%, respectively[13]. Larvicidal activity of crude extract of *Sida acuta* against three important mosquitoes with LC_{50} values ranging from 38 to 48 mg/L. The crude extract had strong repellent action against three species of mosquitoes as it provided 100% protection against *An. stephensi* for 180 min followed by *Ae. aegypti* (150 min) and *Cx. quinquefasciatus* (120 min) respectively[3]. 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 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[14]. The aqueous extracts from leaves of *Ricinus communis* showed that 50% of adult EI_{50} were 374.97 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[15]. The larvicidal activity of extracts of medicinal plants *Plumbago zeylanica* and *Cestrum nocturnum* was tested against *Ae. aegypti*; the LC_{50} values of both the plants were less than 50 mg/L. The larvicidal stability of the extracts at five constant temperatures (19 °C, 22 °C, 25 °C, 28 °C and 31 °C) evaluated against fourth instars larvae revealed that toxicity of both plant extracts increased with increase in temperature[16]. The results of the present study would be useful in promoting research aiming at the development of new agent for mosquito control based on plant source. In view of the recent increased interest in developing plant-based insecticides as an alternative to chemical insecticides, this study was undertaken to assess the adulticidal properties of *D. elata* leaf and seed extracts against the dengue vector mosquito, *Ae. aegypti*.

Conflict of interest statement

We declare that we have no conflict of interest.

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Comments

Background

Mosquitoes are well known for their public health importance since they cause major health problems and diseases. Indiscriminate use of several mosquitocidal agents caused various side effects. Thus there is a need to develop an alternative strategy to control vector mosquitoes. A vector is widely distributed in tropical and subtropical zones. This study aimed to investigate the efficacy of *D. elata* against the adult of *Ae. aegypti* mosquitoes in the laboratory.

Research frontiers

This study aimed to determine the control of vector mosquito *Ae. aegypti* by the plant *D. elata*.

Related reports

In this present experiment, authors have followed standard protocols to assess the mosquito adulticidal action of selected plant. These findings are in close agreement with the earlier reports of Senthilkumar *et al.* (2009), Patil *et al.* (2011), and Elango *et al.* (2012).

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 is very much important to the society. So far, there is no previous record of literature available about the mosquitocidal activity of selected plant.

Applications

Product development is for mosquito control. Plants are always considered as vast repository of natural compounds. The exploration of research leading to their possible utilization certainly paves the way for 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 to the society to control morbidity and other defects caused by mosquitoes and also other insect vectors. This work reports a novel approach for the control of vector mosquitoes.

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