Adulticidal properties of *Cardiospermum halicacabum* plant extracts against three important vector mosquitoes

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Abstract. – OBJECTIVE, To determine adulticidal activitiy of hexane, ethyl acetate, benzene, chloroform and methanol leaf extracts of *Cardiospermum halicacabum* against *Culex quinquefasciatus* (*Cx. quinquefasciatus*), *Aedes aegypti* (*Ae. aegypti*) and *Anopheles stephensi* (*An. stephensi*).

MATERIALS AND METHODS, The bioassay was conducted in an experimental kit consisting of two cylindrical plastic tubes both measuring 125 × 44 mm following the WHO method; Mortality of the mosquitoes was recorded after 24 h.

RESULTS, The adulticidal activity of plant extracts showed moderate toxic effect on the adult mosquitoes after 24 h of exposure period. However, compared to other solvents highest mortality was found in methanol extract of *C. halicacabum* against all the three mosquitoes. Among them *An. stephensi* produce the highest LC_{50} and LC_{90} (186.00 and 346.06 ppm) values.

CONCLUSIONS, From the results it can be concluded the crude extract of *C. halicacabum* was an excellent potential for controlling *Cx. quinquefasciatus*, *Ae. aegypti* and *An. stephensi* mosquitoes.

Key Words:

Adulticidal activity, *Cardiospermum halicacabum, Culex quinquefasciatus, Aedes aegypti, Anopheles stephensi.*

Introduction

Mosquitoes are important vectors of etiological agents of diseases to humans and domestic animals. *Aedes aegypti* (L.) 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 shock syndrome, or with unusual manifestations such as central nervous system involvement[1]. *Cx. quinquefasciatus* (Say.) acts as a vector for filariasis in India. Human filariasis is a major public health hazard and remains a challenging socioeconomic problem in many of the tropical countries[2]. Lymphatic filariasis caused by *Wuchereria bancrofti* and transmitted by mosquito *Cx. quinquefasciatus* is found to be more endemic in the Indian subcontinent. It is reported that *Cx. quinquefasciatus* infects more than 100 million individuals worldwide annually[3]. *Anopheles stephensi* Liston is the primary vector of malaria in India and other West Asian countries, Malaria remains one of the most prevalent diseases in the tropical world. With 200 million to 450 million infections annually worldwide, it causes up to 2.7 million deaths. The disease remains endemic in more than 100 developing tropical countries, and its control is a major goal for improved worldwide health. On a global scale, malaria causes 300-500 million cases and results in 1.5-3 million deaths annually. In India, malaria is one of the most important causes of direct or indirect infant, child, and adult mortality. About 2 million confirmed malaria cases and 1,000 deaths are reported annually, although 15 million cases and 20,000 deaths are estimated by WHO South East Asia Regional Office. India contributes 77% of the total malaria in Southeast Asia[4].

Mosquitoes are also becoming increasingly resistant to traditional chemical pesticides and there is growing concern about the potential health and environmental risks surrounding these products. Environmental protection agencies have banned or placed severe restrictions on the

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use of many pesticides which were formerly used in mosquito control programmes and there are now fewer adulticides available than there have been for the last 20 years[5]. Due to environmental concern of use of synthetic insecticides for vector control and due to existing and further risk of development of widespread insecticide resistance in disease vectors, interest on possible use of environment friendly natural products such as extracts of plant/plant parts increased for vector control[6] listed 346 species from 27 genera and 99 families which has been tested against mosquitoes for various effects such as toxicity, growth inhibition, ovipositional detterrency and repellency. Botanicals can be used as alternative synthetic insecticides or along with other insecticides under integrated vector control programmes. The plant product of phytochemical, which is used as insecticides for killing larvae or adult mosquitoes or as repellents for protection against mosquito bites[7,8,9,10].

Kamaraj and Rahuman[11], to determine the larvicidal and adulticidal activities of hexane, ethyl acetate and methanol extracts of *Momordica charantia* (*M. charantia*), *Moringa oleifera (M. oleifera), Ocimum gratissimum (O. gratissimum), Ocimum tenuiflorum (O. tenuiflorum), Punica granatum (P. granatum)* and *Tribulus terrestris (T. terrestris)* against *Culex gelidus (Cx. gelidus*) and *Culex quinquefasciatus (Cx. quinquefasciatus)*.

Rohani et al[12], has reported the efficacy of few Malaysian essential oils such as *Litsea elliptica, Polygonum minus* and *Piper aduncum* as potential mosquito adulticides while Sulaiman et al[13] has reported the essential oils of *Melaleuca cajuputi* and *Cymbopogon nardus* have adulticidal effects on *Aedes* mosquito at high-rise flats in Kuala Lumpur. The compounds, 4 thiophenes,5-(but-3-ene-1-ynyl)-2,2-bithiophene,5-(but-3-ene-1-ynyl)-5′-methyl-2,2-bithiophene,2,2′,5′,2”-terthiophene, and 5-methyl-2,2′,5′,2”-terthiophene, isolated from floral extract of *T. minuta* were largely responsible for the toxicity exhibited against the adults of *Ae. aegypti* and *An. stephensi*[14]. Botanical phytochemicals with mosquitocidal potential are now recognized as potent alternative insecticides to replace synthetic insecticides in mosquito control programs due to their excellent larvicidal, pupicidal, and adulticidal properties. Insecticide applications, although highly efficacious against the target species vector control, is facing a threat due to the development of resistance to chemical insecticides, resulting in rebounding vectorial capacity[15].

Chaiyasit et al[16] referred that essential oils derived from five plant species, celery (*Apium graveolens*), caraway (*Carum carvi*), zedoary (*Curcuma zedoaria*), long pepper (*Piper longum*), and Chinese star anise (*Illicium verum*), were subjected to investigation of adulticidal activity against mosquito vectors. With concern for the quality and safety of life and the environment, the emphasis on controlling mosquito vectors has shifted steadily from the use of conventional chemicals toward alternative insecticides that are target-specific, biodegradable, and environmentally safe, and these are generally botanicals in origin. Although plants and their derivatives were used for controlling and eradicating mosquitoes and other domestic pests before the advent of synthetic organic chemicals[17], only few insecticides of plant origin have been found commercially available. Plant-derived bioproducts, however, still have encouraging results in the control of mosquito vectors if they are adequately effective and harmless to beneficial nontarget organisms and the environment. Furthermore, the insect resistance to mosquitocidal botanical agents has not been documented[17].

In our previous studies, the larvicidal efficacy of the crude leaf extracts of *Ficus benghalensis* with three different solvents like methanol, benzene, and acetone was tested against the early second, third, fourth-instar larvae of *Cx. quinquefasciatus, Ae. aegypti*, and *An. stephensi*. Among the three solvents, the maximum efficacy was observed in methanol[18]; Govindarajan[19] reported that the leaf methanol, benzene, and acetone extracts of *Cassia fistula* were studied for the larvicidal, ovicidal, and repellent activities against *Ae. aegypti*. The leaf extract of *Acalypha indica* with different solvents viz., benzene, chloroform, ethyl acetate, and methanol were tested for larvicidal, ovicidal activity, and oviposition attractancy against *An. stephensi*[20]. Govindrajan et al[21] have tested extracelluler secondary metabolite of different soil fungi against late third-instar larvae of *Cx. quinquefasciatus*. Govindrajan[22] studied that the mosquito larvicidal and ovicidal activity of crude hexane, ethyl acetate, benzene, chloroform, and methanol extracts of the leaf of three plants, *Eclipta alba, Cardiospermum halicacabum*, and *Andrographis paniculata*, were tested against the early third-instar larvae of *Anopheles stephensi*. The larvicidal and ovicidal efficacy of different extracts of *Cardiospermum halicacabum* L. were tested against *Cx. quinquefasciatus* Say and *Aedes aegypti* L. (Diptera: Culicidae)[23]. Govindarajan et al[24] evaluate the ovicidal and repellent activities of methanol leaf extract of *Ervatamia coronaria* and *Caesalpinia pulcherrima* against *Cx. quinquefasciatus*, *Ae. aegypti* and *An. stephensi*. Larvicidal and ovicidal activities of benzene, hexane, ethyl acetate, methanol and chloroform leaf extract of *Eclipta alba* was investigated against dengue vector, *Ae. aegypti*[25]. Govindarajan investigates the larvicidal and repellent properties of essential oils from various parts of four plant species *Cymbopogan citrates, Cinnamomum zeylanicum, Rosmarinus officinalis* and *Zingiber officinale* against *Cx. tritaeniorhynchus* and *An. subpictus*[26]. The larvicidal and ovicidal efficacy of different extracts of *Andrographis paniculata* against *Cx. quinquefasciatus* Say and *Ae. aegypti* L. (Diptera: Culicidae) was also studied[27]. Govindarajan et al[28] determine the larvicidal efficacy of different solvent leaf extract of *Ficus benghalensis* against *Cx. tritaeniorhynchus* and *An. subpictus*.

Currently, numerous products of botanical origin, have received considerable renewed attention as potentially bioactive agents used in insect vector management. Little work has been performed on their mosquito adulticidal activity. This study was carried out to investigate the efficacy of *C. halicacabum* against adult *Cx. quinquefasciatus, Ae. aegypti* and *An. stephensi* mosquitoes, the laboratory and natural field strains, in search of effective indigenous bioproducts to replace synthetic insecticides for the control of mosquitoes, particularly in cases where susceptibility is decreasing.

Materials and Methods

Plant Collection

The fully developed fresh leaves of *C. halicacabum* were collected from different regions of Cuddalore District, Tamilnadu, India. It was authenticated by a plant taxonomist from the Department of Botany, Annamalai University. A voucher specimen is deposited at the Herbarium of the Plant Phytochemistry Division, Annamalai University.

Preparation of the Extract

The leaves were washed with tap water, shade dried at room temperature, and powdered by electrical blender. The powder (1.0 kg) was extracted with five different solvents at Soxhlet apparatus for 8 h. The extract was filtered through a Buchner funnel with Whatman number 1 filter paper. The filtrate was evaporated to dryness under reduced pressure using rotary evaporator to yield a dark greenish, gummy extract. Standard stock solutions were prepared at 1% by dissolving the residues in ethanol, which was used for the bioassays.

Test Organisms

The mosquitoes, *Cx. quinquefasciatus, Ae. aegypti* and *An. stephensi* 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 one week old chick for blood meal. Mosquitoes were held at 28 ± 2°, 70-85% relative humidity (RH), with a photo period of 14 h light, 10 h dark.

Adulticidal Activity

Five to six day old sugar-fed adult female mosquitoes were used. The *C. halicacabum* plant leaf extracts were diluted with ethanol to make different concentrations. The diluted plants extracts were impregnated on filter papers (140 × 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 fresh prior to testing. The bioassay was conducted in an experimental kit consisting of two cylindrical plastic tubes both measuring 125 × 44 mm following the WHO method[29]. One tube served to expose the mosquitoes to the plants 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 extracts concentration.

Statistical Analysis

The average adult mortality data were subjected to probit analysis for calculating LC_{50},

LC_{90} and other statistics at 95% fiducial limits of upper confidence limit and lower confidence limit, and chi-square values were calculated using the SPSS 12.0 version software. Results with $p < 0.05$ were considered to be statistically significant.

Results

In the present observation adulticidal activity of hexane, ethyl acetate, benzene, chloroform and methanol extract of *C. halicacabum* against blood starved adult female of *Cx. quinquefasciatus, Ae. aegypti* and *An. stephensi* were given in Tables I, II, and III. The adulticidal activity of tested plant extracts showed moderate toxic effect on the adult mosquitoes after 24 h of exposure period. However, compared to other solvents highest mortality was found in methanol extract of *C. halicacabum* against all the three mosquitoes. Among them *An. stephensi* produce the highest LC_{50} and LC_{90} (186.00 and 346.06 ppm) values. The LC_{50} and LC_{90} values of *Cx. quinquefasciatus* and *Ae. aegypti* are 211.78, 227.33 ppm and 395.28, 423.33 ppm, respectively.

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, they may show greater overall bioactivity compared to the individual constituents. Our results showed that crude extract of *C. halicacabum* have significant adulticidal activity against *Cx. quin-*

Table I. Adulticidal activity of different solvent extracts of *Cardiospermum halicacabum* against *Culex quinquefasciatus*.

Solvents	Concentration (ppm)	% of mortality ± SD	LC_{50} (ppm) (LCL-UCL)	LC_{90} (ppm) (LCL-UCL)	χ^2
Hexane	Control	0.0 ± 0.0	264.13	487.07	17.466[a]
	110	28.0 ± 1.2	(195.06-330.95)	(404.57-653.81)	
	220	45.0 ± 0.4			
	330	61.0 ± 0.6			
	440	79.0 ± 1.1			
	550	97.3 ± 0.8			
Ethyl acetate	Control	0.0 ± 0.0	259.54	477.09	11.575[a]
	100	23.0 ± 0.8	208.31-310.88	407.82-600.93	
	200	41.4 ± 1.2			
	300	62.0 ± 0.6			
	400	75.4 ± 1.4			
	500	91.2 ± 0.6			
Benzene	Control	0.0 ± 0.0	243.01	452.85	12.838[a]
	100	26.0 ± 1.2	189.13-295.10	384.63-576.48	
	200	44.2 ± 0.8			
	300	66.0 ± 0.7			
	400	79.2 ± 1.1			
	500	93.2 ± 0.5			
Chloroform	Control	0.0 ± 0.0	227.65	428.17	15.636[a]
	100	29.2 ± 0.6	168.04-283.41	358.38-561.33	
	200	47.0 ± 1.2			
	300	69.8 ± 0.9			
	400	81.0 ± 1.1			
	500	96.2 ± 0.4			
Methanol	Control	0.0 ± 0.0	211.78	395.28	18.183[a]
	100	32.0 ± 1.6	149.45-268.78	327.14-530.27	
	200	49.2 ± 1.7			
	300	73.0 ± 0.4			
	400	85.2 ± 1.0			
	500	99.2 ± 0.4			

[a]Significant at $p < 0.05$. SD: Standard Deviation; LC: Lower Confidence Limits; UCL: Upper Confidence Limits; χ^2: Chi square.

Table II. Adulticidal activity of different solvent extracts of Cardiospermum halicacabum against *Aedes aegypti*.

Solvents	Concentration (ppm)	% of mortality ± SD	LC_{50} (ppm) (LCL-UCL)	LC_{90} (ppm) (LCL-UCL)	χ^2
Hexane	Control	0.0 ± 0.0	274.07	500.35	17.530[a]
	120	29.0 ± 0.8	(201.55-343.06)	(416.64-665.82)	
	240	46.0 ± 0.4			
	360	64.0 ± 1.2			
	480	83.6 ± 1.0			
	600	99.0 ± 0.6			
Ethyl acetate	Control	0.0 ± 0.0	269.40	496.93	12.869[a]
	110	26.4 ± 1.5	210.93-326.36	422.60-631.28	
	220	41.0 ± 0.9			
	330	66.2 ± 0.7			
	440	79.0 ± 1.0			
	550	93.9 ± 0.6			
Benzene	Control	0.0 ± 0.0	255.57	475.41	13.805[a]
	110	28.0 ± 1.0	195.29-312.85	402.28-608.96	
	220	45.4 ± 0.4			
	330	68.0 ± 0.8			
	440	81.4 ± 1.5			
	550	95.8 ± 0.6			
Chloroform	Control	0.0 ± 0.0	240.78	452.59	16.229[a]
	110	31.5 ± 1.2	175.07-301.53	377.84-595.49	
	220	47.0 ± 0.4			
	330	71.7 ± 0.5			
	440	84.0 ± 0.8			
	550	97.4 ± 1.0			
Methanol	Control	0.0 ± 0.0	227.33	423.33	18.856[a]
	110	34.2 ± 1.8	158.40-290.23	349.05-572.33	
	220	48.2 ± 0.4			
	330	73.4 ± 0.5			
	440	88.0 ± 0.7			
	550	99.6 ± 1.2			

[a]Significant at $p < 0.05$. SD: Standard Deviation; LCL: Lower Confidence Limits; UCL: Upper Confidence Limits; χ^2: Chi square.

quefasciatus, An.stephensi and *Aedes aegypti* mosquitoes. The results are comparable with an earlier report by Choochote et al[30] who also tried to demonstrate the adulticidal property of *Kaempferia galangal*. However, it only caused a knockdown effect at the initial stage of exposure but, after transferring to the holding tube the mosquito recovered from the knockdown effect. Choochote et al[31] also reported that the in testing for adulticidal activity, the hexane-extracted *Curcuma aromatica* (LC_{50}: 1.60 microg/mg female) was found to be slightly more effective against female *Ae. aegypti* than volatile oil (LC_{50}: 2.86 microg/mg female). The highest adulticidal effect was established from *Piper sarmentosum*, followed by *Piper ribesoides* and *Piper longum*, with LD_{50} values of 0.14, 0.15 and 0.26 microg/female, respectively[32]. The crude seed extract of celery, *Apium graveolens*, exhibited a slightly adulticidal potency on *Ae. aegypti* with LD_{50} and LD_{95} values of 6.6 and 66.4 mg/cm^2, respectively[33]. The unsaponifiable portion and volatile oil of *Thymus capitatus* showed the highest adulticidal potency (LC_{50} = 0.0070 and 0.0076 mg/cm^2, respectively against *Culex pipiens*[34]. Dua et al[35] who observed that the adulticidal activity of the essential oil of *Lantana camara* was evaluated against different mosquitoes species on 0.208 mg/cm^2 impregnated papers, and 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, An. culicifacies, An. fluvialitis* and *An. stephensi* with their percent mortality of 93.3%, 95.2%, 100%, 100% and 100%, respectively.

The root extract of *Valeriana jatamansi* which exhibited adulticidal activity of 90% lethal concentration against adult *An. stephensi, An. culicifacies, Ae. aegypti, Anopheles albopictus*, and *Cx. quinquefasciatus* were 0.14, 0.16, 0.09, 0.08, and

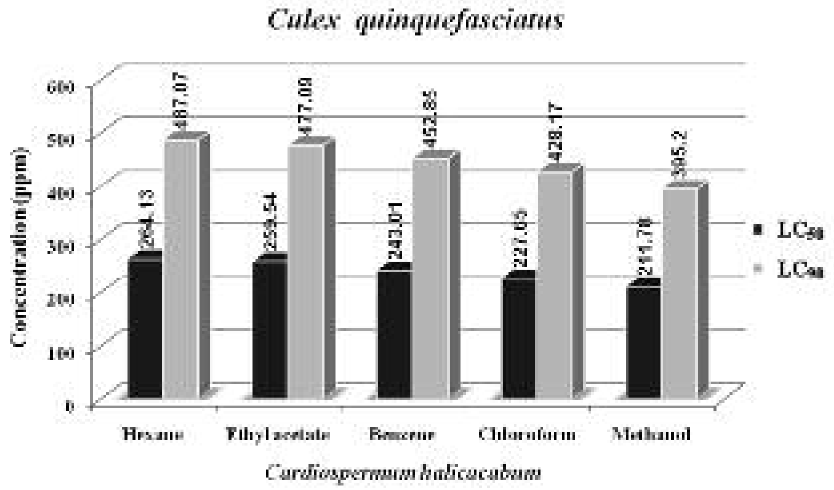
Table III. Adulticidal activity of different solvent extracts of *Cardiospermum halicacabum* against *Anopheles stephensi*

Solvents	Concentration (ppm)	% of mortality ± SD	LC_{50} (ppm) (LCL-UCL)	LC_{90} (ppm) (LCL-UCL)	χ^2
Hexane	Control	0.0 ± 0.0	236.72	428.82	15.931a
	100	27.0 ± 1.3	(179.48-292.11)	(360.16-560.39)	
	200	44.3 ± 0.5			
	300	62.3 ± 1.1			
	400	81.6 ± 0.7			
	500	98.3 ± 0.9			
Ethyl acetate	Control	0.0 ± 0.0	230.58	421.95	13.042[a]
	90	25.6 ± 1.0	(182.08-279.14)	(358.07-539.89)	
	180	39.0 ± 0.6			
	270	61.2 ± 0.8			
	360	76.0 ± 0.4			
	450	93.6 ± 1.3			
Benzene	Control	0.0±0.0	209.01	389.11	14.846[a]
	90	29.4 ± 0.6	157.56-257.92	327.31-505.07	
	180	44.0 ±1.3			
	270	67.2 ± 0.7			
	360	81.6 ± 1.1			
	450	96.2 ± 0.8			
Chloroform	Control	0.0 ± 0.0	198.28	369.18	16.125[a]
	90	30.4 ± 1.3	145.45-247.42	308.79-484.11	
	180	47.0 ± 0.4			
	270	71.2 ± 0.9			
	360	83.6 ± 1.1			
	450	98.0 ± 0.4			
Methanol	Control	0.0 ± 0.0	186.00	346.06	19.036[a]
	90	33.2± 1.1	129.27-237.65	285.21-468.44	
	180	49.4 ± 0.4			
	270	74.2 ± 0.9			
	360	87.0 ± 0.7			
	450	99.8 ± 1.3			

[a]Significant at $p < 0.05$. SD: Standard Deviation; LCL: Lower Confidence Limits; UCL: Upper Confidence Limits; χ^2: Chi square.

0.17 and 0.24, 0.34, 0.25, 0.21, and 0.28 mg/cm^2, respectively[36]. Nathan et al[37] considered pure limonoids of neem seed, testing for biological, larvicidal, pupicidal, adulticidal, and antioviposition-al activity, *An. stephensi* and the larval mortality was dose-dependent with the highest dose of 1 ppm azadirachtin, evoking almost 100% mortality, affecting pupicidal and adulticidal activity and significantly decreased fecundity and longevity of *An. stephensi*. 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[38].

Elango et al[39] evaluate the adulticidal activity and adult emergence inhibition (EI) of leaf hexane, chloroform, ethyl acetate, acetone, and methanol extracts of *Aegle marmelos* (Linn.) Correa ex Roxb, *Andrographis lineate* Wallich ex Nees., *Andrographis paniculata* (Burm.f.) Wall. ex Nees., *Cocculus hirsutus* L. Diels, *Eclipta prostrata* L., and *Tagetes erecta* L. tested against malarial vector, *Anopheles subpictus* Grassi (Diptera: Culicidae). All plant extracts showed moderate adulticidal activity and EI effects after 24 h of exposure at 1,000 ppm; however, the highest adulticidal activity was observed in ethyl acetate extract of *A. lineata*, chloroform extract of *A. paniculata*, acetone extract of *C. hirsutus*, and methanol extract of *T. erecta* (LD_{50}=126.92, 95.82, 109.40, and 89.83 ppm; LD_{90}= 542.95, 720.82, 459.03, and 607.85 ppm); and effective EI was found in leaf acetone extract of the *A. marmelos*, ethyl acetate extract of *A. lineata*, methanol extracts of *C. hirsutus*, and *T. erecta*,



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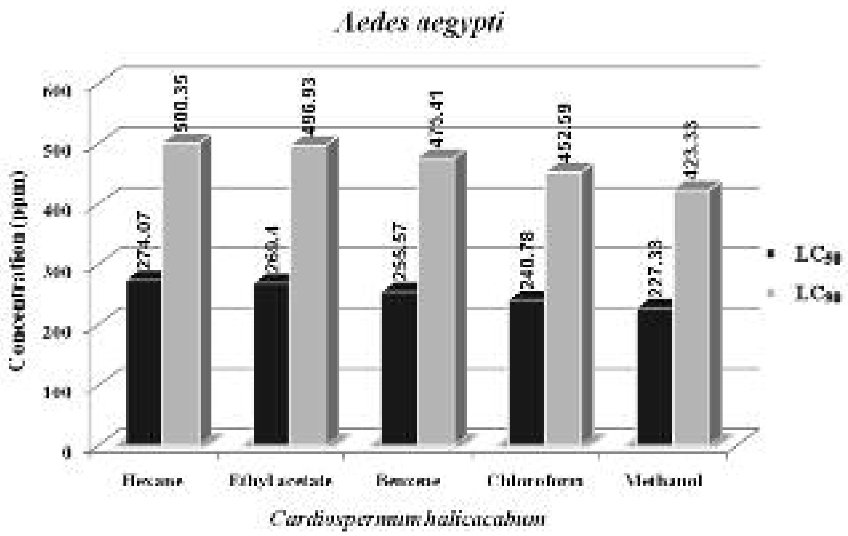

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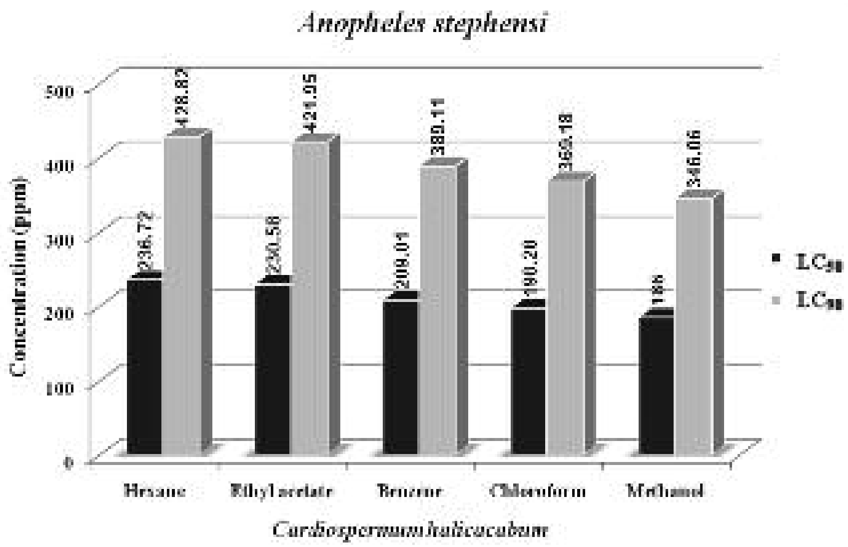

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Figure 1. Figures shows the LC_{50} and LC_{90} values of three important vector mosquitoes.

(EI_{50}=128.14, 79.39, 143.97, and 92.82 ppm; EI_{90} = 713.53, 293.70, 682.72, and 582.59 ppm), respectively, against *An. subpictus*. Kamaraj and Rahuman[11] To determine the larvicidal and adulticidal activities of hexane, ethyl acetate and methanol extracts of *Momordica charantia* (*M. charantia*), *Moringa oleifera* (*M. oleifera*), *Ocimum gratissimum* (*O. gratissimum*), *Ocimum tenuiflorum* (O. tenuiflorum), *Punica granatum* (*P. granatum*) and *Tribulus terrestris* (*T. terrestris*) against *Culex gelidus* (*Cx. gelidus*) and *Culex quinquefasciatus (Cx. quinquefasciatus*). All plant extracts showed moderate larvicidal and adulticidal activities. The adult exposed for 1 h and mortality was recorded at 24 h recovery period. Above 90% mortality was found in the ethyl acetate and methanol extract of all experimental plants at the concentrations of 500 g/mL.

The adulticidal activity of methanol extracts from three Malaysian plants namely *Acorus calamus* Linn., *Litsea elliptica* Blume and *Piper aduncum* Linn. against adult of *Ae. aegypti* (L.) were studied by Hidayatulfathi et al[40]. The hexane fraction from methanol extract of *Acorus calamus* rhizome was the most effective, exhibiting LC_{50} and LC_{90} values of 0.04 $mgcm^{-2}$ and 0.09 $mgcm^{-2}$ respectively. For *L. elliptica*, the methanol fraction also displayed good adulticidal property with the LC_{50} and LC_{90} values of 0.11 $mgcm^{-2}$ and 6.08 $mgcm^{-2}$ respectively. It is found that hexane fraction of the *P. aduncum* crude extract was the least effective among the three plants showing LC_{50} and LC_{90} values of 0.20 $mgcm^{-2}$ and 5.32 $mgcm^{-2}$, respectively. Sulaiman et al[41] evaluated *A. calamus* extract and bifenthrin in the field at high rise flats in Kuala Lumpur. The impact of both plant extract and insecticide on field populations of *Ae. aegypti* and *Ae. albopictus* was monitored weekly. *A. calamus* extract showed adulticidal effect causing 93.9% (inside flats) to 94.9% (outside flats) adult *Ae. aegypti* mortalities compared to bifenthrin with 98.3% (inside flats) and 99.1% (outside flats) adult mortalities. In the control group, the adults of *Ae. aegypti* mortalities were 19.2% (inside flats) and 18.2% (outside flats), respectively 24 h after ULV spraying. Bifenthrin and *Acorus calamus* Linn extract were evaluated against dengue vectors in the laboratory. In testing the adulticidal activity, this plant extract exhibited the LC_{50} and LC_{90} values of 17.4075 and 252.9458 ppm against *Ae. aegypti* and a higher LC_{50} and LC_{90} values of 43.9952 and 446.1365 ppm respectively on *Ae. albopictus*. There was no significant difference on the effect of *A. calamus* extract on both *Aedes* spp adults ($p > 0.05$)[42]. The finding of the present investigation revealed that the leaf extract of *C. halicacabum* possess remarkable adulticidal activity against medically important vector mosquitoes. The extract might be used directly as adulticidal agent in small volume aquatic habitats or breeding sites of limited size around human dwellings. Studies to confirm this hypothesis in field condition are underway in our laboratory.

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