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Larvicidal and adulticidal potential of medicinal plant extracts from south India against vectors

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ABSTRACT

Objective: 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)*. **Methods:** Bioassay test was carried out by WHO method for determination of larvicidal and adulticidal activity against mosquitoes. **Results:** All plant extracts showed moderate larvicidal and adulticidal activities, however the effective larval mortality was found in the leaf ethyl acetate and methanol extract of *O. gratissimum* and bark methanol extract of *M. oleifera* against *Cx.gelidus* with LC_{50} values of 39.31, 66.28, and 21.83 μg/mL respectively, and methanol extract of *O. gratissimum*, *O. tenuiflorum* and *P. granatum* against *Cx. quinquefasciatus* with LC_{50} values of 38.47, 24.90 and 67.20 μg/mL, respectively. 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 μg/mL. **Conclusions:** The present results suggest that the medicinal plant extracts provided an excellent potential for controlling *Cx. gelidus* and *Cx. quinquefasciatus*.

1. Introduction

Lymphatic filariasis stands next to malaria as the most important vector-borne disease in India. *Culex quinquefasciatus (Cx. quinquefasciatus)*, a vector of lymphatic filariasis affects 119 million people living in 73 countries, with India accounting for 40% of the global prevalence of infection[1]. Japanese encephalitis (JE) a mosquito-borne viral disease is a serious public health problem in Asia[2] and it is highly endemic in few districts of Tamil Nadu, Southern India[3,4]. To prevent mosquito-borne diseases and improve public health, it is necessary to control them.

A successive change in the insecticides result in multiple insecticides resistant was developed for vectors. Malaria and filarial vectors in India are resistant to dichlorodiphenyltrichloroethane (DDT), hexachlorocyclohexane (HCH), malathion, and deltamethrin[5]. In this situation, the change of insecticides has hampered the program with increased costs. Thus, the future of vector control mainly relies on the strategies for the management of existing insecticide resistance in the vectors and to limit its further spread. Therefore, it is the hour to launch extensive search to explore eco-friendly biological materials for control of *Culex gelidus (Cx. gelidus)* and *Cx. quinquefasciatus*.

The phytochemicals derived from plant resources can act as larvicides, adulticides, repellents, and ovipositional attractants, having deterrent activities observed by different researchers[6-8] and may be alternative sources of mosquito larval control agents. The plants constitute a rich source of bioactive compounds that are biodegradable into nontoxic products[9].The essential oil of *Momordica charantia (M. charantia)* was evaluated for their topical repellency effects against malarial vector *Anopheles stephensi (An. stephensi)*[10]. The compound lectin isolated from seeds water-soluble extract of *Moringa oleifera(M. oleifera)* and tested against third instar larvae of *Aedes aegypti (Ae. aegypti)*[11] and the seed water extract was assessed on eggs and 3rd instar larvae of *Ae. aegypti*[12]. The essential oil of *Ocimum gratissimum (O. gratissimum)* and *Ocimum americanum (O. americanum)* were tested against *Ae. aegypti*[13]. The acetone, chloroform, ethyl acetate, hexane and methanol leaf extracts of *O. sanctum* were studied against the early fourth-instar larvae

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of *Ae.aegypti* and *Cx. quinquefasciatus*[14].

The fruit rind of *Punica granatum(P. granatum)* acetone, chloroform and ethanol extracts were tested against the 3rd instar larvae of *Chrysomyia albiceps (C. albiceps)*[15]. Leaves and seeds acetone extracts from the *Tribulus terrestris (T. terrestris)* were tested against 3rd-instar larvae of *Anopheles culicifacies (An. culicifacies), An. stephensi, Cx. quinquefasciatus* and *Ae. aegypti*[16]. As part of our continued search for the biodiversity resource available in India for natural products with utilizable bioactivity, the present study assayed larvicidal and adulticidal activity towards *Cx.gelidus* and *Cx. quinquefasciatus* of extracts from 6 medicinal plants commonly available and growing in South India.

2. Materials and methods

2.1. Collection of plant materials

The leaf of *M.charantia, O. gratissimum, Ocimum tenuiflorum (O. tenuiflorum), T. terrestris*, bark of *M. oleifera* and fruit rind of *P. granatum* were collected from Malaiyur Hills and Chitheri Hills Dharmapuri district, Javadhu Hills, Tiruvannamalai district Tamil Nadu.The collected plants were authenticated by Dr. C. Hema, Department of Botany, Arignar Anna Govt. Arts College for Women, Walajapet, Vellore, India. Voucher specimens have been deposited in the laboratory of Zoology, C. Abdul Hakeem College, Melvisharam for further analysis.

2.2. Mosquito rearing

Cx. gelidus and *Cx. quinquefasciatus* larvae were collected from stagnant water area of Melvisharam and identified in Zonal Entomological Research Centre, Vellore, Tamil Nadu. To start the colony, the larvae were kept in plastic and enamel trays containing tap water. They were maintained and reared in the laboratory as per the method of Kamaraj *et al*[6].

2.3. Preparation of the extract

The leaves were dried in *M. charantia* (6 days), *O. gratissimum* (4 days), *O. tenuiflorum* (4 days) *T. terrestris* (10 days), bark of *M. oleifera* (25 days) and fruit rind of *P. granatum* (15 days) in the shade at the environmental temperatures (25-37 ℃ day time). The dried leaves and seeds were powdered mechanically using commercial electrical stainless steel blender and the powdered leaves 800 g, bark 400 g and fruit rind 450 g were extracted with hexane (2 500 mL, Fine Chemicals Ltd. India), ethyl acetate (3 000 mL, Qualigens Chemicals, India), and methanol (3 800 mL, Qualigens Chemicals, India) in a soxhlet apparatus (boiling point range 60-80 ℃) for 8 h. The extracts were filtered through a Buchner funnel with Whatman number 1 filter paper. The extract was concentrated under reduced pressure of 22-26 mmHg at 45 ℃, and the residue obtained was stored at 4 ℃. Polysorbate 80 (Qualigens) was used as an emulsifier at the concentration of 0.05% in the final test solution.

2.4. Larval bioassay

During preliminary screening with the laboratory trial, the larvae of *Cx. gelidus* and *Cx. quinquefasciatus* were collected from the insect-rearing cage and identified in Zonal Entomological Research Centre, Vellore. The larval bioassay was determined as per the procedure of WHO[17] and Rahuman *et al*[18]. Five batches of 20 larvae were used in each bioassay. Six different concentrations (15.63, 31.25, 62, 125, 250 and 500 μg/mL) were papered with 100 mL dechlorinated tap water, in a separate plastic container. Polysorbate 80 (Qualigens) was used as an emulsifier at the concentration of 0.05% in the final test solution. The control was set up with acetone, polysorbate 80 and dechlorinated tap water. The numbers of dead larvae were counted after 24 h of exposure and percentage of mortality was reported from the average of five replicates. The experimental media in which 100% mortality of larvae occurs alone were selected for dose-response bioassay.

2.5. Dose-response bioassay

The different concentrations ranging from 15.63 to 500 μg/mL were prepared. Based on the preliminary screening results, crude different solvents of leaf, bark and fruit rind extracts prepared from *M. charantia, M. oleifera, O. gratissimum, O. tenuiflorum, P. granatum* and *T. terrestris* were subjected to dose-response bioassay for larvicidal activity against the larvae of *Cx. gelidus* and *Cx. quinquefasciatus*. The numbers of dead larvae were counted after 24 h of exposure, and the percentage of mortality was reported from the average of five replicates. However, at the end of 24 h, the selected test samples turned out to be equal in their toxic potential.

2.6. Adulticidal bioassay

Cx. gelidus and *Cx. quinquefasciatus* mosquitoes were selected for this bioassay. The bioassay was performed by WHO method[19]. The adulticidal activity of the plant extracts was evaluated at 500 μg/mL, to produce a range of mortality from 10% to 100% along with control. The different concentrations of the plant extracts were dissolved in 2.5 mL of acetone and applied on Whatman No. 1 filter papers (size 12 cm×15 cm) as described by Dua *et al*[20].Control papers were treated with acetone, polysorbate 80, and distilled water under similar conditions. Twenty female mosquitoes (2-5 days old glucose fed, blood starved) were collected from the insect-rearing cage and gently transferred into a plastic holding tube. The mosquitoes were allowed to acclimatize in the tube for 1 h and then exposed to test paper (filter paper) for 1 h. At the end of exposure period, the mosquitoes were transferred back to the holding tube and kept 24 h for recovery period. A pad of cotton soaked with 10% glucose solution was placed on the mesh screen. Mortality of mosquitoes was determined at the end of 24 h recovery period. The number of mosquitoes knocked down in the exposure tube was recorded at 3 min interval period till the last mosquito was knocked down. Knock down time (KDT) values of KDT_{50} and KDT_{90} were determined using probit analysis. Percent mortality was corrected by using of Abbott's formula[21].

2.7. Statistical analysis

Adult mortality counts were made after 24 h exposure. Bioassay test showing more than 10% control mortality were discarded and repeated. However, when control

mortality ranged from 5% to 10%, the corrected mortality was calculated using Abbott's formula[21]. Median lethal dose (LD_{50}) and LD_{90} with their 95% confidence limits of the extracts were determined using log probit analysis test. The average larval mortality data were subjected to probit analysis for calculating median lethal concentration (LC_{50}), LC_{90} and other statistics at 95% fiducial limits of upper confidence limit and lower confidence limit, and *t*-test were calculated using the software developed by Reddy *et al*[22]. Results with $P<0.05$ were considered to be statistically significant.

3. Results

The results showed that larvicidal and adulticidal, activity of hexane, ethyl acetate, and methanol extracts of six medicinal plant species against *Cx. gelidus* and *Cx. quinquefasciatus* were varied according to plant species.

The preliminary screening is good mean to evaluate the potential larvicidal activity. The larvicidal activity of hexane, ethyl acetate and methanol extracts of *M. charantia*, *M. oleifera*, *O. gratissimum*, *O. tenuiflorum* , *P. granatum* and *T. terrestris* plants were noted(Table 1).

All plant extracts showed moderate toxic effect on the fourth instar larvae after 24 h of exposure; however, the highest mortality was found in leaf ethyl acetate extract of *M. charantia*, ethyl acetate and methanol extracts of *O. gratissimum*, *O. tenuiflorum* and bark methanol extract of *M. oleifera* and fruit rind extract of *P. granatum* against the larvae of *Cx. gelidus* with LC_{50} values of 38.24, 39.31, 66.28, 44.07, 24.90, 21.83, 67.22 μg/mL; LC_{90} values of 180.36, 189.41, 343.50, 261.45, 123.32, 162.34, 323.50 μg/mL; and leaf ethyl acetate extract of *M. charantia*, bark methanol extracts of *M. oleifera*, *O. gratissimum*, *O. tenuiflorum* and fruit rind of *P. granatum* extracts tested against the larvae of *Cx. quinquefasciatus* with LC_{50} values of 41.12, 57.13, 38.47,18.36 and 67.20 μg/mL; the LC_{90} values of 216.61, 247.59, 178.69, 86.29 and 321.58 μg/mL, respectively. *P* values were all significant when compared to the control ($P<0.05$) (Table 2).

The adulticidal activity of tested plant extracts showed moderate toxic effect on the adult mosquitoes after 24 h of exposure at 500 μg/mL; however, the highest mortality was found in methanol, extract of *M. charantia*, *M. oleifera* and ethyl acetate extract of *O. gratissimum*, ethyl acetate and methanol extract of *O. tenuiflorum* and *P. granatum* against adult of *Cx. gelidus* with LD_{50} values of 27.21, 68.03, 72.56, 47.60, 82.66 and 34.26 μg/mL ; LD_{90} values of 146.23, 285.83, 319.14, 264.58, 229.23 and 356.14 μg/mL; and methanol extracts of *M. charantia*, ethyl acetate extract of *M. oleifera*, *O. gratissimum*, *O. tenuiflorum* and *P. granatum* against adult of *Cx.quinquefasciatus* with LD_{50} values of 43.06, 59.34, 62.42, 58.89 and 54.15 μg/mL; LD_{90} values of 248.18, 250.46, 450.72, 326.13 and 423.38 μg/mL, respectively (Table 3).

KDT_{50} and KDT_{90} values were observed in the methanol and ethyl acetate extracts of *M. charantia*, methanol extract of *M. oleifera*, *O. gratissimum* and *P. granatum*, ethyl acetate extract of *O. tenuiflorum* were 25, 16, 24, 15, 12 and 18 min and 30, 32, 30, 20, 20 and 26 min against *Cx. gelidus* with their per cent mortality of 100% and the methanol extract of *M. charantia* and *P. granatum*, ethyl acetate and methanol extract of *M. oleifera*, ethyl acetate extract of *O. gratissimum* and *O. tenuiflorum* were 12, 18, 24, 18, 20 and 20 min and 30, 26, 28, 20, 24 and 22 min against *Cx. quinquefasciatusi* with their per cent mortality of 100 and below 100% was found in hexane extract of *M. charantia* 42.6%, *O. gratissimum* 65.8%, *M. oleifera* and hexane extract *O. tenuiflorum* 91.4% mortality was observed in 24 h recovery period, respectively (Table 4).

4. Discussion

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. In the present observation, hexane, ethyl acetate and methanol extract of leaf, bark and fruit rind of *M. charantia*, *M. oleifera*, *O. gratissimum*, *O. tenuiflorum*, *P. granatum* and *T. terrestris* extracts were tested against two mosquito species, mortality of six plant extracts were recorded, and the findings, related to the larvicidal and adulticidal activities of various plant extracts reported by the earlier authors corroborate the present study results. The larvicidal action of the acetone, chloroform, ethyl acetate, hexane and methanol leaf extracts of *Ocimum canum (O. canum)*, *O. sanctum* and *Rhinacanthus nasutus*

Table 1
Larvicidal activity of crude plant extracts against fourth instar larvae of *Cx. gelidus* and *Cx. quinquefasciatus* at 500 μg/mL (Mean ± SD) (%).

Botanical name	Species	Mortality		
		Hexane	Ethyl acetate	Methanol
M. charantia	*Cx. gelidus*	69.00±2.15	100.00±0.00	82.00±2.62
	Cx. quinquefasciatus	72.00±2.32	100.00±0.00	74.00±2.65
M. oleifera	*Cx. gelidus*	52.00±2.54	78.00±2.64	100.00±0.00
	Cx. quinquefasciatus	61.00 ±2.03	68.00±2.43	100.00±0.00
O. gratissimum	*Cx. gelidus*	93.00±2.13	100.00±0.00	100.00±0.00
	Cx. quinquefasciatus	82.00±1.68	75.00±1.65	100.00±0.00
O. tenuiflorum	*Cx. gelidus*	69.00±1.79	100.00±0.00	100.00±0.00
	Cx. quinquefasciatus	86.00±1.63	76.00±2.48	100.00±0.00
P. granatum	*Cx. gelidus*	72.00±1.14	84.00±1.92	100.00±0.00
	Cx. quinquefasciatus	82.00±2.82	87.00±2.07	100.00±0.00
T. terrestris	*Cx. gelidus*	52.00±2.15	42.00±3.24	48.00±4.26
	Cx. quinquefasciatus	38.00±2.62	56.00±4.68	64.00±2.82

Control – Nil mortality.

Table 2
Larvicidal activity of different solvent crude extracts against fourth instar larvae of *Cx. gelidus* and *Cx. Quinquefasciatus*.

Plant species	Solvents	Species	LC_{50} (μ g/mL)		LC_{90} (μ g/mL)		χ^2(df=4)
			Mean±SD	UCL- LCL	Mean±SD	UCL- LCL	
M. charantia	Ethyl acetate	*Cx. gelidus*	38.24±2.42	86.18-46.61	180.36±10.16	278.34-172.20	8.92
	Ethyl acetate	*Cx.quinquefasciatus*	41.12±2.64	92.24-56.10	216.61±16.86	282.28-186.64	12.62
M. oleifera	Methanol	*Cx. gelidus*	21.83±1.38	24.54-19.14	162.34±8.15	260.40-164.54	6.23
	Methanol	*Cx.quinquefasciatus*	57.13±3.87	64.71-49.54	247.59±27.77	302.01-193.17	15.61
O. gratissimum	Ethyl acetate	*Cx. gelidus*	39.31±2.78	44.77-33.85	189.41±22.79	234.09-144.74	10.36
	Methanol	*Cx. gelidus*	44.07±3.42	89.78-42.37	261.45±35.79	331.61-191.29	10.97
	Methanol	*Cx.quinquefasciatus*	38.47±2.69	43.76-33.17	178.69±21.11	220.06-137.32	12.71
O. tenuiflorum	Ethyl acetate	*Cx. gelidus*	66.28±4.86	75.81-56.75	343.50±44.50	430.03-256.97	12.06
	Methanol	*Cx. gelidus*	24.90±1.78	28.41-21.41	123.32±14.94	152.59-194.05	14.07
	Methanol	*Cx.quinquefasciatus*	18.36±1.32	20.95-15.77	86.29±11.29	108.42-64.16	15.77
P. granatum	Methanol	*Cx. gelidus*	67.22±4.26	72.80-56.45	323.50±40.15	410.03-216.97	10.05
	Methanol	*Cx.quinquefasciatus*	67.20±4.76	76.55-57.86	321.58±38.99	397.99-245.17	11.37

UCL: Upper confidence limit; LCL: Lower confidence limit; df: degree of freedom.

Table 3
LD_{50} and LD_{90} values of adulticidal activity of medicinal plant extract against *Cx. gelidus* and *Cx. Quinquefasciatus*(μ g/mL).

Plant extract	Solvents	Mosquito species	LD_{50}		LD_{90}	
			Mean±SD	UCL-LCL	Mean±SD	UCL-LCL
M. charantia	Methanol	*Cx. gelidus*	27.21±1.56	32.82-22.62	146.23±12.22	194.24-118.92
	Methanol	*Cx. quinquefasciatus*	43.06±2.32	92.51-42.58	248.18±22.34	340.46-242.24
M. oleifera	Methanol	*Cx. gelidus*	68..03±3.21	155.53-102.53	285.83±21.90	387.56-284.09
	Ethyl acetate	*Cx. quinquefasciatus*	59.34±6.46	149.89-98.84	250.46±18.39	347.56-283.37
O. gratissimum	Ethyl acetate	*Cx. gelidus*	72.56±4.51	85.36-57.75	319.14±24.72	481.80-291.48
	Ethyl acetate	*Cx. quinquefasciatus*	62.42±6.43	126.99-92.85	450.72±25.19	354.12-257.30
O. tenuiflorum	Ethyl acetate	*Cx. gelidus*	47.60±3.97	64.38-32.83	264.58±16.65	365.53-252.48
	Methanol	*Cx. gelidus*	82.66±6.19	92.24-51.41	229.23±20.17	335.11-223.55
	Ethyl acetate	*Cx. quinquefasciatus*	58.89±6.46	102.60-71.19	326.13±29.28	345.31-260.94
P. granatum	Ethyl acetate	*Cx. gelidus*	34.26±4.12	126.55-95.72	356.14±24.42	421.47-327.80
	Ethyl acetate	*Cx. quinquefasciatus*	54.15±8.43	152.64-98.62	423.38±25.64	438.63-315.12

(R. nasutus) were studied against fourth instar larvae of *Cx. quinquefasciatus* were documented[23]. Similarly, the larvicidal activity of crude leaf acetone, chloroform, hot water, methanol, petroleum ether and water extracts of *Calotropis procera (C. procera), Canna indica (C. indica), Hibiscus rosa-sinensis (H. rosa-sinensis), Ipomoea carnea (I.carnea)*, and *Sarcostemma brevistigma (S. brevistigma)*[24]. The present study methanol extract of M.charantia showed in 100% larval mortality at concentration of 500 μ g/mL against *Cx. quinquefasciatusi*. The leaf ethanolic extract of *Cassia obtusifolia (C. obtusifolia)* were tested against third instar larvae of *An. stephensi*, the LC_{50} and LC_{90} values were 52.2 and 108.7 mg/L, respectively[25]. The *Ricinus communis (R. communis)* seed extract, showed larval mortality, at the 2 μ g/mL for *An. stephensi* and *Cx. quinquefasciatus*, 4 μ g/mL for *Aedes albopictus (Ae.albopictus)* concentrations[26]. The earlier authors reported that, 100% larval mortality was observed in the essential oils of *Ocimum gratissium (O. gratissium)*, *Cymbopogon citrus (C. citrus),* and *Ageratum conyzoides (A. conyzoides)* against *Ae. aegypti* at concentrations of 120, 200 and 300 ppm, respectively[27].The methyl alcohol extracts of *Acer pseudoplatanus (A.pseudoplatanus), Humulus japonicas (H. japonicas), Acer platanoides (A. platanoides), Satureja hortensis (S. hortensis), Ocimum basilicum (O. basilicum)* and *Thymus vulgaris (T. vulgaris)* showed the LD_{50} 23, 25, 28, 28, 32 and 48 ppm respectively, against the fourth larval instar of the mosquito *Cx. quinquefasciatus*[28]. Our result showed that ethyl acetate extract of the *M. charantia, M. oleifera* have significant adulticidal activity. This result is also comparable to earlier reports of Dua *et al*[29] who observed the adulticidal activity of the essential oil of *Lantana camara (L. 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, An. culicifacies, An. fluvialitis* and *An. stephensi* with their mortality of 93.3%, 95.2%, 100.0%, 100.0% and 100.0% respectively. Ferreira *et al*[12] have reported that the seed extract of *M. oleifera* were tested against third instar larvae of *Ae. aegypti* with their LC_{50} value of 1 260 μ g/mL. The water-soluble lectin was isolated from the seed of *M. oleifera* were tested against fourth-stage larvae of *Ae. aegypti* with the values of LC_{16}, LC_{50} and LC_{84} were 0.153, 0.197, and 0.240 mg/mL, respectively[11]. Similarly, Rahuman and Venkatesan[30] have reported that the petroleum ether extract of *Citrullus colocynthis (C. colocynthis),* methanol extracts of *Coccinia indica (C. indica), Cucumis sativus (C. sativus), Momordica charantia (M. charantia)* and acetone extract of *Trichosanthes anguina (T.anguina)* against early fourth instar larvae of *Ae. aegypti* (LC_{50}=74.57, 309.46, 492.73, 199.14, and 554.20 ppm) and against *Cx. quinquefasciatus* (LC_{50}=88.24, 377.69, 623.80, 207.61, and 842.34 ppm),

Table 4
Adulticidal activity of different solvent extracts against *Cx. gelidus* and *Cx. quinquefasciatus* at 500 μg/mL.

Plant extracts	Mosquito species	Solvents	Knockdown time (min)		Knock-down in 1 h (%)	Mortality 24 h(%)
			KDT_{50}	KDT_{90}		
M. charantia	*Cx. gelidus*	Hexane	20.0±2.4	30.0±2.0	68	42.60±2.26
		Ethyl acetate	25.0±2.1	32.0±1.6	100	100.00
		Methanol	16.0±4.0	25.0±2.1	100	100.00
	Cx. quinquefasciatus	Hexane	18.0±2.3	22.0±3.0	86	68.00±4.24
		Ethyl acetate	15.0±3.0	28.0±3.8	92	90.00±2.64
		Methanol	12.0±2.1	30.0±4.4	100	100.00
M. oleifera	*Cx. gelidus*	Hexane	16.0±4.0	32.0±1.8	80	83.20±5.80
		Ethyl acetate	22.0±2.6	28.0±3.6	96	86.20±4.20
		Methanol	24.0±2.4	30.0±2.4	100	100.00
	Cx. quinquefasciatus	Hexane	16.0±1.8	28.0±1.8	85	91.20±4.80
		Ethyl acetate	24.0±3.0	28.0±2.2	100	100.00
		Methanol	18.0±2.0	20.0±3.6	100	100.00
O. gratissimum	*Cx. gelidus*	Hexane	16.0±3.2	25.0±3.2	60	65.80±5.70
		Ethyl acetate	18.0±1.0	22.0±2.8	94	96.00±4.26
		Methanol	15.0±2.1	20.0±2.2	100	100.00
	Cx. quinquefasciatus	Hexane	18.0±4.6	32.0±1.4	75	78.40±5.80
		Ethyl acetate	20.0±2.8	24.0±2.6	100	100.00
		Methanol	22.0±1.4	30.0±2.8	92	86.40±4.20
O. tenuiflorum	*Cx. gelidus*	Hexane	20.0±3.4	28.0±1.2	86	91.40±4.60
		Ethyl acetate	18.0±2.0	26.0±2.0	100	100.00
		Methanol	15.0±1.0	20.0±3.4	98	82.20±3.60
	Cx. quinquefasciatus	Hexane	10.0±4.6	18.0±4.0	86	92.20±4.10
		Ethyl acetate	20.0±3.2	22.0±3.1	100	100.00
		Methanol	18.0±2.1	24.0±4.0	90	86.20±3.80
P. granatum	*Cx. gelidus*	Hexane	22.0±3.2	32.0±2.2	80	86.40±4.20
		Ethyl acetate	15.0±1.4	22.0±3.4	96	84.20±5.00
		Methanol	12.0±2.1	20.0±2.1	100	100.00
	Cx. quinquefasciatus	Hexane	14.0±2.0	18.0±3.0	60	65.60±4.60
		Ethyl acetate	20.0±3.4	30.0±1.8	94	88.60±5.20
		Methanol	18.0±2.6	26.0±3.4	100	100.00
T. terrestris	*Cx. gelidus*	Hexane	25.0±2.0	35.0 ±2.8	24	18.20±1.60
		Ethyl acetate	20.0±3.6	28.0±4.6	18	14.60±1.40
		Methanol	24.0±2.8	30.0±2.2	36	24.40±2.80
	Cx. quinquefasciatus	Hexane	18.0±2.4	25.0±1.8	28	18.00±1.20
		Ethyl acetate	28.0±1.0	30.0±3.2	32	22.60±2.60
		Methanol	17.0±2.6	24.0±1.6	42	26.40±1.60

respectively. The leaf hexane extract of *M. charantia* showed the LC_{50} values against IV instar larvae of *An. stephensi*, *Cx. quinquefasciatus* and *Ae. aegypti* were 66.05, 96.11 and 122.45 ppm, respectively[31]. Similarly, the fruit wall petroleum ether (LC_{50}=27.60; 17.22 ppm and 41.36; 15.62 ppm) extract was found more effective than carbon tetrachloride (LC_{50}=49.58; 16.15 ppm and 80.61; 27.64 ppm) and methanol (LC_{50}=142.82; 95.98 ppm and 1 057.49; 579.93 ppm) extracts towards *An. tephensi* and *Cx. quinquefasciatus* larvae after 24 and 48 h of exposure respectively[32]. A similar observation has been reported that the adulticidal, repellent, and larvicidal activity of crude hexane, ethyl acetate, and methanol extracts of *Aristolochia indica (A. indica)*, *Cassia angustifolia (C.angustifolia)*, *Diospyros melanoxylon (D. melanoxylon)*, *Dolichos biflorus (D. biflorus)*, *Gymnema sylvestre (G. sylvestre)*, *Justicia procumbens (J. procumbens)*, *Mimosa pudica (M. pudica)*, and *Zingiber zerumbet (Z. zerumbet)* were tested against adult and fourth instar larvae of *Cx. gelidus* and *Cx. quinquefasciatus* showed 100% mortality at 1 000 ppm[33].

In conclusion, the investigation has been made to evaluate the role of medicinal plant extracts in possible to control of mosquitoes. Natural products are generally preferred in vector control measures due to their less deleterious effect on non-target organisms and their innate biodegradability. In the context of resistance developed by the mosquito larvae against chemical insecticides, it is worthwhile to identify new active compounds from natural products against mosquitoes. Hence, the results reported here open the possibility of further elucidations of efficacy on their larvicidal and adulticidal action of the medicinal plant extracts may be exploited against mosquitoes. The bioassay guided fractionation, purification and isolation of pure compounds from the crude ethyl acetate extract of leaves of *M. oleifera*, *O. tenuiflorum*, and methanol extract of *M. charantia* and *P. granatum* are in progress.

Conflict of interest statement

We declare that we have no conflict of interest.

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