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Larvicidal efficacy of medicinal plant extracts against *Anopheles stephensi* and *Culex quinquefasciatus* (Diptera: Culicidae)

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Abstract. Mosquitoes transmit serious human diseases, causing millions of deaths every year. Natural products of plant origin with insecticidal properties have been used in recent years for control of a variety of pest insects and vectors. The present study was based on assessments of the larvicidal activity to determine the efficacies of hexane, chloroform, ethyl acetate, acetone and methanol extracts of ten medicinal plants tested against fourth instar larvae of malaria vector, *Anopheles stephensi* Liston and lymphatic filariasis vector, *Culex quinquefasciatus* Say (Diptera: Culicidae). The larvicidal activity was assessed by the procedure of WHO with some modification. The highest larval mortality was found in leaf acetone of *Adhatoda vasica*, bark ethyl acetate of *Annona squamosa*, methanol leaf and flower of *Cassia auriculata*, leaf ethyl acetate of *Hydrocotyle javanica*, methanol leaf and seed of *Solanum torvum* and leaf hexane extracts of *Vitex negundo* against the fourth instar larvae of *An. stephensi* and *Cx. quinquefasciatus*. The calculated LC_{90} for acetone, ethyl acetate, methanol and hexane extracts of dried leaf and bark of *A. vasica*, *A. squamosa*, *S. torvum*, and *V. negundo* were in the range of 70.38-210.68 ppm. Our results suggest that the leaf methanol extract of *S.torvum* and bark ethyl acetate extract of *A. squamosa* from Southern India have the potential for use to control mosquitoes. Therefore, this study provides the larvicidal activity against *An. stephensi* and *Cx. quinquefasciatus* of plant extracts.

INTRODUCTION

Anopheles stephensi transmits malaria in the plains of rural and urban areas of India. Malaria afflicts 36% of the world population i.e. 2020 million in 107 countries and territories situated in the tropical and subtropical regions. In the South East Asian Region of WHO, out of about 1.4 billion people living in 11 countries, 1.2 billion (85.7%) are exposed to the risk of malaria and most of whom live in India. Of the 2.5 million reported cases in the South East Asia, India alone contributes about 70% of the total cases (Kondrachine, 1992). *Culex quinquefasciatus*, a vector of lymphatic filariasis and it is widely distributed tropical diseases with around 120 million people infected worldwide and 44 million people have common chronic manifestation (Bernhard *et al.*, 2003)

Natural products of plant origin with insecticidal properties have been tried in the recent past for control of variety of insect pests and vectors. Plants are considered as a rich source of bioactive chemicals and they may be an alternative source of mosquito control agents. Natural products are generally preferred because of their less harmful nature to non-target organisms and due to their innate biodegradability.

Many studies on plant extracts against mosquito larvae have been conducted around the world. The leaf powdered preparation of *Adhatoda vasica* (adhatoda), *Azadirachta indica* (neem) and *Ocimum sanctum* (tulsi), which on burning with charcoal produced smoke that repelled *Armigeres subalbatus* and *Cx. quinquefasciatus* to prevent their biting activity for 6–8 h (Pandian *et al.*, 1995). The seeds petroleum ether extract of *Annona squamosa* showed larvicidal activity against *Aedes aegypti*, *Cx. quinquefasciatus* and *An. stephensi* (George & Vincent, 2005) The acetone, chloroform, ethyl acetate, hexane, methanol and petroleum ether extracts of leaf, flower and seed of *Cassia auriculata*, *Solanum torvum* and *Vitex negundo* were tested against fourth instar larvae of *Anopheles subpictus* and *Culex tritaeniorhynchus* (Kamaraj *et al.*, 2009).

Hemidesmus indicus, commonly called Indian sarsaparilla is a climbing vine found throughout India which belongs to the family Asclepiadaceae and the extracts showed antibacterial properties (Aqil & Ahmad, 2007). The methanolic extract *Hydrocotyle javanica* exhibited larvicidal activity against *Cx. quinquefasciatus* (Venkatachalam *et al.*, 2001). The toxicity of *S. torvum* extracts was found effective against the early fourth-instar larvae of *Cx. quinquefasciatus* (Rahuman *et al.*, 2008). Acetone, chloroform, ethyl acetate, hexane, and methanol dried leaf, and seed extracts of *S. trilobatum* were tested against the fourth instar larvae of *An. subpictus* and *Cx. tritaeniorhynchus* (Zahir *et al.*, 2009). The oil obtained from leaves of *V. negundo* was evaluated against *Ae. aegypti* (Hebbalkar *et al.*, 1992). In all probability, the plants contained insecticidal phytochemicals that were predominantly secondary compounds produced by plants to protect themselves against herbivorous insects. The effective mosquito larvicidal plant extracts and their isolated compounds are essential to combat increasing resistance rates, concern for the environment and food safety, the unacceptability and the high cost of synthetic pyrethroids. These studies have exposed an array of botanical insecticides containing a wide spectrum of bioactive insecticides, larvicides and carcinogenic inhibitors.

In the light of earlier literature, it is known that larvicides play a vital role in controlling mosquitoes in their breeding sites, but still vectors resistance to them remains unanswered. In addition they show a negative impact in areas of beneficial and non-target organisms. In view of the recently increased interest in developing plant origin insecticides as an alternative to chemical insecticide, this study was undertaken to assess the larvicidal potential of the extracts from the medicinal plant against two medically important species of malaria vector, *An. stephensi* and filarial vector, *Cx. quinquefasciatus*.

MATERIALS AND METHODS

Collection of plant materials

The leaf of A. *vasica* Nees (Acanthaceae), bark of *A. squamosa* Linn. (Annonaceae), leaf and flower of *C. auriculata* Linn. (Cesalpinaceae), leaf and root of *H. indicus* R.Br. (Asclepiadaceae), leaf of *H. javanica* Linn. (Apiaceae), leaf of *Papaver somniferum* Linn. (Papaveraceae), *Pavonia zeylanica* Cav. (Malvaceae), *Solanum indicum* Linn. (Solanaceae), *S. torvum* Swartz (Solanaceae), and *V. negundo* Linn. (Verbenaceae) were collected from Javadhu Hills, Tiruvannamalai district (12°36'10N, 078°53'07E, altitude 705 m) and Chitheri Hills, Dharmapuri district (11°53'28"N, 078°30'26"E, altitude 959), Tamil Nadu, India in March 2008 and 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.

Preparation of stock solution of plant extracts

The dried (7-10 days in the shade at the environmental temperatures, 27-37ºC day time) stem bark (750g), leaf (550g), flower (450g) and seed (600g) were powdered mechanically using commercial electrical stainless steel blender (Model:ICB-500. 1.5L Capacity, 6 angle blades, 2 speed w/ pulse and made of Stainless Steel Blade. Libra Appliances pvt.lt, Baroda, India) and extracted with hexane 95% 1400 ml, (Fine chemical manufactures Mumbai, India), chloroform 99% 2800 ml, (SRL-Sisco Research Laboratories pvt.ltd, Mumbai), ethyl acetate 98% 3100 ml, (Qualigens Thermo Electron LLS India pvt. Ltd, Navi Mumbai), acetone 99% 1600 ml, (Qualigens Thermo Electron LLS India pvt. Ltd, Navi Mumbai) and methanol 99.5% 4400ml, (Qualigens GlaxoSmithKline pharmaceuticals limited, Mumbai) in a soxhlet apparatus (boiling point range 60–80ºC) for 8 h. The extract was concentrated under reduced pressure 22 – 26 mm Hg at 45ºC and the residue obtained was stored at 4ºC. One gram of crude extract was first dissolved in 100 ml of acetone (stock solution). From the stock solution, 400 – 1.25 ppm were prepared with dechlorinated tap water. 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 and polysorbate 80.

Mosquito culture

Anopheles stephensi and *Cx. quinquefasciatus* larvae were collected from stagnant water area of Melvisharam (12º 56' 23" N, 79º 14' 23" E) and identified Dr. V. Rajagopal, Senior Entomologist, Zonal Entomological Research Centre, Vellore (12º 55' 48" N, 79º 7' 48" E), Tamil Nadu, to start the colony, and larvae were kept in plastic and enamel trays containing tap water. They were maintained, and the experiments were carried out, at 27 ± 2ºC and 75–85% relative humidity under 14:10 light and dark cycles. Larvae were fed a diet of Brewers yeast, dog biscuits and algae collected from ponds in a ratio of 3:1:1, respectively. Pupae were transferred from the trays to a cup containing tap water and were maintained in our insectary (45 x 45 x 40cm) where adults emerged. Adults were maintained in glass cages and continuously provided with 10% sucrose solution in a jar with a cotton wick. On day five, the adults were given a blood meal from a pigeon placed in resting cages overnight for blood feeding by females. Glass petridishes with 50ml of tap water lined with filter paper were kept inside the cage for oviposition (Kamaraj *et al.*, 2009).

Larvicidal bioassay

During screening in the laboratory trial, the larvae of *An. stephensi* and *Cx. quinquefasciatus* were collected from the insect rearing cage and identified in Zonal Entomological Research Centre, Vellore. One gram of crude extract was first dissolved in 100 ml of acetone (stock solution). From the stock solution, 1000 ppm was prepared with dechlorinated tap water. Polysorbate 80 (Qualigens) was used as an emulsifier at the concentration of 0.05% in the final test solution. The larvicidal activity was assessed by the procedure of WHO (1996) with some modification and as per the method of Rahuman *et al.* (2000). For bioassay test, larvae were taken in five batches of twenty in 249 ml of water and 1.0 ml of the desired plant extract concentration. The control was set up with acetone and polysorbate 80. The numbers of dead larvae were counted after 24 hrs of exposure and the percentage mortality was reported from the average of five replicates. The control was set up with acetone, polysorbate 80 and distilled water. The experimental media, in which 100% mortality of larvae occurs alone, were selected for a dose response bioassay.

Dose - response bioassay

From the stock solution, different concentrations ranging from 1.25 to 400 ppm were prepared. Based on the screening results, crude different solvent leaf, flower, seed, bark, and root extracts of *A. vasica*, *A. squamosa*, *C. auriculata*, *H.*

indicus, *H. javanica*, *P. somniferum*, *P. zeylanica*, *S. indicum*, *S. torvum*, and *V. negundo* were subjected to dose response bioassay for larvicidal activity against the larvae of *An. stephensi and Cx. quinquefasciatus*. The numbers of dead larvae were counted after 24 hrs of exposure, and the percentage mortality was reported from the average of five replicates. However, at the end of 24 hrs the selected test samples turned out to be equal in their toxic potential.

Statistical analysis

The average larval 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 software developed (Reddy *et al*., 1992). Results with $p<0.05$ were considered to be statistically significant.

RESULTS AND DISCUSSION

The screening is a better means of evaluation of the potential larvicidal activity of plants popularly used for this purpose. The effect of the leaf, flower, bark and root hexane, chloroform, ethyl acetate, acetone and methanol extracts of *A. vasica*, *A. squamosa*, *C. auriculata*, *H. indicus*, *H. javanica*, *P. somniferum*, *P. zeylanica*, *S. indicum*, *S. torvum*, and *V. negundo* were tested at 1000 ppm and showed activity against the fourth instar larvae of *An. stephensi* and *Cx. quinquefasciatus* (Table 1). All plant extracts showed moderate larvicidal effects after 24 h; however, the highest larval mortality was found in leaf acetone of *An. vasica*, bark ethyl acetate of *An. squamosa* against the fourth instar larvae of *An. stephensi* (LC_{50} = 18.20 and 25.18 ppm; LC_{90} = 96.33 and 94.04 ppm) and against the fourth instar larvae of *Cx. quinquefasciatus* (LC_{50} = 27.24 and 20.56 ppm; LC_{90} = 70.38 and 82.99 ppm) (Table 2).

Nowadays, mosquito control is mostly directed against larvae and only against adults when necessary. This is because the fight against adult is temporary, unsatisfactory and pollutes the environment, while larval treatment is more localized in time and space resulting in less-dangerous outcomes. Larval control can be an effective control tool due to the low mobility of larval mosquitoes, especially where the principal breeding habitats are man-made and can be easily identified (Howard *et al*., 2007).

The screening of plant extract is a better means of evaluation of the potential parasitic activity of plants popularly used for this purpose. The mode of treatment and observations concerning mosquito larvicidal activity are given in Tables 1 and 2. It is evident from our results that a rise in the concentration of plant extracts was the main cause of mortality in *An. stephensi* and *Cx. quinquefasciatus* larvae. In the present observation the leaf acetone extracts of *A. vasica* and bark ethyl acetate extract of *A. squamosa* showed the LC_{50} values of 18.20, 28.18, 27.24 and 43.07 ppm against *An. stephensi* and *Cx. quinquefasciatus* respectively.

The obtained results revealed the larvicidal effect of ten plants corresponding to different botanical families on *An. stephensi* and *Cx. quinquefasciatus*. The highest larval mortality was found in leaf acetone and methanol of *Canna indica* ($LC_{(50)}$ = 29.62 and 40.77ppm; $LC_{(90)}$ = 148.55 and 165.00 ppm) against second instar larvae ($LC_{(50)}$ = 121.88 and 69.76, ppm; $LC_{(90)}$ = 624.35and 304.27ppm) and against fourth instar larvae of methanol and petroleum ether extracts of *Ipomoea carnea* ($LC_{(50)}$ = 41.82 and 39.32 ppm; $LC_{(90)}$ = 423.76 and 176.39 ppm) against second instar larvae ($LC_{(50)}$ = 163.81 and 41.75 ppm; $LC_{(90)}$ = 627.38 and 162.63 ppm) and against fourth instar larvae of *Cx. quinquefasciatus*, respectively (Rahuman *et al*., 2009). Kihampa *et al*. (2009) reported that the stem bark petroleum ether, chloroform and methanol crude extracts of *A. squamosa* showed activity against

III/IV instar larvae of *Anopheles gambiae* with LC_{50} values of 50, 17 and 24 ppm respectively. Previous studies have shown that the alkaloids isolated from *A. squamosa* have shown larvicidal growth-regulating activities against *An. stephensi* at concentrations of 50 to 200 ppm and the mortality in the larvae, pupae and adults produced about a 52-92% decrease in the laboratory experiment (Saxena *et al.*, 1993). The secondary compounds of *An. squamosa* are a vast repository of compounds with a wide range of biological activities, such as palmitone and isomeric hydroxy ketones Shanker *et al.* (2007), annonaceous acetogenins Hopp *et al.* (1998) were well documented. Therefore, the observed larvicidal activity of the stem bark extract was considered to have been exerted by such compounds.

The results from these trials indicate that the flower methanol extract of *C. auriculata* with LC_{50} values of 33.74 and 51.22 ppm against *An. stephensi* and *Cx. quinquefasciatus*.

The ethanolic leaf extract of *Cassia obtusifolia* had significant larvicidal effect against *An. stephensi* with LC_{50} and LC_{90} values were 52.2 and 108.7 mg/l, respectively (Rajkumar & Jebanesan, 2009) and the emodin compound was isolated from seeds and showed the LC_{50} values of

Table 1. Larvicidal activity of crude plant extracts against fourth instar larvae of *Anopheles stephensi* and *Culex quinquefasciatus* at 1000 ppm

Botanical names	Plant Parts	Species	% Mortality * ± SD				
			HE	CH	EA	AC	ME
Adhatoda vasica	LS	*A. stephensi*	78±2.824	88±2.742	71±3.051	100±0.000	100±0.000
		C. quinquefasciatus	78±2.126	91±2.144	62±2.169	100±0.000	96±2.842
Annona squamosa	BK	*A. stephensi*	84±2.280	100±0.000	68±3.049	79±0.577	81±1.924
		C. quinquefasciatus	81±2.588	100±0.000	82±1.140	75±1.581	80±1.673
Cassia auriculata	LS	*A. stephensi*	77±2.702	87±1.140	85±1.581	78±2.408	100±0.000
		C. quinquefasciatus	87±1.140	85±1.581	74±1.304	76±2.863	100±0.000
	FL	*A. stephenasi*	69±1.789	79±2.588	81±1.924	83±1.957	100±0.000
		C. quinquefasciatus	82±1.172	80±1.581	75±2.915	73±2.766	100±0.000
Hemidesmus indicus	LS	*A. stephensi*	42±2.302	64±2.716	89±2.025	90±1.672	100±0.000
		C. quinquefasciatus	53±1.601	44±1.624	72±1.462	100±0.000	100±0.000
	RT	*A. stephensi*	74±2.842	82±3.422	100±0.000	94±2.264	88±3.262
		C. quinquefasciatus	84±3.262	76±2.354	96±2.524	88±3.269	92±2.846
Hydrocotyle javanica	LS	*A. stephensi*	79±2.147	69±2.212	100±0.000	68±2.274	88±1.517
		C. quinquefasciatus	72±2.322	71±2.375	100±0.000	78±2.363	75±2.333
Papaver somniferum	LS	*A. stephensi*	42±2.570	66±1.638	80±3.462	34±3.358	43±2.563
		C. quinquefasciatus	31±2.023	40±2.426	74±2.949	54±1.452	45±1.563
Pavonia zeylanica	LS	*A. stephensi*	93±2.129	67±3.135	82±2.624	96±3.526	100±0.000
		C. quinquefasciatus	92±2.628	82±1.681	74±2.652	100±0.000	100±0.000
Solanum indicum	LS	*A. stephensi*	22±1.342	48±1.615	47±1.703	65±2.949	85±3.272
		C. quinquefasciatus	67±1.629	53±2.483	64±2.824	75±2.487	93±3.224
Solanum torvum	LS	*A. stephensi*	72±1.140	84±1.924	87±2.074	85±1.581	100±0.000
		C. quinquefasciatus	80±2.236	78±2.408	74±1.924	83±1.517	100±0.000
	SD	*A. stephensi*	84±2.280	73±2.966	82±2.408	87±2.074	100±0.000
		C. quinquefasciatus	78±2.302	66±1.861	73±1.844	86±0.837	100±0.000
Vitex negundo	LS	*A. stephensi*	100±0.000	81±0.837	79±2.280	80±1.663	83±2.014
		C. quinquefasciatus	100±0.000	75±1.647	80±1.581	70±1.846	78±2.119

LS=Leaves; BK=Bark; FL=Flower; RT=Root; SD=Seed; Control= Nil mortality.
* Mean value of five replicates. HE=Hexane; CH=Chloroform; EA =Ethyl acetate; AC=Acetone; ME = Methanol.

Table 2. Larvicidal activity of different solvent crude extracts against fourth instar larvae of *Anopheles stephensi* and *Culex quinquefasciatus*

Plant species	Plant Parts	Solvents	Species	LC_{50} (ppm) (UCL- LCL)	LC_{90} (ppm) (UCL -LCL)	Slope±SE	χ2(df = 4)
Adhatoda vasica	LS	Acetone	*A. stephensi*	18.20 (22.11-14.28)	96.33 (131.75-60.91)	4.881±2.521	9.04
			C. quinquefasciatus	27.24 (31.29-23.17)	70.38 (87.49-53.25)	1.325±0.211	4.16
	LS	Methanol	*A. stephensi*	41.42 (49.46-33.38)	192.77 (256.36-129.17)	1.827±0.732	11.27
Annona squamosa	BK	Ethyl acetate	*A. stephensi*	25.18 (28.34-22.03)	94.04 (112.27-75.81)	2.157±0.584	5.93
			C. quinquefasciatus	43.07 (48.62-37.53)	172.27 (208.22-136.31)	2.680±0.872	13.63
Cassia auriculata	LS	Methanol	*A. stephensi*	43.66 (49.13-38.18)	164.70 (197.06-132.34)	3.839±0.475	10.28
			C. quinquefasciatus	58.30 (65.41-51.19)	210.69 (252.26-169.13)	7.678±0.652	12.2
	FL		*A. stephensi*	33.74 (38.03-29.46)	133.07 (160.23-105.92)	4.629±0.258	8.54
			C. quinquefasciatus	51.22 (5782-44.61)	205.12 (248.58-61.67)	5.816±2.365	5.48
Hemidesmus indicus	LS	Acetone	*C. quinquefasciatus*	108.50 (124.72-96.14)	646.36 (743.15-579.63)	7.198±1.585	9.61
	LS	Methanol	*A. stephensi*	155.19 (219.94-106.45)	607.86 (726.04-489.69)	4.650±0.698	11.46
			C.quinquefasciatus	192.83 (224.31-174.35)	760.01 (955.16-605.24)	4.289±1.332	10.80
	RT	Ethyl acetate	*A. stephensi*	217.61 (234.07-87.15)	784.16 (985.16-645.64)	4.182±0.582	12.89
Hydrocotyle javanica	LS	Ethyl acetate	*A. stephensi*	88.28 (109.05-57.55)	560.75 (678.92-484.84)	3.907±0.852	7.46
			C. quinquefasciatus	96.72 (126.92-74.51)	303.20 (420.79-215.62)	6.512±0.678	13.82
Pavonia zeylanica	LS	Acetone	*C. quinquefasciatus*	348.62 (398.64-268.63)	1497.09 (1782.43-1011.72)	5.079±0.563	9.49
	LS	Methanol	*A. stephensi*	142.63 (185.54-69.11)	495.82 (584.00-336.46)	4.063±0.254	10.72
			C. quinquefasciatus	192.84 (272.99-82.79)	565.40 (746.54-454.28)	5.093±2.874	10.57
Solanum torvum	LS	Methanol	*A. stephensi*	29.65 (33.70-25.61)	133.01 (102.71-103.32)	2.547±0.421	11.72
			C. quinquefasciatus	20.56 (23.25-17.87)	82.99 (100.18-65.79)	3.599±0.365	7.36
	SD		*A. stephensi*	31.30 (35.42-27.17)	130.65 (157.20-102.89)	4.798±1.235	4.75
			C. quinquefasciatus	25.60 (28.96-22.24)	105.52 (127.45-83.59)	5.998±0.842	5.82
Vitex negundo	LS	Hexane	*A. stephensi*	33.83 (38.03-29.47)	127.47 (152.53-102.40)	5.502±0.541	7.65
			C. quinquefasciatus	56.981 (64.06-49.91)	210.68 (251.96-169.41)	4.763±2.365	9.26

LS=Leaves; BK=Bark; FL=Flower; RT=Root; SD= Seed; Control = Nil mortality. Significant at $P< 0.05$ level.LC_{50}= Lethal concentration that kills 50% of the exposed larvae; LC_{90} =Lethal concentration that kills 90% of the exposed larvae; UCL=Upper confidence Limit; LCL=Lower confidence Limit; χ^2 =Chi-square; df = degree of freedom.

1.4, 1.9, and 2.2 mg/L against *Culex pipiens pallens*, *Ae. aegypti*, and *Aedes togoi*, respectively (Yang *et al.*, 2003). Compared with earlier authors report, our results revealed that the experimental plant extracts were effective to control *An. stephensi* and *Cx. quinquefasciatus*.

Larval mortality was 100% with the use of 5% concentration of root extract of *H. indicus*, leaves extracts of *Gymnema sylvestre* and *Eclipta prostrata* after 2 days (Khanna & Kannabiran, 2007). Larvicidal efficacy of leaf methanol extracts of *P. zeylanica* and *Acacia ferruginea* were tested against the late third instar larvae of *Cx. quinquefasciatus* with LC_{50} values of 2214.7 and 5362.6 ppm, respectively (Vahitha *et al.*, 2002). The peel methanol extract of *Citrus sinensis* and the leaf and flower ethyl acetate extracts of *Ocimum canum* were tested against the larvae of *An. stephensi* (LC_{50} = 95.74, 101.53, 28.96, LC_{90} = 303.20, 492.43 and 168.05 ppm), respectively (Kamaraj *et al.*, 2008). The present findings revealed that the methanol extract of *S. torvum* was less active compared with the petroleum ether extracts of *S. xanthocarpum*.

Karunamoorthi *et al.* (2008) reported that the petroleum ether (60-80 degrees C) extracts of the leaves of *V. negundo* were evaluated for larvicidal activity against larval stages of *C. tritaeniorhynchus* in the laboratory with LC_{50} and LC_{90} values of 2.4883 and 5.1883 mg/l, respectively. The methanol leaf extracts of *V. negundo*, *V. trifolia*, *V. peduncularis* and *V. altissima* possessed varying levels of larvicidal activity on *Cx. quinquefasciatus* and *An. stephensi* and found with LC_{50} value of 212.57, 41.41, 76.28 and 128.04 ppm respectively (Pushpalatha & Muthukrishnan, 1995).

The results of this study indicate that the plant extracts would likely to be toxic to the target species tested at the concentrations necessary for controlling the fourth instar mosquito larvae. An attempt has been made to evaluate the toxic effect of plant extracts against *An. stephensi and Cx. quinquefasciatus* larvae. Our results suggest that the leaf methanol extract of *S. torvum* and bark ethyl acetate extract of *A. squamosa* have the potential for use to control mosquitoes. Further studies are in progress to evaluate the effect of purified extract on larvicidal activity. The purified plant metabolite of the leaf methanol extracts of *S. torvum* and bark ethyl acetate extract of *A. squamosa* may be used as environment friendly and sustainable insecticides to combat mosquitoes.

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