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**LARVICIDAL PROPERTIES OF *CAESALPINIA PULCHERRIMA*
 (FAMILY: FABACEAE) AGAINST *CULEX TRITAENIORHYNCHUS*, *AEDES
 ALBOPICTUS* AND *ANOPHELES SUBPICTUS* (DIPTERA: CULICIDAE)**

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ABSTRACT

Mosquito-borne diseases have an economic impact, including loss in commercial and labor outputs, particularly in countries with tropical and subtropical climates; however, no part of the world is free from vector-borne diseases. In mosquito control programs, botanical origin may have the potential to be used successfully as eggs, larvae, and adult. The larvicidal activity of crude benzene and ethyl acetate extracts of leaves of *Caesalpinia pulcherrima* were assayed for their toxicity against three important vector mosquitoes, viz., *Culex tritaeniorhynchus*, *Aedes albopictus* and *Anopheles subpictus* (Diptera: Culicidae). The larval mortality was observed after 24 h of exposure. All extracts showed moderate larvicidal effects; however, the highest larval mortality was found in benzene extract of against the larvae of *Cx. tritaeniorhynchus*, *Ae. albopictus* and *An. subpictus* with the LC₅₀ and LC₉₀ values were 150.47, 135.24, and 119.27 ppm and 282.57, 261.55, and 243.37 ppm, respectively. These results suggest that the leaf solvent plant extracts have the potential to be used as an ideal eco-friendly approach for the control of mosquitoes. This is the first report on the mosquito larvicidal activity of the reported *Caesalpinia pulcherrima* plant.

Keywords: Larvicidal activity, *Culex tritaeniorhynchus*, *Aedes albopictus*, *Anopheles subpictus*, *Caesalpinia pulcherrima*.

INTRODUCTION

Mosquitoes are the vectors of a number of human and zoonotic disease pathogens affecting human and animal hosts, including those that cause malaria, filariasis, Japanese encephalitis (JE), and dengue and yellow fevers. In view of the fact that mosquitoes develop genetic resistance to synthetic insecticides and even to bio pesticides such as *Bacillus sphaericus* the application of easily degradable botanicals for the control of mosquitoes is recommended. Mosquitoes are cosmopolitan and common and transmit a variety of diseases, as well as

being nuisance pests (Mullen *et al.*, 2009). Continued and repeated use of conventional mosquitocides, such as organophosphorus (OP) and carbamate insecticides, insect growth regulators and bacterial larvicides (Rozendaal, 1997; WHO, 2006) has often resulted in the widespread development of resistance and has undesirable effects on non-target organisms. In particular, the use of conventional synthetic insecticides for the control of rice and horticultural and veterinary pests has accelerated these adverse effects. Increasing public concern for the environmental effects of insecticides, groundwater

contamination, human health effects and undesirable effects on non-target organisms (Rozendaal, 1997; Cooper, 1991).

Mosquito control has become increasingly difficult because of the indiscriminate uses of synthetic chemical insecticides which have an adverse impact on the environment and disturb the ecological balance. The majority of the chemical pesticides is harmful to man and animals, some of which are not easily degradable and spreading toxic effects. The increased use of these insecticides may enter into the food chain, and thereby, the liver, kidney, etc., may be irreversibly damaged. They even result in the mutation of genes and these changes become prominent only after a few generations (Ghosh, 1991). Chemical insecticides are very costly. In larval mosquito control, application of insecticides in ponds, wells, and other water bodies may cause health hazards to human and larvivorous fishes. Nowadays, mosquito coils containing synthetic pyrethroids and other organophosphorus compounds cause so many side effects, such as breathing problem, eye irritation, headache, asthma, itching, and sneezing to the users. With the use of mosquito repellent, people complained of ill health effects and sometimes required medical treatment. In addition, pests were becoming resistant to chemical treatments. Indoor residual spraying of insecticides stains the walls and leaves a long lasting unpleasant odor. These problems have highlighted the need for the development of new strategies for selective mosquito control. Extracts or essential oils from plants may be alternative sources of mosquito larval control agents, since they constitute a rich source of bioactive compounds that are biodegradable into nontoxic products and potentially suitable for use in the control of mosquito larvae. In fact, many researchers have reported on the effectiveness of plant extracts or essential oils against mosquito larvae (Amer and Mehlhorn, 2006a, b, c; Govindarajan, 2010a, b).

Phytochemicals are advantageous due to their eco-safety, target-specificity, non-development of resistance, reduced number of applications, higher acceptability, and suitability for rural areas. Botanicals can be used as an alternative to synthetic insecticides or along with other insecticides under integrated vector control programs. The plant produces phytochemicals, which are used as insecticides for killing larvae or adult mosquitoes or as repellents for protection against mosquito bites. Phytochemicals obtained from the whole plant or specific part of the plant by the extraction with different types of solvent such as aqueous, methanol,

chloroform, benzene, acetone, etc., depending on the polarity of the phytochemical. Some phytochemicals act as a toxicant (insecticide) both against adult as well as larval stages of mosquitoes, while others interfere with growth and growth inhibitor or with the reproduction or produce an olfactory stimulus, thus acting as repellent or attractant (Markouk *et al.*, 2001).

Plants may be a source of alternative agents for control of mosquitoes because they are rich in bioactive chemicals, are active against a limited number of species including specific target insects, and are biodegradable. They are potentially suitable for use in integrated pest management programs (Alkofahi *et al.*, 1989): the mosquito larvicidal properties of leaf and seed extract of plant *Agave Americana* (Dharmshaktu *et al.*, 1987); the mosquito larvicidal activity in the extract of *Tagetes minuta* flowers against *Aedes aegypti* (Green *et al.*, 1991); the methanolic fraction of leaves of *Mentha piperita*, *Phyllanthus niruri*, *Leucas aspera*, and *Vitex negundo* against larvae of *Culex quinquefasciatus* (Pandian *et al.*, 1994); the methanolic extracts of *Solanum suratense*, *Azadirachta indica*, and *Hydrocotyle javanica* exhibited larvicidal activity against *Culex quinquefasciatus* (Muthukrishnan *et al.*, 1997); the benzene and methanol extracts of *Artemisia vulgaris* has been repellent activity against *Aedes Aegypti* (Yit *et al.*, 1985); the *Zanthoxylum armatus*, *Zanthoxylum alatum* (Rutaceae), *Azadirachta indica* (Mailiaceae), and *Curcuma aromatica* (Zingiberaceae) were possessing repellent properties against mosquitoes (Das *et al.*, 2000); the repellent activity of active compound *Octacosane* from *Moschosma polystachyum* against the vector *Culex quinquefasciatus* (Rajkumar and Jebanesan, 2004); and the essential oil of *Zingiber officinalis* as a mosquito larvicidal and repellent agent against the filarial vector *Culex quinquefasciatus* (Pushpanathan *et al.*, 2008).

Today, more than 1,005 plant species are found to possess insecticidal properties, 384 contain antifeedants, 297 contain repellents, and 27 contain attractants and possess growth inhibitors (Jayaraj, 1993). The Larvicidal efficacy of the crude leaf extract of *Ficus benghalensis* with three different solvents like methanol, benzene, and acetone were tested against the early second, third, and fourth instar larvae of *Culex quinquefasciatus*, *Aedes aegypti*, and *Anopheles stephensi* (Govindarajan, 2010c). The acetone, chloroform, ethyl acetate, hexane, and methanol leaf extracts of *Acalypha indica*, *Achyranthes aspera*, *L. aspera*, *Morinda tinctoria*, and *Ocimum sanctum* were studied against

the early fourth instar larvae of *Aedes aegypti* and *Culex quinquefasciatus* (Bagavan *et al.*, 2008a). The larvicidal activity of crude hexane, ethyl acetate, petroleum ether, acetone, and methanol extracts of the leaf of five species of cucurbitaceous plants, *Citrullus colocynthis*, *Coccinia indica*, *Cucumis sativus*, *Momordica charantia*, and *Trichosanthes anguina*, were tested against the early fourth instar larvae of *Aedes aegypti* L. and *Culex quinquefasciatus* (Rahuman *et al.*, 2008).

Govindarajan (2009) reported that the leaf methanol, benzene, and acetone extracts of *Cassia fistula* were studied for the larvicidal, ovicidal, and repellent activities against *Aedes aegypti*. The leaf extract of *Acalypha indica* with different solvents, viz., benzene, chloroform, ethyl acetate, and methanol, was tested for larvicidal, ovicidal activity, and oviposition attractancy against *Anopheles stephensi* (Govindarajan *et al.*, 2008a). Mullai *et al.* (2008) have reported that the leaf extract of *Citrullus vulgaris* with different solvents, viz., benzene, petroleum ether, ethyl acetate, and methanol, were tested for larvicidal, ovicidal, repellent, and insect growth regulatory activities against *Anopheles stephensi*. Ovicidal effects of the seed extract of *Atriplex canescens* were reported against *Culex quinquefasciatus* (Ouda *et al.*, 1998). Su and Mulla (1998) reported the ovicidal activity of the Neem product azadirachtin against the mosquitoes *Culex tarsalis* and *Culex quinquefasciatus*.

In view of the recent increased interest in developing plant origin insecticides as an alternative to chemical insecticide, this study was undertaken to assess the larvicidal potential of the different solvent crude extracts from the medicinal plant *Caesalpinia pulcherrima* against the medically important mosquito vectors, *Cx. tritaeniorhynchus*, *Ae. albopictus* and *An. subpictus*.

MATERIALS AND METHODS

Collection of plants: Fully developed leaves of the *Caesalpinia pulcherrima* were collected from in and around Annamalai University Campus, Annamalainagar, 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.

Extraction: The leaves were washed with tap water, shade-dried, and finely ground. The finely ground

plant leaf powder (3.0 kg/ solvent) was loaded in Soxhlet apparatus and was extracted with two different solvents, namely, benzene and ethyl acetate, individually (Vogel, 1978). 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 acetone. From this stock solution, different concentrations were prepared and these solutions were used for larvicidal bioassays.

Test organisms: The mosquitoes, *Cx. tritaeniorhynchus*, *Ae. albopictus* and *An. subpictus*. 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 a blood meal. Mosquitoes were held at 28±2°C, 70–85% relative humidity, with a photo period of 14-h light and 10-h dark.

Larvicidal bioassay: The larvicidal activity of the plants crude extracts was evaluated as per the method recommended by World Health Organization (2005). Batches of 25 third instar larvae were transferred to small disposable test cups, each containing 200 ml of water. The appropriate volume of dilution was added to 200 ml water in the cups to obtain the desired target dosage, starting with the lowest concentration. Six replicates were set up for each concentration, and an equal number of controls were set up simultaneously using tap water. To this, 1 ml of appropriate solvent was added. The LC₅₀ value was calculated after 24 h by probit analysis (Finney, 1979).

Statistical analysis: The average larval mortality data were subjected to probit analysis for calculating LC₅₀, LC₉₀, 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 SPSS12.0 (Statistical Package of Social Sciences) software. Results with p<0.05 were considered to be statistically significant.

RESULTS

The results of the larvicidal activity of benzene and ethyl acetate extract of *Caesalpinia pulcherrima* against the larvae of three important vector mosquitoes, viz., *Cx. tritaeniorhynchus*, *Ae. albopictus* and *An. subpictus*. are presented in Tables 1 and 2. The highest larvicidal activity was observed in benzene extract of *Caesalpinia pulcherrima* against *Cx. tritaeniorhynchus*, *Ae. albopictus* and *An.*

subpictus, with the LC₅₀ and LC₉₀ values were 150.47, 135.24, and 119.27 ppm and 282.57, 261.55 and 243.37 ppm respectively. The chi-square values are significant at p < 0.05 level. The chi-square values in the bioassays indicated probably the heterogeneity of the test population. The 95% confidence limits

(LC₅₀ (LCL–UCL)) and (LC₉₀ (LCL– UCL)) were also calculated. In this observation, these one plant crude extracts gave protection against mosquito bites without any allergic reaction to the test person, and also, the repellent activity is dependent on the strength of the plant extracts.

Table 1. Larvicidal activity of crude benzene extract of *Caesalpinia pulcherrima* against *Culex tritaeniorhynchus*, *Aedes albopictus* and *Anopheles subpictus*.

Mosquitoes	Concentration (mg/ L)	% of mortality±SD	LC ₅₀ (LCL-UCL) (mg/L)	LC ₉₀ (LCL-UCL) (mg/L)	χ ²
<i>Culex tritaeniorhynchus</i>	60	25.2±0.8	150.47 (111.76-188.40)	282.57 (234.43-381.09)	16.641*
	120	45.3±1.2			
	180	64.5±1.6			
	240	73.0±1.8			
	300	93.2±0.8			
	Control	0.0±0.0			
<i>Aedes albopictus</i>	60	30.6±0.2	135.24 (93.00-173.70)	261.55 (214.53-359.68)	18.949*
	120	48.3±0.6			
	180	73.0±0.4			
	240	79.2±0.8			
	300	94.5±2.0			
	Control	0.0±0.0			
<i>Anopheles subpictus</i>	60	38.0±0.8	119.27 (66.64-162.58)	243.37 (193.33-360.19)	24.511*
	120	57.2±1.6			
	180	76.3±1.4			
	240	83.5±1.2			
	300	96.1±0.8			
	Control	0.0±0.0			

Each value ($\bar{X} \pm S.D.$) represents mean of six values . LCL- Lower Confidence Limit, UCL- Upper Confidence Limit. *Significant at P<0.05 level.

Table 2. Larvicidal activity of crude ethyl acetate extract of *Caesalpinia pulcherrima* against *Culex tritaeniorhynchus*, *Aedes albopictus* and *Anopheles subpictus*.

Mosquitoes	Concentration (mg/ L)	% of mortality±SD	LC ₅₀ (LCL-UCL) (mg/L)	LC ₉₀ (LCL-UCL) (mg/L)	χ ²
<i>Culex tritaeniorhynchus</i>	60	23.4±0.8	158.17 (120.54-196.34)	296.61 (246.85-397.89)	15.322*
	120	43.5±1.6			
	180	64.5±1.2			
	240	70.6±1.8			
	300	90.1±0.8			
	Control	0.0±0.0			
<i>Aedes albopictus</i>	60	28.5±0.6	142.43 (101.93-180.57)	261.55 (214.53-359.68)	17.661*
	120	46.2±0.8			
	180	69.3±0.4			
	240	76.4±0.2			
	300	93.6±2.0			
	Control	0.0±0.0			
<i>Anopheles subpictus</i>	60	28.5±0.8	127.80 (78.70-170.04)	254.79 (204.90-367.21)	22.570*
	120	46.2±1.2			
	180	69.3±1.6			
	240	76.4±2.0			
	300	93.6±0.8			
	Control	0.0±0.0			

Each value ($\bar{X} \pm S.D.$) represents mean of six values. LCL- Lower Confidence Limit, UCL- Upper Confidence Limit. *Significant at $P < 0.05$ level.

DISCUSSION

Today, the environmental safety of an insecticide is considered to be of paramount importance. An insecticide does not have to cause high mortality on target organisms in order to be acceptable (Kabar and Gichia, 2001). Phytochemicals may serve as suitable alternatives to synthetic insecticides in future as they are relatively safe and inexpensive and are readily available in many areas of the world. According to Bowers *et al.* (1995), the screening of locally available medicinal plants for mosquito control would generate local employment, reduce dependence on expensive imported products, and stimulate local efforts to enhance public health. Different parts of plants contain a complex of chemicals with unique biological activity (Govindarajan *et al.* 2008a, b, c), which is thought to be due to toxins and secondary metabolites which act as mosquitocidal agent (Niraimathi *et al.* 2010). Our result showed that the crude benzene and ethyl acetate extracts of leaves of *Caesalpinia pulcherrima* have significant larvicidal, against three important vector mosquitoes, viz., *Cx. tritaeniorhynchus*, *Ae. albopictus* and *An. subpictus*. This result is also comparable to earlier reports of Singh *et al.* (2003) who observed the larvicidal activity of *Ocimum canum* oil against vector mosquitoes, namely, *Aedes aegypti* and *Culex quinquefasciatus* (LC_{50} , 301 ppm) and *Anopheles stephensi* (234 ppm). Larvicidal efficacy of leaf methanol extracts of *Pavonia zeylanica* and *Acacia ferruginea* were tested against the late third instar larvae of *Culex quinquefasciatus* with LC_{50} values of 2,214.7 and 5,362.6 ppm, respectively (Vahitha *et al.* 2002). The peel methanol extract of *Citrus sinensis* and the leaf and flower ethyl acetate extracts of *O. canum* were tested against the larvae of *Anopheles stephensi* (LC_{50} =95.74, 101.53, and 28.96 ppm; LC_{90} =303.20, 492.43 and 168.05 ppm), respectively (Kamaraj *et al.* 2008a).

Karunamoorthi *et al.* (2008) reported that the petroleum ether (60–80°C) extracts of the leaves of *V. negundo* were evaluated for larvicidal activity against larval stages of *Culex 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*, *Vitex trifolia*, *Vitex peduncularis*, and *Vitex altissima* possessed varying levels of larvicidal activity on *Culex quinquefasciatus* and *Anopheles stephensi* and were found with LC_{50} value of 212.57, 41.41, 76.28, and

128.04 ppm, respectively (Pushpalatha and Muthukrishnan 1995). The ethanolic leaf extract of *Cassia obtusifolia* had a significant larvicidal effect against *Anopheles stephensi* with Table 5 Ovicidal activity of *Caesalpinia pulcherrima* plant extracts against *Culex quinquefasciatus*, *Aedes aegypti*, and *Anopheles stephensi*. The compound was isolated from seeds and showed the LC_{50} values of 1.4, 1.9, and 2.2 mg/L against *Culex pipiens*, *Aedes aegypti*, and *Aedes togoi*, respectively (Yang *et al.* 2003). The findings of our results are in corroboration with important findings of Sumroiphon *et al.* (2006) who have reported that the effect of water extract of citrus seed extract showed LC_{50} values of 135,319.40 and 127,411.88 ppm against the larvae of *Aedes aegypti* and *Culex quinquefasciatus*.

Amer and Mehlhorn (2006a) have reported that the five most effective oils were those of *Litsea cubeba*, *Cajuput (Melaleuca leucadendron)*, *Niaouli (Melaleuca quinquenervia)*, *Violet (Viola odorata)*, and *Catnip (Nepeta cataria)*, which induced a protection time of 8 h at the maximum and a 100% repellency against *Aedes aegypti*, *Anopheles stephensi*, and *Culex quinquefasciatus*. The essential oil of *T. minuta* providing a repellency of 90% protection for 2 h against *Anopheles stephensi*, *Culex quinquefasciatus*, and *Aedes aegypti* was observed by Tyagi *et al.* (1994). Mullai and Jebanesan (2006) reported the complete ovicidal activity (100% mortality) was attained at 300 ppm for methanol, benzene, petroleum ether, and ethyl acetate extracts of *Citrullus pubescens* against *Culex quinquefasciatus*. The leaf extract of *Solanum trilobatum* reduced egg laying by gravid females of *Anopheles stephensi* from 18% to 99% compared with ethanol-treated controls at 0.01%, 0.025%, 0.05%, 0.075%, and 0.1% (Rajkumar and Jebanesan 2005). Bagavan *et al.* (2008b) have reported that peel chloroform extract of *Citrus sinensis*, leaf ethyl acetate extracts of *O. canum* and *O. sanctum*, and leaf chloroform extract of *Rhinacanthus nasutus* against the larvae of *Anopheles subpictus* (LC_{50} =58.25, 88.15, 21.67, and 40.46 ppm; LC_{90} =298.31, 528.70, 98.34, and 267.20 ppm) and peel methanol extract of *Citrus sinensis*, leaf methanol extract of *O. canum*, and ethyl acetate extracts of *O. sanctum* and *R. nasutus* against the larvae of *Culex tritaeniorhynchus* (LC_{50} =38.15, 72.40, 109.12, and 39.32 ppm; LC_{90} =184.67, 268.93, 646.62, and 176.39 ppm) respectively.

Kamaraj *et al.* (2008b) reported that the highest larval mortality was found in leaf petroleum

ether, flower methanol extracts of *Cryptocoryne auriculata*, flower methanol extracts of *L. aspera* and *R. nasutus*, leaf and seed methanol extracts of *Solanum torvum*, and leaf hexane extract of *V. negundo* against the larvae of *Anopheles subpictus* (LC₅₀=44.21, 44.69, 53.16, 41.07, 35.32, 28.90, and 44.40 ppm; LC₉₀=187.31, 188.29, 233.18, 142.66, 151.60, 121.05, and 192.11 ppm, respectively) and against the larvae of *Culex tritaeniorhynchus* (LC₅₀=69.83, 51.29, 81.24, 71.79, 44.42, 84.47, and 65.35 ppm; LC₉₀=335.26, 245.63, 300.45, 361.83, 185.09, 351.41, and 302.42 ppm, respectively). Ansari *et al.* (2005) observed the larvicidal activity of *Pinus longifolia* oil against three vector mosquitoes namely *Aedes aegypti* (LC₅₀, 82.1 ppm), *Culex quinquefasciatus* (LC₅₀, 85.7 ppm), and *Anopheles stephensi* (LC₅₀, 112.6 ppm). Cavalcanti *et al.* (2004) reported that the larvicidal activity of essential oils of Brazilian plants against *Aedes aegypti* and observed the LC₅₀ to range from 60 to 533 ppm. Prajapati *et al.* (2005) reported that the larvicidal activity of different plants essential oil showed varied LC₉₅ values against *Culex quinquefasciatus*. They were *Pimpinella anisum* (149 µg/ml), *Z. officinalis* (202 µg/ml), *Junipers macropoda* (204 µg/ml), *Cinnamomum zeylanicum* (277 µg/ml), *Curcuma longa* (292 µg/ml), *Cyperus scariosus* (408 µg/ml), *Ocimum basilicum* (315 µg/ml), *Cuminum cyminum* (344 µg/ml), and *Nigella sativa* (365 µg/ml). Rajkumar and Jebanesan (2004) studied ovicidal activity of *M. polystachyum* leaf extract against *Culex quinquefasciatus* and observed 100% egg mortality at 100 ml/l.

Larvicidal activity of crude extract of *Sida acuta* against *Culex quinquefasciatus*, *Aedes aegypti*, and *Anopheles stephensi* with LC₅₀ values ranging between 38 to 48 mg/l (Govindarajan, 2010d); the lethal concentration (LC₅₀) values of *Ficus benghalensis* against early second, third, and fourth instar larvae of *Culex quinquefasciatus*, *Aedes aegypti*, and *Anopheles stephensi* were 41.43, 58.21, and 74.32 ppm; 56.54, 70.29, and 80.85 ppm; and 60.44, 76.41, and 89.55 ppm, respectively (Govindarajan, 2010c). Rahuman *et al.* (2008b) have reported that the LC₅₀ value of petroleum ether extracts of *Jatropha curcas*, *Pedilanthus tithymaloides*, *Phyllanthus amarus*, *Euphorbia hirta*, and *Euphorbia tirucalli* were 8.79, 55.26, 90.92, 272.36, and 4.25 ppm, respectively, against *Aedes aegypti* and 11.34, 76.61, 113.40, 424.94, and 5.52 ppm, respectively, against *Culex quinquefasciatus*, and the larvicidal effect of ten plants corresponding to different botanical families on *Anopheles stephensi* and *Culex quinquefasciatus*. The highest larval

mortality was found in leaf acetone and methanol of *Canna indica* (LC₅₀=29.62 and 40.77 ppm; LC₉₀=148.55 and 165.00 ppm) against second instar larvae (LC₅₀=121.88 and 69.76, ppm; LC₉₀=624.35 and 304.27 ppm) and against fourth instar larvae of *Anopheles stephensi* and in methanol and petroleum ether extracts of *Ipomoea carnea* (LC₅₀=41.82 and 39.32 ppm; LC₉₀= 423.76 and 176.39 ppm) against second instar larvae (LC₅₀=163.81 and 41.75 ppm; LC₉₀=627.38 and 162.63 ppm) and against fourth instar larvae of *Culex quinquefasciatus*, respectively (Rahuman *et al.* 2009).

Govindarajan *et al.* (2008a) reported that the younger age groups of egg rafts or eggs showed poor hatchability rate when exposed to higher concentrations of extract and that older age groups of egg rafts or eggs showed high hatchability rate when exposed to lower concentrations of extract. The methanol containing water that served as a control showed 94% hatchability in 0–3-h-old egg rafts/eggs, but the 100% hatchability was noted in egg percentage protection was 45.8% at 1.0 mg/cm² and 59.0% at 2.5 mg/cm² for 10 h. Compared with earlier authors' reports, our results revealed that the experimental plant extracts were effective to control *Anopheles stephensi*, *Aedes aegypti*, and *Culex quinquefasciatus*.

CONCLUSION

From these results, it was concluded that the plants *Caesalpinia pulcherrima* exhibit larvicidal activities against three important vector mosquitoes. Further analysis to isolate the active compound for larval control is under way in our laboratory. The flora of India has rich aromatic plant diversity with potential for development of natural insecticides for control of mosquito and other pests.

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