



Adulticidal and repellent activities of *Melaleuca leucadendron* (L.) and *Callistemon citrinus* (Curtis) against filarial and dengue vectors

E. Pushpalatha * and K. Anju Viswan

*Biopesticides and Toxicology Division, Department of Zoology,
University of Calicut, Kerala, 673635 India
E mail: drepushpalatha@yahoo.co.in*

ABSTRACT: Essential oils extracted by steam distillation from two tropical plants viz; *Melaleuca leucadendron* (L.) and *Callistemon citrinus* (Curtis) evaluated against *Aedes aegypti* (L.) and *Culex quinquefasciatus* (Say), *in vitro* for their adulticidal and repellent potentiality showed a 100% protection up to 5 hrs after treatments. *M. leucadendron* showed 100% repellency against *Cx. quinquefasciatus* and 80.9% repellency against *Ae. aegypti* at its 8th hour of exposure. Adulticidal activities of these two essential oils observed after 24 hour exposure showed 100% adult mortality, indicating their potentiality in the control of mosquitoes. © 2013 Association for Advancement of Entomology

KEYWORDS: *Aedes aegypti*, *Culex quinquefasciatus*, *Melaleuca leucadendron*, *Callistemon citrinus*, essential oil, repellent activity, adulticidal activity.

INTRODUCTION

Mosquitoes are well known for public health importance, as they are the vectors which transmit pathogens of several diseases like malaria, chikun guniya, dengue fever, yellow fever, Japanese encephalitis and so on (Service, 1983). Mosquitoes alone transmit diseases affecting more than 700 million annually (Jang *et al.*, 2000). Man started using synthetic chemical insecticide DDT since 1940 to control mosquitoes (Metcalf and Lukaman, 1975). Indiscriminate use of synthetic insecticides lead to various environmental consequences (Agarwal *et al.*, 1981), development of genetic resistance in mosquito species (Sharma *et al.*, 1986) and disrupted natural biological control systems (Brown, 1986). Plant-derived

* Author for correspondence

essential oils can be considered as a valuable alternative for insect control (Govindarajan *et al.*, 2008).

Repellents of plant origin should be non-toxic, non-irritating and long lasting and eco-friendly in nature. Plants of terrestrial origin have been reported to be an important source of mosquito repellents (Hwang *et al.*, 1985). The essential oils from medicinal herbs in Lebanon proved as an environmentally safe measure to control the seaside mosquito (Knio *et al.*, 2008). Several essential oils from herbs act as *Culex* and *Aedes* larvicides (Sukumar *et al.*, 1991). Essentials oils from *Cannabis sativa* (Thomas *et al.*, 2000) and *Tagetes patula* L. (Dharmaggada *et al.*, 2005) were reported to have activity against *Aedes aegypti* Linn., *Anopheles stephensi* Liston and *Culex quinquefasciatus* Say. Prajapati *et al.* (2005) studied the larvicidal, adulticidal, oviposition deterrent and repellent activities of essential oils from 10 medicinal plants against *An. stephensi*, *Ae. aegypti* and *Cx. quinquefasciatus*. The undiluted oils of *Cymbopogon nardus* Linn., *Pogostemon cablin* Benth., *Syzigium aromaticum* Linn. and *Zanthoxylum limonella* Alston were the most effective and provided two hours of complete repellency (Trongtokit *et al.*, 2005).

MATERIALS AND METHODS

Target organism: Mosquito species chosen for the present study were *Ae. aegypti* and *Cx. quinquefasciatus* obtained from the laboratory culture maintained as described in Pushpalatha and Muthukrishnan (1995).

Essential Oil extraction: Leaves of *Melaleuca leucadendron* Linn. and *Callistemon citrinus* (Curtis) Skeels were collected from the field, washed with distilled water and essential oils were extracted by steam distillation in Clevenger-type apparatus (Craveiro *et al.*, 1976).

Repellency Assay: Repellency of volatile oils was evaluated using human bait technique. Each test was conducted for a period of 8hrs, depending on the response. *Ae. aegypti* was tested between 07.00 h and 15.00 h while *Cx. quinquefasciatus* was tested between 17.00 h and 01.00 h. Each oil was tested 100, 70, 50 and 10 percent concentrations. An arm of a human volunteer was covered with a paper sleeve with 3x10cm window and 0.1ml of desired concentrations of the oil was applied. The uncovered arm was exposed into a standard mosquito emergence cage having 100 hungry 4 to 5 days old female mosquitoes for one minute. Prior to the commencement of each exposure, the mosquitoes were tested for their readiness to bite by placing an untreated bare hand of a volunteer for 30 seconds. An arm of human volunteer without any oil application was kept as control. The number of incidence of landing without biting and those of biting ones were recorded at each interval until the rate of bite fell into 1 to 1.5 per minute. The duration between the application of repellent and the commencement of bite was recorded as the protection time. The percentage of repellency was calculated at the end of every test using the formula mentioned by Tawastsin *et al* (2001) viz; $(C-T/C) \times 100$ where, C is total number of mosquitoes landing or biting the control area and T is total number of mosquitoes landing or biting the treated area.

Adulticidal Bioassay: Sugar fed adult mosquitoes (4-6 days old) was used for bioassay. Different concentrations of the essential oils were impregnated on filter papers of 1cm² size. The bioassay was conducted in a cylindrical glass tube (15cm X 5cm) following the method of WHO (1981). The experiment was carried out in triplicate for each essential oil. For each replicate two tubes were used; one was used to expose the mosquitoes to the essential oil and another to hold the mosquitoes before and after the exposure period. Each tube was closed at one end with a wire mesh screen. Twenty sucrose fed mosquitoes were released in to the tube, and the mortality rate was observed every 15 min for 3h exposure. At the end of the exposure period, the mosquitoes were transferred in to the holding tube. A cotton pad soaked in 10% sugar solution was placed in the tube during the holding period. Mortality of mosquitoes recorded after 24h.

RESULTS AND DISCUSSION

Observations of the present study showed that the essential oils of *M. leucadendron* and *C. citrinus* have significant adulticidal and repellent activity against *Cx. quinquefasciatus* and *Ae. aegypti* mosquitoes. The present findings are comparable to earlier reports of Amer and Mehlhorn (2006), who reported forty one essential oils against different species of mosquitoes

Table 1. Evaluation of different concentrations of essential oils of *M. leucadendron* and *C. citrinus* against selected species of mosquitoes

Plants	Mosquito species	Conc. (%)	Percentage of repellency at different intervals after treatment					
			330 min	360 min	390 min	420 min	450 min	480 min
<i>M. leucadendron</i>	<i>Cx. quinquefasciatus</i>	100	100	100	100	100	100	100
		50	100	100	100	100	100	100
		10	100	100	100	100	96	84
		0	0	0	0	0	0	0
	<i>Ae. aegypti</i>	100	100	100	100	100	97	90
		50	100	100	100	96.5	88	80.9
		10	100	94	87	82	75	70
		0	0	0	0	0	0	0
<i>C. citrinus</i>	<i>Cx. quinquefasciatus</i>	100	100	100	100	100	100	97
		50	100	100	100	100	94.9	85.7
		10	100	100	94.2	88.7	83	79
		0	0	0	0	0	0	0
	<i>Ae. aegypti</i>	100	100	100	100	100	96	88.5
		50	100	100	94	88.2	81	75.7
		10	94.2	89	80	76	70.3	66
		0	0	0	0	0	0	0

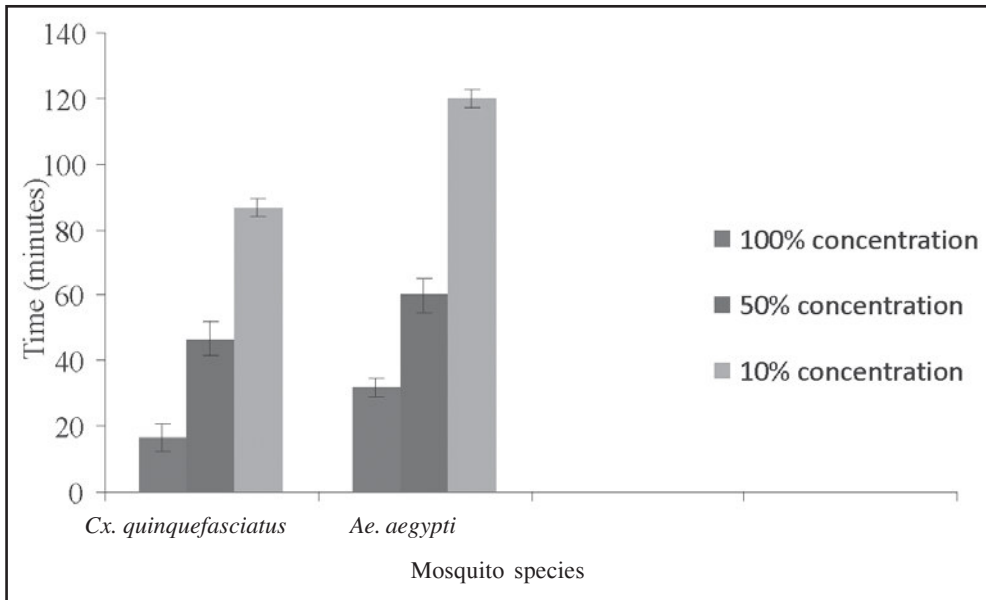


Figure 1. Time (minutes) taken for 100% adult mortality of mosquitoes tested with different concentration of *M. leucadendron* essential oil for an exposure time of 3 hours

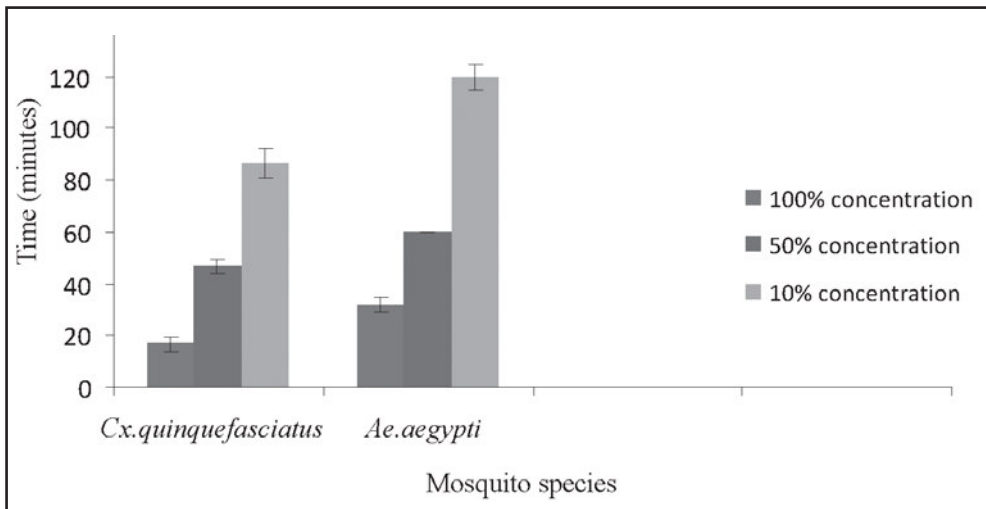


Figure 2. Time (minutes) taken for 100 % adult mortality of mosquitoes tested with different concentration of *C. citrinus* essential oil for an exposure time of 3 hours

and found out five most effective essential oils viz; *Litsea cubeba* (Lour.) Pers., *M. leucadendron*, *M. quinquenervia* (Cav.) S. T. Blake, *Viola odorata* Linn. and *Nepeta cataria* Linn., which induced 100% repellency over a protection period of 480 min against *Ae. aegypti*, *An. stephensi* and *Cx. quinquefasciatus*. Observations on the efficacy of essential oils, the repellent effect and adulticidal effect against *Cx. quinquefasciatus* and *Ae. aegypti* are provided in table 1.

Considering protection time and repellency, *M. leucadendron* was shown to have higher activity than that of *C. citrinus*. The percentage repellency showed a dose depended effect on the tested mosquitoes. In *Cx. quinquefasciatus*, the percentage repellency of *M. leucadendron* ranged between 84% - 100%. The 100% protection period lasted up to 480 min for 100% and 50% concentrations. At 10% concentrations the 100% protection period lasted up to 420 min. *M. leucadendron* demonstrated 420 min protection period against *Ae. aegypti*. In case of *C. citrinus* essential oil, the protection period for both *Cx. quinquefasciatus* and *Ae. aegypti* were 420 min. The percentage repellency for 480 min at 100%, 50% and 10% concentrations were 97%, 85.7% and 79% for *Cx. quinquefasciatus* and 88.5%, 75.7% and 66% for *Ae. aegypti* respectively.

Observations on adulticidal activity of *M. leucadendron* and *C. citrinus* were provided in figure 1 and 2. Among the two essential oils tested, the highest adulticidal activity was observed in *C. citrinus* against *Cx. quinquefasciatus*. At higher concentrations, the adults showed the restless movement for sometimes with abnormal wagging and died. The results show a dose depended effect on adult mortality. From these results, it is proved that both *M. leucadendron* and *C. citrinus* are active agents against mosquitoes. This study opens the possibility of further investigations of these plant products against other pest populations. Further studies are needed to develop appropriate formulations. The isolation and purification studies are in progress.

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