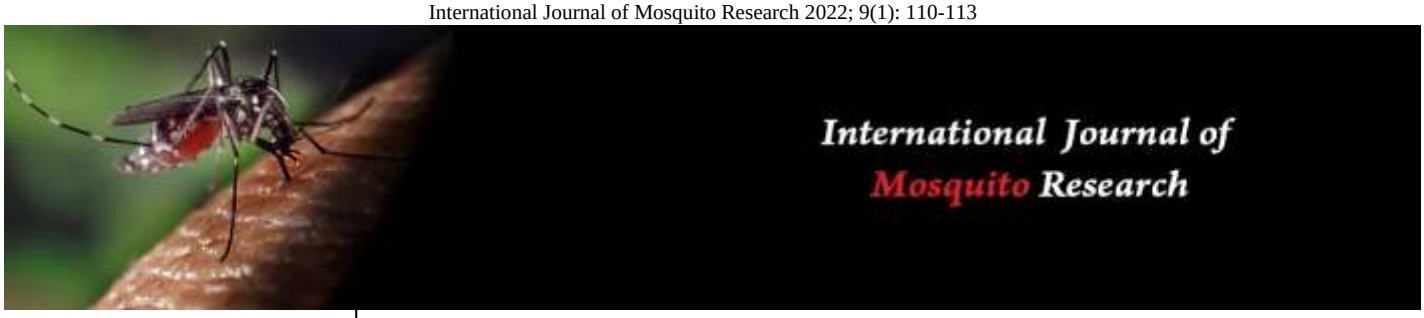

ISSN: **2348-5906**
CODEN: **IJMRK2**
IJMR 2022; 9(1): 110-113

www.dipterajournal.com
Received: 07-11-2021
Accepted: 10-12-2021

K Dass
Department of Zoology, Annai Vailankanni Arts and Science College, Thanjavur, Bharathidasan University, Tiruchirappalli, Tamilnadu, India

S. Sujitha
Department of Zoology, Annai Vailankanni Arts and Science College, Thanjavur, Bharathidasan University, Tiruchirappalli, Tamilnadu, India

P Mariappan
P.G and Research Department of Zoology, Rajah Serfoji Govt. College (Autonomous), Thanjavur. Bharathidasan University, Tiruchirappalli, Tamilnadu, India

Corresponding Author:
K Dass
Department of Zoology, AnnaiVailankanni Arts and Science College, Thanjavur, Bharathidasan University, Tiruchirappalli, Tamilnadu, India

Larvicidal activity of selected medicinal plants against dengue vector *Aedes aegypti*

K Dass, S Sujitha and P Mariappan

DOI: https://doi.org/10.22271/23487941.2022.v9.i1b.588

Abstract
The use of synthetic chemical pesticides to control mosquitoes has resulted in a slew of issues, including practical, ecological, insecticide susceptibility, financial, and toxicity to co-inhabitant organisms. Therefore, new ways for eradicating *Aedes aegypti* using medicinal plant active bio compounds. The purpose of this study was to assess the larvicidal effectiveness of *Aegle marmelos* and *Coleus aromaticus* plants methanolic extracts against *Ae. aegypti* larvae and pupa. The 24 hour LC_{50} values for II, III, IV instars, and pupa for methanolic solvent leaf extracts of *A. marmelos* are 124.27ppm, 145.07ppm, 178.87ppm, and 225.99ppm, respectively. The LC_{50} value for *C. aromaticus* plant extract was 62.46 ppm in the II instar, 81.94 ppm in the III instar, 101.19 ppm in the IV instar, and 124.34 ppm in the pupa. *A. marmelos* II, III, IV instars, and pupa have LC_{90} values of 222.74ppm, 283.43ppm, 354.02ppm, and 439.73ppm, respectively, after 24 hr. *C. aromaticus*, respectively, 162.87 ppm for the II instar and 202.83ppm for the III instar.213.63 for the IV instar and 254.14ppm in the pupa.

Keywords: Larvicidal activity, insecticidal, *Aegle marmelos*, *Coleus aromaticus*, *Aedes aegypti*, plant extracts

1. Introduction
Mosquitoes spread many terrible diseases than other group of insect vectors, and world wild millions of people affects every year [1, 2]. *Ae. aegypti* was a major vector for dengue fever, Zika fever, Chikungunya and yellow fever [3, 4].
To prevent and control the *Ae. aegypti* mosquitoes used by synthetic chemical insecticides created a number of problems and were not successful due to technical, environmental, insecticide resistance, economical and also toxic to co-inhabitant organisms [5-7].
Phytochemicals based mosquitocidal activity are now widely acknowledged as effective alternatives to synthetic insecticides in mosquito control strategies [8, 9]. In this time we need to discover alternate strategies to eradicate *Ae. aegypti* use of plant based nature products. In this study aimed to evaluate the larvicidal activity of two plant methanolic extracts tested against *Ae. aegypti* larval stages and pupa.
The experimental plants were chosen based on a survey of the literature on preliminary phytochemical, familial activity, and ethnopharmacological characteristics. *A.marmelos* has antidiarrhoeal, antimicrobial, antiviral, radioprotective, anticancer, chemopreventive, antipyretic, ulcer healing, antigenotoxic, diuretic, antifertility, and anti-inflammatory activities, according to many experimental and clinical investigations [10, 11].
C. aromaticus medicinal properties used in traditional, ayurvedic, and folklore medicine. This plant has been shown to have antioxidant, antiepileptic, antiurolithic, antidiabetic, anticancer, anthelmintic, antiprotozoal, and antiviral and antimycobacterial properties. The plant is useful against cardiovascular illnesses, respiratory disorders, and digestive diseases [12, 13]. The focus of this research was to see how efficient *A. marmelos* and *C. aromaticus* leaves methanolic crude extracts were as a larvicide against *Ae. Aegypti.*

2. Materials and methods
2.1 Extract collection and preparation
The leaves of *A. marmelos* (Linn) and *C. aromaticus* (*Benth*) were obtained in Pappanadu, Thanjavur, Tamil Nadu, India. The plant leaves were cleaned with distilled water and shade dried for 10-14 days at 27-37 °C [14]. 900 gms powder of *A. marmelos* and *C. aromaticus* put in Soxhlet extractor with methanol solvent.

The solvent was evaporated from the crude extract using a rotary evaporator, and the crude extract powder was kept at 4°C [15, 16]. A stock solution was made by combining 1 gram of crude leaf extract with 100 ml of acetone. Various concentrations of this stock were made and evaluated in a larvicidal experiment [17, 18].

2.2 Maintenance of experimental animal

The eggs of *Ae. aegypti* (*Lin*) were procured from the vector control research centre in Madurai, Tamil Nadu India. The eggs hatched into the first instar, which was kept in the research lab at ambient temperature (27±2 °C) and 75-85 percent humidity levels [19, 20]. The larvae were fed a 3:1 mixture of dog biscuit and yeast power [21]. The second, third, and fourth instars, as well as the pupa of *Ae. aegypti* were used in this experiment.

2.3 Larvicidal bio-assay

The larvicidal activity was assayed in accordance with WHO guidelines, with minor modifications [22]. Each 250ml jar contains 200 mL of dechlorinated water. The test concentrations of *A. marmelos* and *C. aromaticus* methanol leaf extract range from 100 to 300ppm. A separate control was also kept by adding 2ml of acetone to 200ml of dechlorinated water in a 250ml container. Ten larvae were used in each experiment. Dead larvae were removed as soon as possible to avoid decomposition. The two plant extracts were tested against a 24 hour larval stage and pupa mortality data. The corrected mortality rate was calculated using Abbott's formula.

2.4 Statistical analysis

Finney's [23] log probit analysis method was used to calculate larval mortality data. SPSS was used to compute chi-square values. A two-way ANOVA was used to determine the differences in death rates as a combination of plant species and larval stages.

3. Results

The larvicidal activity of methanolic solvent leaves extracts of *A. marmelos*, the 24 hr LC_{50} values for II, III, IV instars, and pupa are 124.27ppm, 145.07ppm, 178.87ppm, and 225.99ppm,. Accordingly the LC_{50} value for *C. aromaticus* plant extract was 62.46 ppm in the II instar, 81.94 ppm in the III instar, 101.19 ppm in the IV instar, and 124.34 ppm in the pupa Similarly, the 24 hr LC_{90} value for *A. marmelos* II, III, IV instars, and pupa is 222.74ppm, 283.43ppm, 354.02ppm, and 439.73ppm, respectively, after 24 hr. *C. aromaticus*, respectively, 162.87 ppm for the II instar and 202.83ppm for the III instar.213.63 for the IV instar ppm, 254.14ppm in the pupa given in the table 1.The ANOVA test was used to determine the connection between plant species and larval and pupal mortality (Table 2).

The present investigation exhibited dose dependent larval mortality; that is, as the concentration increased, larval mortality also increased. These plant based natural products are safe for the environment and co-inhabitant organisms.

Following a preliminary screening with crude leaf extracts, significant larvicidal activity against *Ae. aegypti* were found in this study.

Furthermore, more research is required to do crude extract fractionation, which will aid in the identification of active toxic compound(s) responsible for larval mortality.

4. Discussion

Several researchers have discussed the insecticides derived from various plant parts, the plant parts exist naturally play an important role in mosquito control. According to a study conducted by Kaushik and Saini [24] *Millingtonia hortensis* leaf extract is effective *against An. stepensi, Ae. aegypti,* and *Cx. quinquefasciatus* larva.

Vepris soyauxii (Rutacea) and *Momordica foetida*, *Gnidia glauca* aqueous and methanol extracts shown good biological effects against malaria vector species, *An. gambiae* and *An. coluzzii* [25].

According to Aziz experiments, the crude extract of *Vitex ovata* bioactive compounds could be produced as biolarvicides for *Ae.aegypti* [26].

The effect of two plant extracts (*Spilanthes acmella* and *Andrographis paniculata*) on distinct larval instars and pupae of the mosquito vector *Ae.aegypti* has been studied. *Spilanthes acmella* flower extract outperformed *Andrographis paniculata*. These plants with insecticidal capabilities appear to be more effective vector control agents than synthetic insecticides [27].

Clausena anisata acetone, dichloromethane, and hexane crude leaf extracts were tested against *Ae.aegypti.* The findings of this investigation suggest that *C. anisata* hexane extracts have the potential to be used in the control of mosquito populations [28].

The larvicidal and ovicidal activity of *Clausena excavata* (Rutaceae) extracts hexane, diethyl ether, dichloromethane, ethyl acetate, and methanol against *Ae.aegypti*, *An. stephensi* (Diptera: Culicidae), and *Cx. quinquefasciatus.* The study concludes that *C. excavata* may be a source of natural larvicidal and ovicidal activity against vector insects [29].

Citrus sinensis and *Murraya koenigii* leaves extracts were tested independently on *Ae.aegypti* and *Cx. quinquefasciatus* third instar larvae. This study validates and suggests that the usage of *C. sinensis* and *M. koenigii* is safe and environmentally friendly, and that it could be used as an alternative to synthetic pesticides in vector control [30].

The current study's findings indicate that using crude extract of *C. aromaticus* larvicide against *Ae. aegypti* as a potential larvicide and isolating bioactive components from these plants will aid in mosquitoes population management.

5. Conclusion

This finding contributes to the current search for new bio-active elements that can be used as alternatives to synthetic pesticides in plants. According to the findings of this study plant leaves have a high larvae potential and are safe for the environment. The methanol leaf extracts of *A. marmelos* and *C. aromaticus* can contribute immensely to reduction of *Ae.aegypti* population density. Furthermore, the harmful effects of specific plant chemicals on larvae must be investigated before they can be deemed environmentally benign. The death rate of the larvae increases as the concentration of the plant extract rises.

Table 1: The LC_{50} and LC_{90} values of *A. marmelos and C. aromaticus* leaves were determined against the II, III, IV instars, and pupa of *A. aegypti* after 24 hr of exposure.

Plant species	Larval stages	LC_{50} (ppm)(LCL-UCL)	LC_{90} (ppm)(LCL-UCL)	$\chi 2$
A. marmelos	II	124.47(80.57-152.87)	222.74(162.78-348.37)	.840
	III	145.07(107.59-174.92)	283.43(182.71-398.98)	2.011
	IV	178.87(148.19-250.43)	354.02(274.57-502.30)	1.364
	Pupa	225.99(191.55-277.88)	439.7 (330.66-543.30)	.765
C. aromaticus	II	62.46(48.46-90.95)	162.87(132.78-154.98)	1.388
	III	81.94(70.17-108.22)	202.83(148.93-363.81)	.355
	IV	101.19(88.68-131.79)	213.63(175.31-379.94)	2.260
	Pupa	124.34(101.13-192.19)	254.14(197.08-448.84)	1.440

Table 2: ANOVA was used to evaluate the reliability of the connection between mortality (LC_{50}) and plant extracts and larval stages.

Source of Variation	SS	df	MS	F	P-value
Plant species	7457	3	2485.66	15.438	0.02500
Larval stages	11552	1	11552	71.751	0.00345
Error	483	3	161		
Total	19492	7			

****P*< 0.001; ***P*< 0.001

6. References

1. Kaushik R, Saini P. Screening of some semi-arid region plants for larvicidal activity against *Aedes aegypti* mosquitoes. J Vector Borne Dis. 2009;46(3):244-246.
2. Kamaraj C, Bagavan A, Elango G, *et al*. Larvicidal activity of medicinal plant extracts against *Anopheles subpictus* and *Culex tritaeniorhynchus.* Indian J Med Res. 2011;134(7):101-106.
3. Elango G, AbduzZahir A, Bagavan A, *et al.* Efficacy of indigenous plant extracts on the malaria vector *Anopheles subpictus*grassi (Diptera: Culicidae). *Indian J Med Res*. 2011;134(9):375-383.
4. Kuppusamy C, Murugan K. Mosquitocidal effect of *Andographis paniculata* Nees against the malaria vector, *Anopheles stephensi* Liston (Diptera: culicidae). Int J Integr Biol. 2009;5(2):75-81.
5. Maneemegalai S, Kumar MS. Evaluation of Larvicidal Effect of *Lantana camara* Linn against Mosquito Species *Aedes aegypti* and *Culex quinquefasciatus*. Adv Biol Res (Rennes). 2008;2(4):39-43.
6. Subramaniam J, Murugan K, Kovendan K, Kumar PM. Larvicidal and pupcidal efficacy of *Momordica charantia* leaf extract and bacterial insecticide, *Bacillus thuringiensis* against malarial vector, *Anopheles stephensi* Liston. (Diptera: Culicidae). JBiopestic. 2012;163-169.
7. Das NG, Goswami D, Rabha B. Preliminary evaluation of mosquito larvicidal efficacy of plant extracts. J Vector *Borne Dis*. 2007;44(2):145-148.
8. Bagavan A, Kamaraj C, Elango G, AbduzZahir A, Abdul Rahuman A. Adulticidal and larvicidal efficacy of some medicinal plant extracts against tick, fluke and mosquitoes. *Vet Parasitol*. 2009;166(3-4):286-292. doi:10.1016/j.vetpar.2009.09.007
9. Rathy MC, Sajith U, Harilal CC. Plant diversity for mosquito control: a preliminary study. Int J Mosq Res. 2015;2(1):29-33.
10. Baliga MS, Bhat HP, Joseph N, Fazal F. Phytochemistry and medicinal uses of the bael fruit (*Aegle marmelos* Correa): A concise review. *Food Res Int*. 2011;44(7):1768-1775. doi:10.1016/j.foodres.2011.02.008
11. Rahman S, Parvin R. Therapeutic potential of *Aegle marmelos* (L.): An overview. *Asian Pacific J Trop Dis*. 2014;4(1):71-77. doi:10.1016/S2222-1808(14)60318-2
12. Rout OP, Acharya R, Mishra SK, Sahoo R. Pathorchur (*Coleus aromaticus*): a review of the medicinal evidence for its phytochemistry and pharmacology properties. International Journal of Applied Biology and Pharmaceutical Technology. 2012;3(4):348-55.
13. Jimmy JL. *Coleus aromaticus* Benth.: an update on its bioactive constituents and medicinal properties. *All Life*. 2021;14(1):756-773. doi:10.1080/26895293.2021.1968959
14. Patil CD, Borase HP, Salunkhe RB, *et al*. Mosquito larvicidal potential of Gossypium hirsutum (Bt cotton) Leaves extracts against *Aedes aegypti* and *Anopheles stephensi* larvae. *J Arthropod Borne Dis*. 2013;8(1):91-101.
15. Mathivanan T, Govindarajan M, Elumalai K, Krishnappa K, Ananthan A. Mosquito larvicidal and phytochemical properties of *Ervatamiacoronaria* Stapf. (Family: Apocynaceae). *J Vector Borne Dis*. 2010;47(3):178-180.
16. Ahmad B, Ali J. Evaluation of larvicidal activity of *Hippophae rhamnoides* L. leaves extracts on *Aedes aegypti* and *Anopheles stephensi* (Diptera: Culicidae). *Middle East J Sci Res*. 2013;13(5):703-709. doi:10.5829/idosi.mejsr.2013.13.5.2939
17. Kovendan K, Murugan K, Vincent S, Barnard DR. Mosquito larvicidal properties of *Orthosiphon thymiflorus* (Roth) Sleesen. (Family: Labiatae) against mosquito vectors, *Anopheles stephensi*, *Culex quinquefasciatus* and *Aedes aegypti* (Diptera: Culicidae). *Asian Pac J Trop Med*. 2012;5(4):299-305. doi:10.1016/S1995-7645(12)60043-1
18. Govindarajan M. Evaluation of *Andrographis paniculata* Burm.f. (Family: Acanthaceae) extracts against *Culex quinquefasciatus* (Say.) and *Aedes aegypti* (Linn.) (Diptera: Culicidae). *Asian Pac J Trop Med*. 2011;4(3):176-181. doi:10.1016/S1995-7645(11)60064-3
19. Amerasan D, Murugan K, Kovendan K, Mahesh Kumar P, Panneerselvam C, Subramaniam J, *et al*. Adulticidal and repellent properties of *Cassia tora* Linn. (Family: Caesalpiniaceae) against *Culex quinquefasciatus, Aedes aegypti*, and *Anopheles stephensi*. Parasitology research. 2012 Nov;111(5):1953-64.
20. Promsiri S, Naksathit A, Kruatrachue M, Thavara U. Evaluations of larvicidal activity of medicinal plant extracts to *Aedes aegypti* (Diptera: Culicidae) and other effects on a non target fish. *Insect Sci*. 2006;13(3):179-188. doi:10.1111/j.1744-7917.2006.00080.x
21. Arivoli S, John Ravindran K, Tennyson S. Larvicidal efficacy of plant extracts against the malarial vector *Anopheles stephensi* Liston (Diptera: Culicidae). *World J*

Med Sci. 2012;7(2):77-80. doi:10.5829/idosi.wjms.2012.7.2.62134
22. WHOPES. Guidelines for laboratory and field testing of mosquito larvicides. *World Heal Organ*. Published online, 2005, 1-41.
23. Finney DJ. Probit Analysis Probit Analysis. *Cambride Univ Press*. Published online, 1952, 330.
24. Kaushik R, Saini P. Larvicidal activity of leaf extract of *Millingtoni ahortensis* (Family: Bignoniaceae) against *Anopheles stephensi*,*Culex quinquefasciatus* and *Aedes aegypti*. *J Vector Borne Dis*. 2008;45(1):66-69.
25. Njuabe MT, Fru CT, Desire S, Awono-Ambene HP. Larvicidal Activity of *Momordica foetida* (Cucurbitaceae), *Gnidia glauca* (Thymelaeaceae) and *Vepris soyauxii* (Rutaceae) Extracts on *Anopheles gambiae* Mosquitoes and their Acute Toxicity on Rats. *J Environ Sci Public Heal*. 2021;5(2):358-370. doi:10.26502/jesph.96120153.
26. Aziz M, HashanArif EI, Muhammad Dimyati NI, *et al.* Larvicidal Effect of *Vitex ovata* Thunb. (Lamiales: Lamiaceae) Leaf Extract towards *Aedes* (Stegomyia) *aegypti* (Linnaeus, 1762) (Diptera: Culicidae). *Parasitologia*. 2021;1(4):210-217. doi:10.3390/parasitologia1040022.
27. Remia KM. Larvicidal and pupicidal effect of *Spilanthes acmella* and *Andrographis paniculata* on the mosquito *Aedes aegypti*. International Journal of Institutional Pharmacy and Life Sciences. 2012;2(2):71-6.
28. Mukandiwa L, Eloff JN, Naidoo V. Larvicidal activity of leaf extracts and seselin from *Clausena anisata* (Rutaceae) against *Aedes aegypti*. *South African J Bot*. 2015;100:169-173. doi:10.1016/j.sajb.2015.05.016
29. Thangarasu M, Kaliyamoorthy K, Kuppusamy E. Pesticidal and Mosquitocidal Activities of *Clausena excavata* Burm. F. (Rutaceae) against *Spodoptera litura* (Fab.) (Noctuidae: Lepidoptera) and *Aedes aegypti* L., *Anopheles stephensi* Liston, and *Culex quinquefasciatus* Say. (Diptera: Culicidae) *Adv Zool*. 2014;2014:1-7. doi:10.1155/2014/745015
30. Marin G, Arivoli S, Tennyson S. Larvicidal activity of two rutaceae species against the vectors of dengue and filarial fever. *J Exp Biol Agric Sci*. 2020;8(2):166-175. doi:10.18006/2020.8(2).166.175