Biotechnology Frontiers
An International Journal
OPEN ACCESS
ISSN 3117-5791
Original Research Article
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Phytochemical composition and Larvicidal properties of ethanolic extract from *Laurencia papillosa* (C. Agardh) Greville (Rhodophyceae), against *Aedes aegypti,Anopheles stephensi* and *Culex quinquefasciatus* (Diptera: Culicidae)

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Citation: Gnana Suriya P, Nagarajan R, Adaikala Raj G (2025). Phytochemical composition and Larvicidal properties of ethanolic extract from *Laurencia papillosa* (C. Agardh) Greville (Rhodophyceae), against *Aedes aegypti,Anopheles stephensi* and *Culex quinquefasciatus* (Diptera: Culicidae). *Biotechnology Frontiers: An International Journal.* **DOI: https://doi.org/10.51470/BF.2025.5.2.29**

10 August 2025: Received | 15 September 2025: Revised | 14 October 2025: Accepted | 10 November 2025: Available Online

Abstract

The research aim to assess the larvicidal efficacy of its ethanolic extracts against medically significant mosquito larvae and determine the presence of secondary metabolites. The algal material was extracted using ethanol in a Soxhlet apparatus for a duration of 48 hours. The extract was mixed with 1 mL of acetone and diluted to achieve concentrations ranging from 12.5 to 400 ppm. Bioassays for larvicidal activity were performed on early fourth instar larvae of Aedes aegypti, Anopheles stephensi, and Culex quinquefasciatus, including five replicates for each concentration. Mortality was noted after 12 and 24 hours. The GC–MS analysis was conducted with an Agilent system featuring an HP-5MS column, and compounds were recognized utilizing the NIST mass spectral library. FT–IR spectra (4000–400 cm^{-1}) identified functional groups through the KBr pellet technique. Probit analysis was utilized to ascertain LC_{50} and LC_{90} values, including their 95% confidence intervals. Phytochemical analysis of the ethanolic extract of L. papillosa showed the presence of small amounts of alkaloids, cardiac glycosides, and saponins, while tannins, steroids, and phenolics were found in moderate quantities; flavonoids, terpenoids, and coumarins were not detected. Of the species tested, A. stephensi exhibited the greatest susceptibility with LC_{50} = 174.6 ppm and LC_{90} = 519.5 ppm at 12 hours, and LC_{50} = 110.6 ppm and LC_{90} = 391.8 ppm at 24 hours. A. aegypti showed the least sensitivity. GC–MS analysis revealed key compounds such as 3,7,11,17-tetramethyl-2-hexadecen-1-ol, Decane (2,2,3-trimethyl), and sulfur-bearing derivatives, suggesting a variety of hydrocarbons and terpenoids. FT-IR analysis verified the existence of hydroxyl, carbonyl, aromatic, and halogen groups, reinforcing the presence of phenolics and halogenated compounds that contribute to the extracts notable biological and environmentally friendly larvicidal capability. The findings indicate that the ethanolic extract of L. papillosa has strong bioactive constituents with larvicidal effects, reinforcing its promise as an environmentally friendly biological control agent for mosquito vectors.

Keywords: *Laurencia papillosa, Larvicidal activity, Phytochemical screening, Mosquito control.*

Introduction

Globalization, climate change, and increased human movement have greatly facilitated the worldwide spread of highly invasive species [1]. Among these, arthropods are particularly important as they serve as carriers for numerous pathogens that cause serious diseases, potentially leading to epidemics or pandemics [2]. Mosquitoes (Diptera: Culicidae) are especially concerning because they are vectors for a variety of dangerous pathogens and parasites [3]. The mosquito genera *Anopheles, Aedes*, and *Culex* are particularly noteworthy, as they are responsible for transmitting major diseases such as malaria, dengue, yellow fever, filariasis, Japanese encephalitis, and Zika virus infection [4]. To combat mosquito-borne diseases, a range of strategies has been implemented, including behavioral, chemical, biological, and mechanical methods. While these approaches have had varying levels of success, their effectiveness is often limited by the absence of efficient vaccines and the slow development of antiviral drugs for most arboviruses. Additionally, the rapid development of insecticide resistance in mosquito populations has severely compromised existing vector control programs and hindered the creation of new chemical control agents. The shortcomings of traditional vector control methods, combined with the spread of invasive mosquito species and increased human-vector interactions, have led to the persistent re-emergence of arboviral diseases [5]. As a result, mosquito control programs are facing rapidly changing challenges, highlighting the urgent need for innovative strategies in disease surveillance and vector management as a critical public health priority.

Vector-borne diseases continue to be a significant global health challenge, leading to nearly one million deaths each year. Malaria is responsible for about 400,000 of these deaths, with an estimated 219 million cases occurring worldwide. Dengue fever, which is mainly spread by Aedes mosquitoes, impacts nearly four billion people globally and results in around 40,000 deaths annually [6]. Although several commercial insecticides are available, the emergence of resistance in mosquito vectors significantly hinders effective disease control [7]. Globally, numerous plant species are being investigated for their bioefficacy against vector-borne diseases, including their larvicidal and adulticidal activities [8]. The use of plants and herbal remedies for the treatment of human ailments has been practiced in the region since ancient times. Mosquito-borne diseases such as dengue fever, Zika virus infection and chikungunya continue to pose serious threats to public health, especially in tropical and subtropical regions. It has been projected that nearly half of the world's population may be at risk of arboviral infections by the year 2050 [9]. Covering approximately 70% of the Earth's surface, the oceans represent the largest reservoir of bioactive compounds and offer immense potential for the discovery of novel pharmaceutical agents [10]. Several marine-derived anticancer compounds have already been approved for clinical use or are currently undergoing clinical trials [11].

Seaweeds are a significant natural asset in the oceanic ecosystem, having been used for a long time as functional foods and in traditional medicinal practices [12]. Numerous studies have shown that marine algae are abundant in structurally distinct natural compounds with a variety of biological activities, making them promising candidates in the quest for effective and specific therapeutic agents [13]. The anticancer properties of marine algae have mainly been investigated through the use of crude extracts or partially purified fractions [14]. Over the last fifty years, the genus *Laurencia* has been thoroughly studied, with many new metabolites still being discovered [15]. These metabolites encompass sesquiterpenes, C15 acetogenins, diterpenes, triterpenes, indoles, steroids, aromatic compounds, and other miscellaneous metabolites, many of which are naturally halogenated [16]. Several of these compounds have shown a broad spectrum of biological activities, including cytotoxic, antibacterial, antifungal, antiviral, antiparasitic, and anti-inflammatory effects. *L. papillosa* is commonly found along the rocky shores of Lebanon. Various studies have explored this species from different regions, such as Japan, the Red Sea, the Caribbean Sea, and India, resulting in the isolation and identification of numerous bioactive metabolites [17]. Recent research has indicated that crude ethanol–water (50:50, v/v) and ethanol–chloroform (50:50, v/v) extracts of *L. papillosa* collected from the Lebanese coast showed moderate cytotoxic activity against the Jurkat human acute T-cell leukemia cell line, with IC_{50} values of 121.6 µg/mL and 57.7 µg/mL, respectively [18].

In recent times, there has been a growing interest in discovering natural products that could be used in vector control. Plants are known to produce a vast range of bioactive compounds that are effective against insects that feed on plants and plant diseases. Utilizing plant-derived compounds for vector control is increasingly seen as an eco-friendly method, as these substances typically have minimal negative impacts on non-target organisms and the environment. Additionally, plant-based bioactive compounds are structurally varied and often have unique mechanisms of action, making them promising candidates for creating new larvicidal agents.

Materials and Methods

Collection of algae samples

Rhodomenia palmata (Rhodophyceae) and *Sargassum swartzii* (Phaeophyceae) seaweeds were gathered by hand from the underwater marine rocks at Manappad, (Lat. 8°30′N; Long. 78°8′E), Tuticorin district, the Marine biosphere of Gulf of Mannar, Tamil Nadu, India. Collections of seaweeds were conducted in December 2024. The specimens of the museum are housed in the Department of Botany, Annamalai University, Annamalainagar.

2.2. Preparation for extraction

The gathered algal samples were meticulously washed to eliminate epiphytes, attached debris, and foreign substances, and any dead sections were carefully removed. The samples were first rinsed with seawater, then washed with freshwater to remove surface contaminants. Afterward, the seaweeds were brought to the lab in sterile polyethylene bags while maintaining chilled conditions using an ice box filled with slush ice. When reaching the laboratory, the samples were washed with sterile distilled water to eliminate residual salts and impurities. The processed samples were air-dried at ambient temperature, chopped into small bits, and ground into a fine powder with a mixer grinder. Five hundred grams of the powdered algal material underwent Soxhlet extraction with ethanol as the solvent for six hours. The extracts collected were combined, and the solvent was eliminated under reduced pressure with a rotary evaporator (Heidolph, Germany) at 40 °C. The dried extracts were kept at 4 °C in a refrigerator until they were needed for further experimental analysis.

Phytochemical analysis

The ethanolic extracts of Laurencia papillosa underwent qualitative phytochemical analysis to detect the presence of significant secondary metabolites. The extracts were assessed for terpenoids, tannins, cardiac glycosides, saponins, alkaloids, flavonoids, phenolic compounds, and coumarins following standard qualitative methods outlined in the literature [19,20].

Mosquito Larvicidal Bioassessment

The *A. aegypti* and *A. stephensi* eggs were sourced from the Field Station at the Centre for Research in Medical Entomology (CRME), Indian Council of Medical Research (ICMR), located in Madurai, India.

Egg rafts of *C. quinquefasciatus* were gathered from drainage water in a nearby residential area of Karumathur (11°23′17″ N, 79°42′57″ E) and then raised in laboratory conditions (29 ± 3 °C and 75–85% relative humidity). Larvae were kept on a diet consisting of Brewer's yeast and dog biscuit in a 1:3 proportion. The ethanolic extracts of *L. papillosa* were assessed for larvicidal activity against *A. aegypti, A. stephensi* and *C. quinquefasciatus* in accordance with the standard protocols outlined by the World Health Organization (WHO) [21]. The extract was first dissolved in 1 mL of acetone and then diluted with distilled water to achieve test concentrations of 12.5, 25, 50, 100, 200, and 400 ppm. In each assay, twenty early fourth instar larvae were placed into a 100 mL beaker with the test solution, and five replicates were kept for every concentration. During the experimental period, the larvae were not given any food. Larval death rates were noted after 12 and 24 hours of exposure. Larvae that did not move or could not reach the water's surface when lightly nudged were deemed dead. Control experiments were carried out using distilled water and DMSO individually.

Chemical Analysis

Gas chromatographic assessment of the ethanolic extract of *L. papillosa* was conducted using a Thermo GC-Trace Ultra Version 5.0 system partnered with a Thermo DSQ II mass spectrometer. The device was fitted with a DB-5 MS typical non-polar capillary column. The injector temperature was held constant at 300 °C. The oven temperature protocol started at 80 °C, rose to 200 °C at a speed of 5 °C/min, and was sustained for 10 minutes. Then, the temperature was increased to 300 °C at a pace of 20 °C/min and maintained for 5 minutes. Helium served as the carrier gas with a steady flow rate of 1.0 mL/min. Samples were introduced in split mode utilizing a split ratio of 1:100. The relative percentage composition of the compounds in the ethanolic extract was determined using GC peak area normalization.

GC-MS Analysis

GC–MS analysis of the ethanolic extract from *L. papillosa* was performed using a Varian 3800 gas chromatograph linked to a Varian 1200 C single quadrupole mass spectrometer. The conditions for chromatography and the specifications of the column were the same as those utilized for GC analysis. The mass spectrometer functioned in electron impact (EI) mode with an ionization energy of 70 eV. The temperatures of the ion source and transfer line were held at 300 °C. Mass spectra were collected in centroid scan mode across a mass range of 40–800 amu. The identification of chemical constituents was accomplished by comparing the acquired mass spectra with those in the NIST and WILEY mass spectral libraries included with the instrument's software. The compounds identified were further validated through comparison with literature data that has been published.

Fourier Transform Infrared (FT-IR)

The FT-IR spectrum of the ethanolic extract of *L. papillosa* was obtained with an AVATAR-330 FT-IR spectrophotometer (Thermo Nicolet, USA). The sample was thoroughly blended with spectroscopic-grade potassium bromide (KBr), and pellets were formed utilizing the KBr pellet technique. The spectra were obtained to determine the functional groups in the bioactive components of the extract.

Statistical analysis

All experimental outcomes are presented as mean ± standard deviation (SD). Statistical analyses were conducted utilizing SPSS statistical software version 22.0 (SPSS Inc., Chicago, IL, USA). The Student's t-test was utilized to assess significant differences amongethanolic extracts in the larvicidal tests. One-way analysis of variance (ANOVA) was utilized to compare several treatment groups, with Duncan's multiple range test applied afterwards to identify differences in group means. A p-value under 0.05 was regarded as statistically significant.

Results

The phytochemical examination of the ethanolic extract from *L. papillosa* revealed the existence of various significant bioactive compounds. Alkaloids, cardiac glycosides, and saponins were identified in minimal quantities (+), while tannins, steroids, and phenolic compounds were observed in moderate levels (++), highlighting their crucial role in the biological activity of the extracts. Flavonoids, terpenoids, and coumarins were not present (–), and the findings are shown in Table 1. The dominance of phenolic compounds, tannins, and steroids indicates that these metabolites might be crucial for larvicidal effects, reinforcing their possible application in medical and environmentally sustainable vector management strategies.

Table 1: Phytochemical analysis of ethanolic extracts of L. papillosa

S.No	Secondary metabolites	*Laurencia papillosa*
1	Alkaloids	+
2	Flavonoids	-
3	Tannins	++
4	Cardic glycosides	+
5	Steroids	++
6	Terpenoids	-
7	Phenolic compounds	++
8	Saponins	+
9	Coumarins	-

- = Absence, + = weak, ++ = medium, +++ = strong

The ethanolic extract of L. papillosa showed significant larvicidal effects against *A. aegypti, A. stephensi,* and *C. quinquefasciatus* in a manner that depended on both dose and time. Of the species tested, *A. stephensi* exhibited the highest susceptibility, with LC_{50} and LC_{90} values of 110.6 and 391.8 ppm following a 24-hour period. *A. aegypti* showed moderate sensitivity with an LC_{50} of 150.1 ppm and an LC_{90} of 491.6 ppm, while *C. quinquefasciatus* was the least impacted, recording an LC_{50} of 216.6 ppm and an LC_{90} of 514.1 ppm after 24 hours, with the findings displayed in Table 2. Extended exposure duration raised larval mortality for all species.

The increased mortality rate observed in *A. stephensi* suggests its heightened vulnerability to algal bioactive compounds, likely resulting from variations in cuticular permeability and metabolic processes. These results indicate that the ethanolic extract of L. papillosa has significant larvicidal potential, backing its application as a sustainable option for managing vector mosquitoes. The ethanolic extract of L. papillosa was analyzed using GC–MS, which identified several bioactive components, highlighting five primary compounds based on their relative abundance. The predominant compound was observed at a retention time of 5.04 minutes with a composition of 17.98%, relating to a complex hydrocarbon derivative (partly identified as 17-(1,5-dimethylhexyl)-10,13-dimethyl compound). The second most prevalent compound was 3,7,11,17-tetramethyl-2-hexadecen-1-ol (17.13%), a long-chain terpenoid alcohol recognized for its biological functions. 2,2,3-trimethyl decane (16.40%) was also found in significant amounts, indicating it is a branched aliphatic hydrocarbon. A significant component, Di-n-decyl sulfone or 1,2-bis(trimethylsilyl)benzene (14.21%), emerged as an important sulfur-containing or aromatic compound, with the findings detailed in Table 3 and fig.1

Table 2: Larvicidal properties of ethanolic extract from Laurencia papillosaagainst the larvae of A. aegyptiA. stephensi, and C. quinquefasciatus after 12 and 24 h of exposure period

Name of the mosquito species Time Concentration (ppm) % mortality ± SE LC_{50} (LCL- UCL)[a] LC_{90} (LCL- UCL)[a] x2(d=4)[b]						
A .aegypti	After 12h	12.5	10 ± 0.44	253.2 (146.5 – 723.5)	647.7 (410.4 – 2514.0)	21.3
		25	18 ± 0.83			
		50	30 ± 0.85			
		100	43 ± 0.57			
		200	50 ± 0.50			
		400	62 ± 1.50			
	After 24h	12.5	18 ± 0.76	150.1 (59.7 – 275.9)	491.6 (335.1 – 1144.8)	16.5
		25	28 ± 0.83			
		50	42 ± 0.37			
		100	53 ± 0.57			
		200	62 ± 0.28			
		400	78 ± 0.50	174.6 (101.6 – 289.8)	519.5 (367.9 – 1021.4)	12.86
A. stephensi	After 12h	12.5	17 ± 0.28			
		25	26 ± 0.76			
		50	37 ± 0.57			
		100	48 ± 0.28			
		200	60 ± 0.34			
		400	75 ± 0.50			
	After 24h	12.5	24 ± 0.50	110.6 (86.7 – 134.5)	391.8 (340.1 – 467.0)	8.37
		25	31 ± 0.28			
		50	46 ± 1.52			
		100	58 ± 0.28			
		200	72 ± 0.40			
		400	88 ± 0.76			
C.quinquefasciatus	After 12h	12.5	5.0 ±1.04	309.7 (207.4 – 680.5)	662.8 (443.6 – 1756.9)	18.8
		25	11 ± 0.34			
		50	20 ± 0.57			
		100	31 ± 0.78			
		200	44 ± 0.50			
		400	56 ± 0.28			
	After 24h	12.5	7.0± 0.28	216.6 (130.9 – 414.7)	514.1 (315.7 – 1212.6)	22.69
		25	16 ± 0.57			
		50	29 ± 0.60			
		100	41 ± 0.28			
		200	56 ± 0.36			
		400	72 ± 0.50			

LCL lower confidence level, UCL upper confidence level
a 95 % Confidence interval
b Degrees of freedom

The fifth dominant compound, bicyclo[3.2.1]oct-3-en-2-one derivative (12.87%), represents a complex oxygenated bicyclic structure. These major chemical constituents indicate that the ethanolic extract of *L. papillosa* contains a diverse mixture of hydrocarbons, terpenoids, and oxygenated aromatic compounds that may contribute to its potent biological and larvicidal properties.

Table 2: Chemical Composition of ethanolic extract fromLaurencia papillosa

Peak No.	Retention Time (min)	Chemical Constituents[a,b]	Composition (%)
1.	4.74	Decane2,2,3-trimethyl	16.40
2.	4.79	3,7,11,17-Tetramethyl-2- hexadecen-1-ol	17.13
3.	5.04	17 1,5-Dimethylhexyl)-10,13- dimethyl-2,3,4,	17.98
4.	5.18	Methyl hexa-decanoate	5.34
5.	5.46	N-(5-chloro-2-hydroxyphenyl)dodecanamide	3.40
6.	5.61	Pentadecanoic acid	4.37
7.	5.78	(E, E)- Methyl 9, 11-Octa-decadienoate	3.64
8.	6.05	Cis-13-Eicosenoic acid	4.61
9.	6.46	Di-n-decylsulfone1, 2-Bis (trimethylsilyl) benzene	14.26
10	6.64	Bicyclo[3.2.1]oct-3-en-2-one, 3,8-dihydroxy-1-methoxy-7- (7-methoxy-1,3-benzodioxol-5-yl)-6-methyl-5	12.87
Total			100

[A]Compounds listed in order of elution from DB 35-MS Capillary Standard non-polar column.
[b]Components identified based on computer matching of the mass peaks with the WILEY and NIST Library

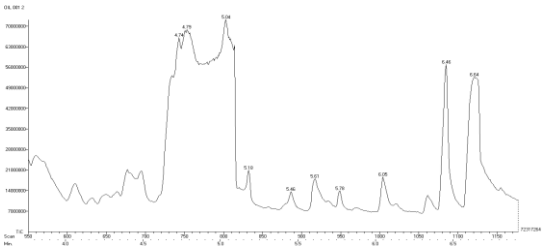

Fig 1: GC-MS analysis of ethanolic extract from Laurencia papillosa

The FT-IR spectrum of the ethanolic extract from L. papillosa displayed multiple distinct absorption bands linked to various functional groups. The wide and intense band seen at 3405.71 cm^{-1} relates to O–H stretching vibrations, suggesting the existence of hydroxyl or phenolic groups. The absorption peaks at 2919.18 and 2849.97 cm^{-1} indicate C–H stretching vibrations of aliphatic $-CH_2$ and $-CH_3$ groups, affirming the existence of alkane or fatty acid elements. The pronounced peak at 1707.22 cm^{-1} is linked to the C=O stretching of carbonyl groups, including ketones and aldehydes. The peaks observed at 1610.49, 1541.59, and 1511.99 cm^{-1} result from C=C stretching vibrations in aromatic compounds. The band at 1103.82 cm^{-1} is related to the C–O stretching found in alcohols and esters. The band at 430.89 cm^{-1} in the low-frequency range is linked to C–Br stretching, indicating the existence of halogenated compounds, often found in Laurencia species, with results shown in fig.2. These functional groups verify the existence of various bioactive compounds like terpenoids, phenolics and halogenated metabolites in the extract.

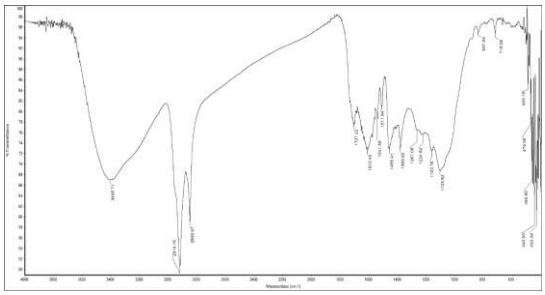

Fig 2: FT-IR spectrum analysis of ethanolic extracts of Laurencia papillosa

The phytochemical analysis of the ethanolic extract from*L. papillosa* showed the presence of various important bioactive compounds. Alkaloids, cardiac glycosides, and saponins were found in low amounts (+), while tannins, steroids, and phenolic compounds were identified at moderate levels (++), indicating their significant contribution to the extract's biological activities. These results align with earlier research. Manilal et al. [22] state that different species of Laurencia are recognized for generating terpenoids and alkaloids that possess antimicrobial and cytotoxic effects. Likewise, El Gamal [23] noted that red algae, such as Laurencia spp., are abundant producers of halogenated secondary metabolites, especially terpenoids and phenolics. The lack of flavonoids observed in this study was similarly reported in certain marine algal screenings by Kumaran et al. [24], indicating variation specific to species. Additionally, Johnson et al. [25] noted the occurrence of tannins and cardiac glycosides in *Laurencia* spp., corroborating the present results. The lack of flavonoids indicates metabolic differences that are specific to each species. The larvicidal properties of Laurencia papillosa are attributed to its content of terpenoids, tannins, steroids, cardiac glycosides, alkaloids, and phenolic compounds. Terpenoids are known to interfere with the nervous systems of larvae, resulting in paralysis and mortality [26]. Tannins function by attaching to digestive enzymes and proteins, disrupting larval nutrition [27]. Steroids may disrupt hormonal control, influencing metamorphosis [28]. Cardiac glycosides interfere with ion exchange processes, resulting in cellular toxicity [29]. Alkaloids have neurotoxic effects, while phenolic compounds cause oxidative stress, leading to larval death [30]

The ethanolic extract of *L. papillosa* displayed significant larvicidal effects against *A. aegypti, A. stephensi,* and *C. quinquefasciatus* in a manner dependent on both dose and time. Of the species examined, *A. stephensi* exhibited the highest susceptibility, demonstrating LC_{50} and LC_{90} values of 110.6 and 391.8 ppm following a 24-hour exposure. *A. aegypti* showed moderate sensitivity, having an LC_{50} of 150.1 ppm and an LC_{90} of 491.6 ppm, while *C. quinquefasciatus* was the least impacted, with an LC_{50} of 216.6 ppm and an LC_{90} of 514.1 ppm after 24 hours. The current research demonstrated that ethanolic extracts from marine macroalgae show considerable larvicidal properties against *A. aegypti, Anopheles stephensi,* and *C. quinquefasciatus,* with the most pronounced impact observed on *A. stephensi* (LC_{50} = 148.8 ppm and LC_{90} = 252.1 ppm after 24 hours). These results correspond with previous studies emphasizing the capability of marine algae for mosquito management. Manilal et al. [22] discovered that red algae, especially Laurencia spp., have larvicidal effects attributed to their bioactive terpenoids and alkaloids. Govindarajan et al. [26] also showed significant larvicidal effects of seaweed extracts on mosquito vectors. In a similar results, Elumalai et al. [31] demonstrated that marine macroalgae can act as environmentally friendly larvicides because of their secondary metabolites.

Extracts of *Sargassum* species, both ethanol and methanol, have demonstrated strong larvicidal effects on *A. stephensi* and *A. gambiae*. Research shows that death rates surpass 80% at comparatively low levels of extract. Green seaweeds such as Ulva have demonstrated considerable larvicidal properties against Anopheles species, mainly because of their elevated levels of fatty acids and polysaccharides. Extracts from seaweed have shown strong larvicidal effects on Anopheles mosquitoes, the main carriers of malaria. Research has indicated that extracts from species like *Ulva, Sargassum*, and *Gracilaria* demonstrate considerable mortality rates against Anopheles larvae [32].

Extracts from Padina, a brown seaweed, have shown significant effectiveness against *Culex quinquefasciatus,* with mortality rates reaching as high as 90% at specific concentrations. The extracts have been recognized to contain phenolic compounds as the main larvicidal agents.

Gracilaria species. I'm sorry, but it seems you've provided only a reference "[33]." Could you please provide the specific text you would like paraphrased? Red algae like Gracilaria have demonstrated encouraging effects on *Culex* larvae, with polysaccharides significantly contributing to their larvicidal efficacy. The effectiveness of seaweed extracts as larvicides against *Culex* mosquitoes has been thoroughly researched. Studies have shown that different species of seaweed have considerable larvicidal effects on Culex larvae [34]. For example, extracts from *G. corticata, S. wightii, Ulva lactuca,* and *Turbinaria conoides* have demonstrated encouraging outcomes. Extract of *G. corticata,* at a concentration of 250 ppm, resulted in 100% mortality of larvae from *C. quinquefasciatus.* In the same way, *Sargassum wightii* extract exhibited 93.33% larval mortality at a concentration of 500 ppm. *Ulva lactuca* extract showed 90.91% mortality in larvae at a concentration of 300 ppm, whereas *Turbinaria conoides* extract had 85.71% larval mortality at a concentration of 400 ppm [33]. The larvicidal activity of seaweed extracts is attributed to the bioactive compounds they contain, including alkaloids, flavonoids, and terpenoids [35]. These substances hinder larval growth, disturb mosquito neurotransmission, and cause oxidative stress, ultimately resulting in larval death.

The benefits of utilizing seaweed extracts as larvicides consist of their eco-friendliness, biodegradability, and minimal toxicity to non-target species. Nonetheless, the differences in effectiveness based on seaweed species, extraction techniques, and concentrations, along with the requirement for additional research on stability, formulation, and real-world application, represent limitations that must be tackled [36]. To maximize the potential of seaweed extracts as larvicides, upcoming studies should concentrate on standardizing methods for extraction and formulation, exploring synergistic interactions with other larvicides, and performing field trials to evaluate effectiveness and practicality.

β-Sitosterol extracted from *Abutilon indicum* has been previously shown to demonstrate notable larvicidal efficacy against *A. aegypti* larvae, with an LC_{50} value of 11.49 ppm [37]. The findings of this study align with previous research indicating that the effectiveness of larvicides against mosquitoes is closely linked to certain chemical properties, especially lipophilicity [38]. Compounds with lipophilic characteristics are recognized to more effectively penetrate the larval cuticle, thus amplifying their toxic impact. In this context, long-chain aliphatic fatty acids derived from the green seaweed *Cladophora glomerata* have demonstrated significant larvicidal effects against *Aedes triseriatus*, with LC_{50} values between 3 and 14 ppm. Particularly, saturated fatty acids like capric acid, lauric acid, and myristic acid, along with monounsaturated fatty acids such as palmitoleic acid, extracted from *C. glomerata*, exhibited significant larvicidal activity within the identical concentration spectrum [39]. Likewise, octacosane derived from *Moschoma polystachyum* was noted to demonstrate larvicidal effects on early third instar larvae of *C. quinquefasciatus*, with an LC_{50} value of 7.2 ppm.

The FT-IR spectral analysis of the ethanolic extract of *L. papillosa* in this study revealed the presence of various significant functional groups, such as hydroxyl, carbonyl, aromatic, and aliphatic components, highlighting the intricate biochemical makeup of the alga. The notable O–H stretching band detected at 3405.71 cm^{-1} indicates the existence of hydroxyl or phenolic compounds, which are often linked to antioxidant and antimicrobial properties. Moreover, the C–H stretching vibrations detected at 2919.18 and 2849.97 cm^{-1} relate to aliphatic chains, suggesting the existence of fatty acids and hydrocarbons typical of marine red algae, which may play a role in the noted larvicidal activity.

Dhinakaran et al. [40] revealed through FT-IR spectral analysis that the red alga *Hypnea musciformis* contains a range of bioactive compounds such as polysaccharides, terpenes, alkenes, sterols, and associated metabolites. Red algae are especially abundant in polysaccharides, where galactans make up the primary carbohydrate constituents. These galactans consist of agar, carrageenan, floridean starch, and xylan, which have important structural and functional functions in the cell walls of red algae [41]. Galactose acts as the main monosaccharide component in red algae and is the essential constituent of galactans like agar and carrageenan. FT-IR spectral analysis of the red algae *Kappaphycus alvarezii* showed the existence of aliphatic components, such as carbon compounds, ketones, alkyl halides, and hydroxyl functional groups, suggesting a complicated biochemical profile [42]. Likewise, FT-IR analysis of the methanolic extract from *Acanthophora specifera* validated the existence of various functional groups, including phenols, alkanes, alkenes, carboxylic acids, aromatic compounds, nitro compounds, alcohols, benzene derivatives, and bromoalkanes [43]. Furthermore, Pereira et al. [44] indicated that *L.obtusa* has sulfated galactans and agar-like polysaccharides, which further emphasizes the chemical abundance and structural variation within the genus Laurencia. These results collectively affirm the existence of various bioactive compounds in red algae and strengthen their potential as important sources of biologically active natural products.

The GC–MS analysis of the ethanolic extract of *L. papillosa* showed a wide variety of bioactive compounds, encompassing hydrocarbons, terpenoids, and fatty acid derivatives, which together enhance its notable biological potential. The primary components recognized including 17-(1,5-dimethylhexyl)-10,13-dimethyl compound, 3,7,11,17-tetramethyl-2-hexadecen-1-ol, and Decane, 2,2,3-trimethyl suggest the prevalence of long-chain aliphatic and terpenoid frameworks. These substances are recognized for their antimicrobial, antioxidant, and cytotoxic effects, as noted in several species of Laurencia [45;46]. The occurrence of fatty acids like palmitic, oleic, and linoleic acids in L. papillosa corresponds with results from Cortes et al. [47], who indicated that palmitic acid is the main component in *Ceramium rubrum*. In the current research, comparable long-chain saturated and unsaturated fatty acids were identified, reinforcing the idea that fatty acids play a significant

role in the bioactivity of red algal extracts. These compounds are essential for disrupting microbial membranes, blocking cell wall formation, and hindering nutrient absorption [48].

Additionally, the significant presence of palmitic acid noted in related research has been linked to cytotoxic and antimicrobial characteristics. Earlier studies showed that palmitic acid triggers apoptosis in human leukemic cell lines (MOLT-4) and serves as a selective inhibitor of DNA topoisomerase I, implying its promise as a key compound in anticancer drug research [49]. The presence of both saturated and unsaturated fatty acids, along with hydrocarbon derivatives, boosts the extract's total bioactivity, offering a synergistic impact against pathogenic microorganisms. Consequently, the GC–MS profile of L. papillosa verifies its chemical diversity and pharmacological significance, akin to other bioactive marine red algae. Fatty acids, terpenoids, and hydrocarbon compounds may account for the wide-ranging biological effects noted, which include antimicrobial, antioxidant, and cytotoxic properties. These results emphasize L. papillosa as a potential candidate for deeper investigation in natural product-driven drug development.

In conclusion, the ethanolic extract of *L. papillosa* showed considerable promise as a natural source of bioactive compounds possessing larvicidal and antimicrobial properties. The integrated findings from phytochemical, FT-IR and GC–MS analyses indicated the existence of various functional groups and key components including terpenoids, phenolics, and fatty acids, notably palmitic and oleic acids recognized for their biological effectiveness. These compounds probably work together to damage cell membranes, hinder enzyme function, and obstruct larval growth. The results align with earlier studies on red algae, validating *L. papillosa* as a species that is chemically abundant and pharmacologically significant. Thus, it offers significant potential for creating environmentally safe, natural larvicidal and antimicrobial products.

Acknowledgement

The authors wish to convey their heartfelt thanks to the Director of the Centre for Research in Medical Entomology (CRME), Indian Council of Medical Research (ICMR), Madurai, for supplying mosquito eggs for the research. The authors express gratitude to the Department of Rural Development Science at Arul Anandar College for offering the laboratory resources required to conduct this research.

Funding

The research received no external funding

Declaration

Consent to participate: Not applicable

Consent for publication: Not applicable

Conflicts of interest: The authors declare no competing interests.

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