

# Application of *Eucheuma Cottonii* Carrageenan as an Emulsifier in the Making of Mosquito Repellent Lotion from Clove Leaf Essential Oil (*Syzygium Aromaticum*)

Nurhayati<sup>1</sup>, Hasnah Natsir<sup>1,\*</sup>, Indah Raya<sup>1</sup>, Paulina Taba<sup>1</sup>,  
Abdul Wahid Wahab<sup>1</sup>, Herlina Rasyid<sup>1</sup> and Rizal Irfandi<sup>2</sup>

<sup>1</sup>Department of Chemistry, Faculty of Mathematics and Natural Sciences, Hasanuddin University, South Sulawesi 90245, Indonesia

<sup>2</sup>Department of Chemistry, Faculty of Mathematics and Natural Science, Universitas Negeri Makassar, Jalan Daeng Tata Raya Makassar 90244, Indonesia

(\*Corresponding author's e-mail: [hasnahnatsir@unhas.ac.id](mailto:hasnahnatsir@unhas.ac.id))

Received: 16 April 2025, Revised: 24 May 2025, Accepted: 5 June 2025, Published: 20 August 2025

## Abstract

Dengue fever in Indonesia continues to increase every year. One form of prevention is the utilisation of clove leaf essential oil (*Syzygium aromaticum*) as an anti-mosquito lotion. The abundance of *Eucheuma cottonii* in Indonesia has the potential to increase the production of carrageenan which can function as an emulsifier for lotions. The objectives of this research to analyze the characteristics and effectiveness of anti-mosquito lotion from clove leaf essential oil with the addition of carrageenan based on red algae *E. cottonii*. The method used was carrageenan extraction with 0.1 N KOH solvent, antimicrobial test with disc diffusion method and antimosquito effectiveness test. Analysis of carrageenan using FTIR showed the presence of galactose 4-sulfate functional group which identified kappa type in the spectrum of 846 - 927  $\text{cm}^{-1}$ . Microstructure characteristics of carrageenan using SEM 2000 $\times$  magnification showed a rough surface and irregular clumps. Characterization of clove leaf essential oil (*Syzygium aromaticum*) using GC-MS instrument showed the main content of the compound 4-allyl-2-methoxyphenol with an area of 78.89% which includes eugenol compounds. The optimal anti-mosquito lotion was F2 with 4% carrageenan and 15% clove leaves essential oil. Formula 2 has a level of liking in the sensory test in the neutral category, but has good effectiveness with the support of several characteristics that meet quality standards including a pH value of 7.1, a viscosity value of 5,481.8 cP, a moisture content of 4.91% and microbial inhibition of *E. coli* and *S. aureus* at a good concentration of 40%.

**Keywords:** Anti-mosquito lotion, Carrageenan, Clove leaf essential oil, *Eucheuma cottonii*

## Introduction

Indonesia is an archipelago with a tropical climate, so it has a lot of biodiversity on land and sea. Seaweed is an abundant and important marine resource for the national economy, one of which *Eucheuma* species produces agar and carrageenan. Indonesia imports 1,380 tonnes or about 70% of domestic carrageenan needs, while local production has not met the growing demand [1]. Seaweed from red algae is more widely cultivated, because it has higher biological activity, so the content of primary and

secondary metabolite compounds is also

higher than other types of algae [2].

In addition to abundant marine natural resources, Indonesia also has abundant terrestrial natural resources. One of its superior products is the clove plant, which produces essential oil. Clove contain eugenol (70% - 96%) with many advantages, such as anticancer, antibacterial, antifungal, antioxidant, anti-inflammatory and anti-insect. Eugenol is neurotoxic which can disrupt the nervous system of mosquitoes

and its punget aroma can provide better protection against mosquito bites. These advantages can be utilised as a natural and safe anti-mosquito product for humans. Various studies on anti-mosquito lotions from essential oils of various plants rely solely on aroma to repel mosquitoes, such as citronella-containing lemongrass [3] and evodiamine-containing zodia leaves [4].

Currently, health problems, especially dengue fever, are still a big problem. Since the beginning of 2024, more than 14 million cases of dengue fever and more than 10,000 deaths due to dengue fever worldwide [5]. In Indonesia, in 2024 dengue cases will be reported to reach 88,593 with a death toll of 621 people [6]. Dengue fever is an infectious disease caused by the dengue virus which is transmitted through the bite of the *Aedes aegypti* or *Aedes albopictus* mosquito [7].

One way to prevent dengue fever is to eliminate or reduce the population of vector mosquitoes such as *Aedes* sp. The main vector control activities in Indonesia are carried out on mosquito larvae and adult mosquitoes. Control of adult mosquitoes is done by fogging to break the chain of transmission from infected mosquitoes to humans. Specifically for mosquito larvae, mosquito nest eradication is carried out by draining, covering and burying programs chemically with insecticides, biologically using natural enemies such as predators, bacteria, and other methods such as using mosquito nets, using mosquito repellents and installing wire mesh [6,8]. Individual self-protection from the risk of DHF virus transmission can be done in various ways including using repellents, and wearing clothes that reduce mosquito bites.

People generally use mosquito coils, mosquito sprays, and electric mosquito repellents that are derived from chemicals and have negative impacts on the human body such as shortness of breath, and also cause air pollution [9]. Most of the mosquito repellents available today contain diethyltoluamide (DEET) chemicals as active ingredients. DEET compounds are easily absorbed through the skin and enter the bloodstream, affecting the nervous system. In particular, DEET causes seizures and even death in

some human individuals. The dangers and warnings are a clear enough reason that alternative safe active ingredients are needed in the formulation of mosquito repellent to facilitate its use, mosquito repellent will be made in the preparation of mosquito repellent lotion.

Based on the above problems, in this study, an anti-mosquito lotion was made from clove leaf essential oil, using carrageenan from red algae (*E. cottonii*) as an emulsifier.

## Materials and methods

### Preparation of red algae (*E. cottonii*)

The red algae obtained were soaked with clean water for 30 min, then washed thoroughly to remove dirt and attached objects such as sand and gravel. Furthermore, it is drained and cut with a size of 2 - 4 cm.

### Extraction of carrageenan from red algae (*E. cottonii*)

Prepared red algae were then weighed as much as 160 g. Furthermore, it was extracted using 0.1 N KOH solvent with a ratio of solvent and raw material of 5:1 for 60 min at a temperature of 70 - 80 °C. The extracted *E. cottonii* filtrate was washed with water and filtered. Then dried in the oven at 60 °C, and mashed using a hammer mill and sieved using 100 mesh sieving.

### Extraction of clove leaf essential oil

Weighed 5 kg of washed and dried clove leaves, distilled for 7 h, then separated the oil and water.

### Lotion formulation

The formulation of anti-mosquito lotion refers to the research conducted by Dewi *et al.* [10] modified. The modification is done by replacing the active ingredient extract with clove leaf essential oil extract (*Syzigium aromaticum*) and adding an emulsifier in the form of carrageenan extracted from red algae (*E. cottonii*). This test was carried out 3 times. The formulation of anti-mosquito lotion can be seen in **Table 1**.

**Table 1** Anti-mosquito lotion formula with carrageenan.

Material	Formula				Usability
	F0 (%)	F1 (%)	F2 (%)	F3 (%)	
Clove leaf essential oil ( <i>Syzigium aromaticum</i> )	-	10	15	20	Active substance
Stearic acid	15	15	15	15	Emulsifiers
<i>Eucheuma cottonii</i> carrageenan	-	2	4	6	Thickener
Glycerin	15	15	15	15	Emollient
Adeps Lanae	3	3	3	3	Emollient
Methyl Paraben	0.02	0.02	0.02	0.02	Preservatives
Propyl Paraben	0.05	0.05	0.05	0.05	Preservatives
Oil Rosae	1.5	1.5	1.5	1.5	Fragrance
Aquadest	100 mL	100 mL	100 mL	100 mL	Solvent

Description: From 100%, 60 mL of lotion preparation was made.

### Sensory test of anti-mosquito lotion

The sensory test used hedonic scale with 15 panelists and was processed using SPSS V. 25 with Kruskal-Wallis test and Mann-Whitney test as the next step. The parameters observed were appearance, color, aroma, and texture. Observations with a hedonic scale worth 1 to 5: 1) Very dislike, 2) Dislike, 3) Neutral, 4) Like, 5) Very like [11].

### Characteristic test of anti-mosquito lotion

Characteristic tests on anti-mosquito lotions consist of pH measurements, viscosity [12], specific gravity and water content.

### Antimicrobial test against *E. coli* and *S. aureus* bacteria

Testing the antibacterial activity of the lotion was done by disc diffusion method using paper discs with a diameter of 5 mm. Paper discs were dipped into the sample at each concentration of 20%, 30% and 40%, then placed on NA media that had been inoculated with test bacteria (*E. coli* and *S. aureus*). Then incubated at 37 °C for 2×24 h. Positive control was 0.1% chloramphenicol solution and negative control was aquades [13].

### Effectiveness test of anti-mosquito lotion

The effectiveness test was conducted by

introducing 20 test mosquitoes into each of the 5 test cages (control, F0, F1, F2 and F3). Then the left hand was smeared with antimosquito lotion evenly and the right hand was not smeared (as a comparison). Replication was done twice. The observation time was 20 min with an interval of 5 min [19].

### Characterization of clove leaf essential oil extract (*Syzigium aromaticum*) using Gas Chromatography-Mass Spectrometry (GC-MS)

Essential oil from clove leaves is then analyzed using GC-MS with RTx-5MS column conditions with a length of 30 meters (0.25 mm), column oven temperature of 70 °C, injection temperature of 300 °C, water control mode used in the form of pressure with the pressure used of 13.7 kPa, total flow of 50 mL/min, and column flow of 0.5 mL/min [14].

### Characterization of *E. cottonii* carrageenan using Fourier Transform Infrared (FTIR)

Characterization of carrageenan structure using FTIR spectrophotometry was carried out by mixing 1 mg of fucoidan powder with 100 mg of KBr, crushed in a mortar until homogeneous to obtain a pellet. The pellet was put into a simple pan to record the infrared spectrum at a wavelength of 4,000 - 500 cm<sup>-1</sup>.

**Characterization of *E. cottonii* carrageenan using Scanning Electron Microscope (SEM)**

As much as 200 mg of carrageenan powder was printed on carbon tape, dried and then coated with gold. Placed on the device and observed the surface of the sample directly with 10 - 1500× magnification, dept of field 4 - 0.4 mm and resolution of 1 - 10 nm.

**Results and discussion**

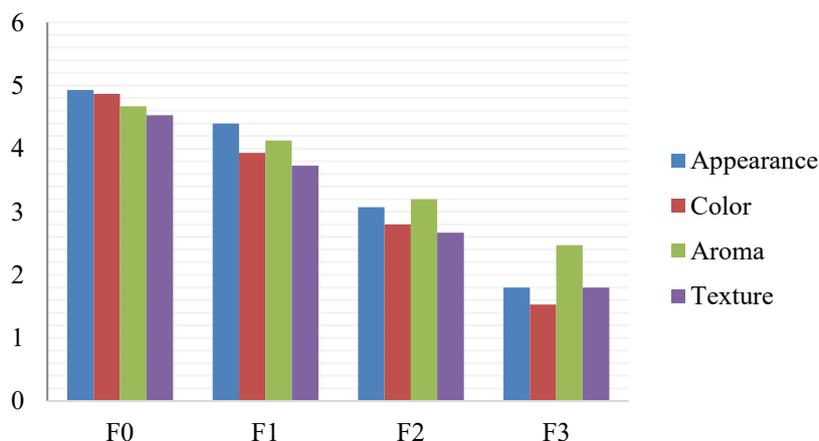
**Sensory test of anti-mosquito lotion**

Extract of Clove Leaf Essential Oil Sensory testing was conducted by 15 panelists by measuring the level of appearance, color, aroma, and texture using a hedonic scale. Data were analyzed using Kruskal-Wallis test followed by Mann-Whitney test using SPSS 25.

**Table 2** Average value of sensory test of anti-mosquito lotion preparations of clove leaf essential oil extracts.

Parameters	Sample			
	F0	F1	F2	F3
Appearance	4.93± 0.258 <sup>a</sup>	4.40 ± 0.737 <sup>b</sup>	3.07 ± 0.799 <sup>c</sup>	1.80 ± 0.775 <sup>d</sup>
Color	4.87± 0.352 <sup>a</sup>	3.93 ± 0.704 <sup>b</sup>	2.80 ± 0.676 <sup>c</sup>	1.53 ± 0.640 <sup>d</sup>
Aroma	4.67 ± 0.488 <sup>a</sup>	4.13 ± 0.915 <sup>a</sup>	3.20 ± 0.775 <sup>b</sup>	2.47 ± 0.915 <sup>c</sup>
Texture	4.53 ± 0.640 <sup>a</sup>	3.73 ± 0.799 <sup>b</sup>	2.67 ± 0.617 <sup>c</sup>	1.80 ± 0.775 <sup>d</sup>

Description: 1 = Strongly dislike, 2 = Dislike, 3 = Neutral, 4 = Like, 5 = Strongly like a, b, c, d = Similar letter notation means no significant difference at the Mann-Whitney test level ( $p > 0.05$ ).



**Figure 1** Distribution of panelists' level of liking for the preparation of anti-mosquito lotion.

The panelist liking test was conducted to determine the level of panelist preference for the lotion preparation that had been made. The appearance of F0 tends to be preferred because it has the characteristics of lotions in general that are not given the addition of clove leaf essential oil extract and *E. cottonii* carrageenan. Based on the level of panelists' preference for color, panelists preferred F0 compared to other formulas. This is because the color of F0 is white and the other formulas are brown. The color change that occurs is due to the addition of clove leaf essential oil and *E. cottonii* carrageenan. The white

color tends to have a greater appeal because the majority of lotions spread in the community are white so that the level of acceptance of panelists for lotions other than white is still low. The clove leaf essential oil lotion preparation has a distinctive aroma, namely the distinctive aroma of clove leaf essential oil. This is due to the eugenol content that dominates clove essential oil. The aroma of cloves is still not a special attraction for lotion preparations because of its distinctive aroma characteristics that tend to be sharp and spicy. Several studies have found good percentage for lotion preparations. Commonly used percentage are around

5% - 15%, depending on the type of essential oil and the intended use. Generally, higher concentrations are applied for more than just moisturizing the skin, such as antiinflammatory and antimosquito. The texture liking level of the lotion tended to favor F0 by the panelists because of its smooth and soft texture. When compared to the other formulas which tended to be rough according to the variation in concentration of *E. cottonii* carrageenan addition and slightly oily with the addition of clove leaf essential oil.

Based on the sensory test, the ideal antimosquito lotion formula is F2 with 15% clove leaf essential oil

concentration which is good for warding off mosquitoes. Some other plant essential oils with a percentage of about 15% can provide 50% protection for 6 h. Regarding antimosquito lotion products on the market, the percentage of essential oil used is not specified. However, some studies can serve as a reference for the percentage that is effective and safe for the skin ranging from 5% - 20%.

#### Characteristic test of anti-mosquito lotion

Characteristic of anti-mosquito lotion can be seen in **Table 3**.

**Table 3** Characteristic of anti-mosquito lotion.

No.	Parameters	Formula				Standard
		F0	F1	F2	F3	
1	pH	7.74 ± 0.01	7.12 ± 0.015	7.1 ± 0	6.98 ± 0.015	4.5 - 8.0 <sup>a</sup>
2	Viscosity (cP)	7,094.37 ± 56.602	6,992.70 ± 31.624	5,481.80 ± 24.278	2,610.53 ± 213.057	2,000 - 50,000 <sup>a</sup>
3	Specific gravity (g/mL)	1.004 ± 0.005	1.005 ± 0.001	1.028 ± 0.006	1.037 ± 0.002	0.95 - 1.05 <sup>a</sup>
4	Moisture content (%b/b)	6.22 ± 0.006	5.06 ± 0.01	4.91 ± 0.015	3.92 ± 0.025	≤ 10% <sup>b</sup>

Description: <sup>a</sup> = SNI 16-4399-1996; <sup>b</sup> = Herbal Pharmacopoeia Edition II.

Measuring the pH of clove essential oil extract anti-mosquito lotion aims to ensure the safety of the user's skin. A pH value that is too acidic can irritate the skin while a pH value that is too alkaline can dry the skin [15]. Based on the SNI 16-4399-1996 standard, the ideal pH value for lotions is between 4.5 - 8.0. Based on the pH measurement data of the lotion using a pH meter, from F0 to F3, the pH decreased. This is in accordance with research conducted by Dewi *et al.* [10], which states that the higher the clove essential oil content, the smaller the pH obtained. Referring to SNI standards, all formulas meet the requirements to be applied to the skin with a pH value of 6.98 - 7.74.

The viscosity test of anti-mosquito lotion aims to determine the viscosity level of the lotion so that it can have good spreadability on the skin. According to Dewi *et al.* [10], the presence of thickeners in lotion formulations is useful to prevent particle separation from emulsions so as to maintain lotion stability. Based on the results of the table above, all formulas meet the quality requirements of anti-mosquito lotion preparations. According to Dewi *et al.* [10], the higher

the concentration of clove oil added, the lower the viscosity, due to the consistency of clove oil which is more liquid than the lotion preparation base. Good viscosity is indicated by the fact that the higher the viscosity value, the harder particles movement will tend to move, so the lotion will be more stable.

Based on **Table 3** all lotion formulas meet the SNI 16-4399-1996 standard and exceed the specific gravity of water. According to Rowe *et al.* [16], the specific gravity of lotion tends to be greater than the specific gravity of water depending on the increase in emulgator concentration in the preparation, the increase in specific gravity of lotion preparation is directly proportional to the concentration of emulgator (carrageenan *E. cottonii*).

The anti-mosquito lotion moisture content test is one of the tests to measure the quality of lotion preparations that are effective and safe to use. Too high moisture content can cause rapid growth of microorganisms. The right moisture content will make the lotion have good adhesion and effectiveness in its function as an anti-mosquito lotion. There is no

reference in SNI standards or other standards, but based on Herbal Pharmacopoeia Edition II [17], a good moisture content to maintain the quality of simplisia is  $\leq 10\%$ . The following is the water content of each formula. Based on the table above, all formulas have a moisture content of  $< 10\%$  so that the quality of the lotion can be maintained properly. This is also in accordance with the formulation of each lotion produced in the addition of distilled water.

**Antimicrobial test against the growth of *E. coli* and *S. aureus* bacteria**

Antimicrobial testing of anti-mosquito lotions uses the *Agar Diffusion Disk* method. According to Rivera *et al.* [18], the *Agar Diffusion Disk* method is

based on the diffusion of a certain amount of antimicrobial agent contained in a paper disk placed on the surface of an agar medium that has been previously seeded with a standardized microbial inoculum. As the sample diffuses, it will form a zone of inhibition, the concentration of which will be sufficient to inhibit the growth of the inoculated microorganism from growing. The larger the diameter of the inhibition zone produced and exceeds that of the positive control, the greater the potential in inhibiting the growth of the inoculated microorganisms. In the antimicrobial testing of this lotion, the test bacteria used were *E. coli* and *S. aureus*. The positive control used chloramphenicol 0.1% and the negative control used DMSO 0.1%.



**Figure 2** Lotion antimicrobial test.

**Table 4** Diameter of the clear zone of the anti-mosquito lotion dosage formula.

Formula (F)	Concentration	Diameter of clear zone of test bacteria (mm)	
		<i>E. coli</i>	<i>S. aureus</i>
F0	20%	8.4	9.9
	30%	10.4	10.3
	40%	11.4	11.8
	+	27	27.8
	-	0	2
F1	20%	11	9.3
	30%	11.4	11.4
	40%	11.7	11.9
	+	29	25.3
	-	0	0

Formula (F)	Concentration	Diameter of clear zone of test bacteria (mm)	
		<i>E. coli</i>	<i>S. aureus</i>
F2	20%	10.6	10.4
	30%	10.8	11.6
	40%	11.3	12.1
	+	28.9	27.5
	-	10.1	10
F3	20%	9.8	10.4
	30%	10.5	11.6
	40%	11.8	12.3
	+	29.4	28.1
	-	9.4	9.8

Based on the observation results (**Figure 2**), clear zone diameter data was obtained for each anti-mosquito lotion formula. The diameter of the clear zone in each anti-mosquito lotion preparation formula with 2 test bacteria showed significant potential inhibition of microorganism growth as the concentration of the lotion preparation formula increased (**Table 4**). Although still not exceeding the inhibitory ability of the positive control (chloramphenicol), each formula can increase its inhibitory power with increasing concentration. So it can be concluded that the higher the concentration of the lotion preparation formula, the higher the antimicrobial ability. Antimicrobial testing of clove leaf mosquito repellent lotion is conducted to increase

the usability of the lotion as skin protector from pathogenic microorganisms. In addition, the presence of antimicrobial activity in the lotion may indicate that the lotion is more effective as a mosquito repellent due to the lethal effect of the same antimicrobial active compound in the antimosquito effectiveness test [19].

#### Effectiveness test of anti-mosquito lotion

The anti-mosquito lotion effectiveness test was conducted in cages containing mosquitoes that had never sucked blood with a total of 5 cages. The test used 4 lotion formulas and 1 comparison (no lotion). The results of the lotion effectiveness test can be seen in **Table 5**.

**Table 5** Effectiveness test of anti-mosquito lotion.

No.	Formula (F)	Jumlah nyamuk yang menempel (ekor)
1	Tanpa losion	$5 \pm 0.577$
2	F0	$2 \pm 0.577$
3	F1	$2 \pm 0$
4	F2	$1 \pm 0.577$
5	F3	$0 \pm 0.577$

Description:

F0 = Without the addition of clove leaf essential oil extract.

F1 = Addition of clove leaf essential oil extract by 10%.

F2 = Addition of 15% clove leaf essential oil extract.

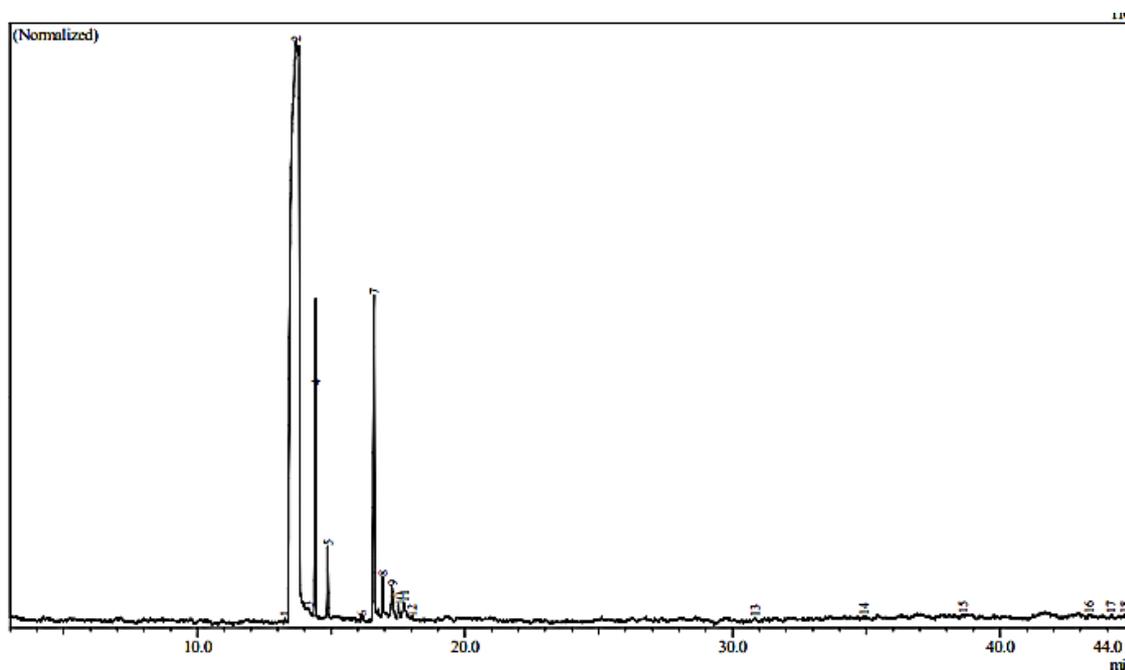
F3 = Addition of clove leaf essential oil extract by 20%.

Based on the lotion effectiveness test, it shows that the higher the concentration of clove leaf essential oil extract added to the lotion formula, the more effective it is as an anti-mosquito lotion. This is due to the sharp aroma of clove leaf essential oil that can prevent mosquitoes from sticking to the skin. The eugenol content in clove leaves has neurotoxic properties that can cause insects to become inactive. Neurotoxic works in the process of suppressing the nervous system of insects which can be characterized by the insect's body which when touched feels soft and limp. In a study conducted by Juniyanti *et al.* [20], showed that the effectiveness of clove leaf extract lotion was good at a concentration of 3% with a repulsion percentage of 85.5% of 50 test mosquitoes.

This was also stated by Ndalú [21], which resulted in an average value of 1.3 mosquitoes from 25 test mosquitoes.

#### GC-MS analysis of clove leaf essential oil (*Syzygium aromaticum*)

Analysis of the constituent compound components of clove leaf essential oil extract (*Syzygium aromaticum*) using GC-MS instrument. The results obtained in the form of spectra and chromatograms. Mass spectra analysis is based on the *base peak*, similarity index (SI) and fragmentation pattern of the spectrum compared to the spectrum of the *Wiley 229 LIB library*. The analysis results can be seen in **Figure 3** and The interpretation results can be seen in **Table 6**.



**Figure 3** GC-MS results of clove leaf essential oil.

**Table 6** Interpretation of GC chromatogram of clove leaf essential oil.

Peak	RT (Retention Time)	Area (%)	Compounds name
1	13.258	0.19	Alpha-cubebene
2	13.700	78.89	4.allyl-2-methoxyphenol
3	14.150	0.13	Methyleugenol
4	14.425	5.61	Caryophyllene
5	14.877	1.17	1,4,8cycloundecatriene, 2,6,6,9-tetramethyl,(E,E,E)-
6	16.150	0.22	Caryophyllene oxide
7	16.599	7.08	(-)-5-oxatricyclo[8.2.0.0(4,6)]dodecane,12-trimethyl-9-methylene

Peak	RT (Retention Time)	Area (%)	Compounds name
8	16.925	1.39	(1R,3E,7E,11R)-1,5,5,8-Tetramethyl-12-oxabicyclo[9.1.0]dodeca-3,7-diene
9	17.285	1.33	10,10-dimethyl-2,6-dimethylenebicyclo[7.2.0]undecan-5.beta-ol
10	17.525	0.52	Isoaromadendrene epoxide
11	17.725	1.06	1,3B,6,6-Tetramethyldecahydro-1H-Cyclopropa[7,8]azuleno[4,5-B
12	18.000	0.12	2-Ethyl-5,6,7-trimethoxyisoquinoline-1,3,4-trione
13	30.848	0.18	Terephthalic acid, butyl isopropyl ester
14	34.900	0.27	Dotriacontane
15	38.609	0.70	1H-Benzocyclohepten-7-ol,2,3,4,4a,5,6,7,8-octahydro-1,1,4a,7-tetramethyl-,cis-
16	43.325	0.55	3-(6-Nitro-1-oxo-1,3 dihydro-isoindol-2-yl)-propionic acid
17	44.150	0.29	2-(3,5-dichloro-6-dimethylaminopyridyloxy)-2-propionic acid
18	44.662	0.31	Succinic acid,hept-2-yl 2,4,6-trichlorophenyl ester

In the GC chromatogram data in **Table 6**, there are 3 peaks that have a larger area than other peaks, namely peaks number 2, 4, and 7. The compound at peak number 2 with a retention time of 13.7 min and S1 of 95 is similar to the compound 4-allyl-2-methoxyphenol with the molecular formula  $C_{10}H_{12}O_2$ . The compound at peak number 4 with a retention time of 14.425 min and S1 of 97 is similar to the compound caryophyllene with the molecular formula  $C_{15}H_{24}$ . The compound at peak number 7 with a retention time of 16.6 min and S1 of 97 is similar to the compound (-)-5-oxatricyclo[8.2.0.0(4,6)]dodecane,12-trimethyl-9-methylene with the molecular formula  $C_{15}H_{24}O$ . Among the 3 compounds, the most is the compound 4.allyl-2-methoxyphenol with an area of 78.89% which is included in the eugenol compound. The same thing was reported by Sanga *et al.* [22], the most compound of clove essential oil is eugenol (59.38%) with a retention time of 17.946; and Budiman *et al.* [23] eugenol is 87.24%.

#### Characterization of carrageenan using FTIR

The wavelength absorption in **Table 7** shows a broad absorption at a wavelength of  $3,479\text{ cm}^{-1}$  indicating an -OH group, and a sharp and narrow absorption at a wavelength of  $1,070\text{ cm}^{-1}$  indicating a galactose glycosidic bond. In addition, there are sharp and narrow absorptions at wavelengths of 927 and  $846\text{ cm}^{-1}$  that identify the presence of 3,6-anhydrogalactose group and galactose-4-sulfate group. The presence of 3,6-anhydrogalactose group shows the characteristics of kappa carrageenan. This is in line with research by Amin *et al.* [24], that the characteristics of carrageenan extracted from *E. cottonii* belong to the kappa type with an absorption region of  $848.53\text{ cm}^{-1}$  showing galactose-4-sulfate bonds and  $928.56\text{ cm}^{-1}$  showing 3,6-anhydrogalactose bonds. The characteristic of kappa carrageenan is the presence of D-galactose-4-phosphate and 3,6-anhydro-D-galactose groups [25].

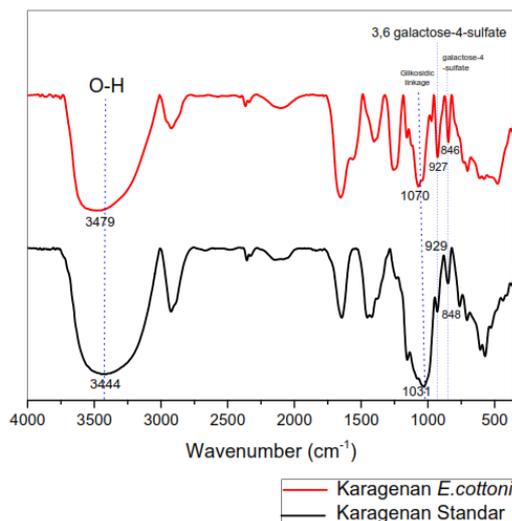


Figure 4 Identification results of *E. cottonii* carrageenan and standard carrageenan.

Table 7 infrared absorption data of *E. cottonii* carrageenan.

Wavelength (cm <sup>-1</sup> )	Bonding	Function group	Intensity
3479	-OH	Hydroksyl	Medium
1070	C-O-C	Galactose glycosidic	Strong
927	C-O	3,6 anhydrogalactose	Strong
846	C-O-SO <sub>3</sub>	galactose-4-sulfate	Strong

**Characterization of carrageenan *E. cottonii* using SEM**

SEM analysis is useful to determine the

microstructure (including porosity and crack shape) of solid objects [26].

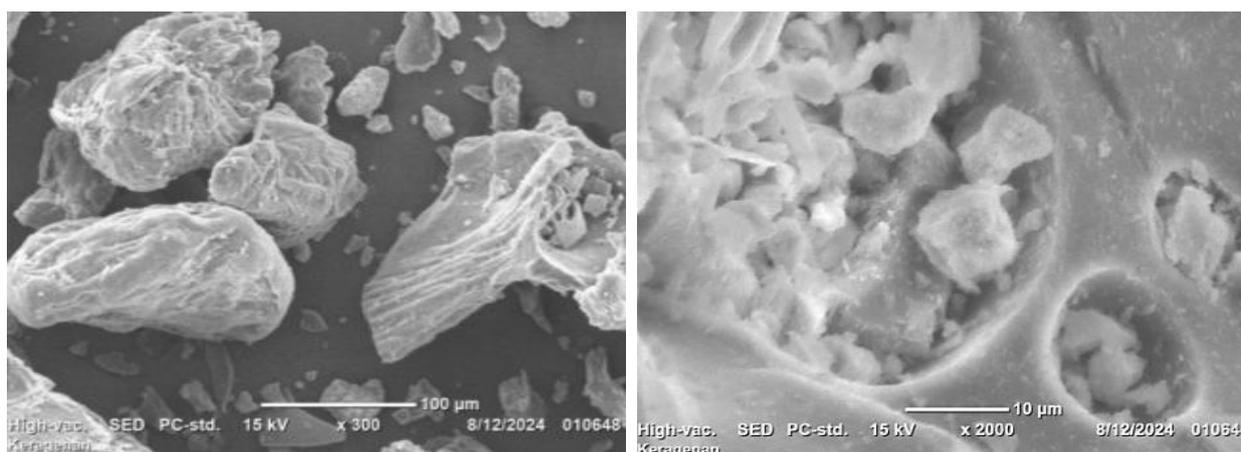


Figure 5 Structure of *E. cottonii* carrageenan magnification (a) 300× and (b) 2000×.

Based on Figure 5, the microstructure characteristics of carrageenan using SEM magnification of 300× shows a rough surface and irregular clumps, while at 2000× magnification shows pores on the surface that look more clearly there are irregular clumps. This is due to the strong reaction

between carrageenan and KOH which can increase the ionic strength of the carrageenan polymer chain and form a more stable gel. This is in accordance with research by Bhernama [27], who compared the structure of carrageenan with KOH and NaOH. The results obtained, carrageenan with KOH solvent has an

uneven surface structure with gel lumps, while with NaOH solvent the surface tends to be flat and smooth. According to Winarno [28], this is because the solvents potassium hydroxide and sodium hydroxide have a major influence on the extraction of seaweed *E. cottonii* into carrageenan, which  $K^+$  ions and  $Na^+$  have a different role in the strength of the gel.  $K^+$  ions produce a better surface structure than  $Na^+$  ions. KOH solvent significantly affects the viscosity of carrageenan through molecular degradation and sulfate reduction, but its strong ionic interactions improve functional properties such as gel strength, making it a versatile choice in various applications such as antimosquito lotions [29].

### Conclusions

Characterization of carrageenan from red algae (*E. cottonii*) using FTIR indicated that it is a type of kappa carrageenan with the characteristics of the 3,6-anhydrogalactose functional group at a wavelength of  $927\text{ cm}^{-1}$ . The results of analysis using SEM instrument showed a rough surface and irregular clumps. Characterization of clove leaf essential oil (*Syzygium aromaticum*) using GC-MS showed the presence of the main component contained in clove leaf essential oil, namely 4-allyl-2-methoxyphenol. Based on the tests to determine the conformity of the lotion with the predetermined quality standards, the optimal percentage of carrageenan in making antimosquito lotion is F2 with 4% carrageenan. Formula 2 has a level of liking in the sensory test in the neutral category, but has good effectiveness with the support of several characteristics that meet quality standards including a pH value of 7.1, a viscosity value of 5,481.8 cP, a moisture content of 4.91% and microbial inhibition of *E. coli* and *S. aureus* at a good concentration of 40%.

### Acknowledgements

The greatest appreciation goes to the research team who have fully contributed to the completion of this research, and thanks to hasanuddin university for providing equipment that supports this research as well as the panelists who have participated in completing the necessary data.

### Declaration of Generative AI in Scientific Writing

The author acknowledge the use of generative tools (e.g., QuillBot) in the preparation of this manuscript, specifically for language editing and grammar correction. No content generation or data interpretation was performed by AI. The authors take full responsibility for the content and conclusions of this work.

### CRedit Author Statement

**Nurhayati:** Conceptualization, Methodology, Software, Investigation, Funding Acquisition, Resources, and Writing – original draf.

**Hasnah Natsir:** Conceptualization, Methodology, Recources, Validation, Formal analysis, Data Curation, and Project Administration..

**Indah Raya:** Conceptualization, Methodology, Formal analysis, Validation, and Data Curation.

**Paulina Taba:** Data curation, Formal Analysis, Validation, and Supervision.

**Abdul Wahib Wahab:** Validation, Formal analysis, Data Curation, Writing-Reviewing and Editing.

**Herlina Rasyid:** Formal analysis, Data curation, Validation, Software, and Visualization.

**Rizal Irfandi :** Data Curation, Software, and Visualization.

### References

- [1] M Borg, S Krueger-Hadfield, C Destombe, J Collén, A Lipinska and SM Coelho. Red macroalgae in the genomic era. *New Phytologist* 2023; **240(2)**, 471-488.
- [2] V Dolganyuk, D Belova, O Babich, A Prosekov, S Ivanova, D Katserov, N Patyukov and S Sukhikh. Microalgae: A promising source of valuable bioproducts. *Biomolecules* 2020; **10(8)**, 1153.
- [3] YO Nainggolan, Masrullita, R Dewi, N Za and E Kurniawan. Pembuatan formula *Lotion* antinyamuk dari minyak atsiri sereh wangi (*Citronella oil*) (in Indonesian). *Chemical Engineering Journal Storage* 2022; **2(5)**, 29-41.
- [4] ARY Eff, RDLA Pertiwi and TP Utami. Efektivitas repelan losion minyak atsiri daun zodia (*Evodia suaveolens*) terhadap nyamuk *Aedes aegypti* linnaeus (in Indonesian). *Majalah Farmasetika* 2020; **4(1)**, 119-124.

- [5] European Centre for Disease Prevention and Control. *Dengue worldwide overview*. European Centre for Disease Prevention and Control, Solna, Sweden, 2025.
- [6] Indonesian Ministry of Health. *Demam berdarah masih mengintai*, Available at: <https://sehatnegeriku.kemkes.go.id/baca/mediakom/20240521/2845637/mediakom-165/>, accessed November 2024.
- [7] Y Agnesia, Nopianto, SW Sari and DW Ramadhani. *Demam Berdarah Dengue (DBD): Determinan dan pencegahan (in Indonesian)*. NEM Pekanbaru, Riau, Indonesia, 2023.
- [8] Soegijanto. *Demam berdarah dengue edisi kedua (in Indonesian)*. Airlangga University Pres, Surabaya, Indonesia, 2006.
- [9] RD Halim and A Fitri. Aktivitas minyak sereh wangi sebagai antinyamuk (in Indonesian). *Jurnal Kesehatan Masyarakat Jambi* 2020; **1**, 28-34.
- [10] B Dewi, T Wulandari and S Yanti. Formulasi dan uji efektivitas *Lotion* antinyamuk minyak cengkeh (*Syzygium aromaticum*) (in Indonesian). *Jurnal Ilmiah Farmacy* 2020; **7(2)**, 277-286.
- [11] D Kurnianingsih. Uji efektivitas sediaan krim kombinasi ekstrak daun bakau hitam (*Rhizophora mucronata*) dan jeruk purut (*Citrus hystrix*) terhadap bakteri *Staphylococcus aureus* (in Indonesian). *Jurnal Ilmiah JOPHUS: Journal of Pharmacy UMUS* 2021; **2(1)**, 28-35.
- [12] R Siskayanti and ME Kosim. Analisis konsentrasi minyak atsiri sereh sebagai aditif dalam pembuatan lotion antinyamuk. *Jurnal Redoks* 2021; **6(1)**, 26-34.
- [13] IH Goetie, R Sundu and R Supriningrum. Uji aktivitas antibakteri ekstrak kulit batang sekilang (*Embelia borneensis* Scheff) terhadap bakteri *Escherichia coli* dan *Staphylococcus aureus* menggunakan metode disc diffusion (in Indonesian). *Jurnal Riset Kefarmasian Indonesia* 2022; **4(2)**, 144-155.
- [14] IBD Kapelle, HJ Sohilit and ML Haluruk. Analisis minyak atsiri dari bunga dan ganggang cengkeh (*Syzygium aromaticum* L.) asal pulau saporua maluku (in Indonesian). *Tekonotan* 2023; **17(2)**, 131-136.
- [15] D Pertiwi, R Desnita and S Luliana. Effect of pH on alpha arbutin stability in niosomal gel (in Indonesian). *Majalah Farmasetik* 2020; **16(1)**, 91-100.
- [16] RC Rowe, PJ Sheskey and PJ Weller. *Handbook of pharmaceutical excipient*. The Pharmaceutical Press, London, 2006.
- [17] Indonesia Ministry of Health. *Pharmacopeia Edition II*. Ministry of Health of the Republic of Indonesia, Jakarta, Indonesia, 2017.
- [18] A Rivera, B Vinado, N Benito, F Docobo-Pérez, F Fernández-Cuenca, J Fernández-Domínguez, J Guinea, A López-Navas, M Ángel Moreno, MN Larrosa, A Oliver and F Navarro. Recommendation of the Spanish Antibiogram Committee (COESANT) for *in vitro* susceptibility testing of antimicrobial agents by disk diffusion. *Enfermedades Infecciosas y Microbiología Clínica* 2023; **41(9)**, 571-576.
- [19] P Setiawan, N Farid, M Yusuf and RM Latelay. Uji stabilitas sediaan losion antinyamuk dengan variasi konsentrasi ekstrak bunga sukun (*Artocarpus altilis*) terhadap nyamuk *Aedes aegypti* (in Indonesian). *Journal of Experimental and Clinical Pharmacy* 2021; **1(2)**, 103-111.
- [20] AR Juniyanti, A Retnaningsih and S Marcellia. Uji formulasi sediaan lotion dari ekstrak daun cengkeh (*Syzygium aromaticum* L.) sebagai repellan terhadap nyamuk *Aedes aegypti* (in Indonesian). *Journal of Pharmacy and Tropical Issues* 2021; **1(1)**, 27-35.
- [21] MK Nдалu. 2020, Tugas akhir efektivitas ekstrak daun cengkeh (*Syzygium aromaticum*) sebagai repellent antinyamuk *Aedes sp* (in Indonesian). Ph. D. Dissertation. Poltekkes Kemenkes Kupang, Nusa Tenggara Timur, Indonesia.
- [22] AG Sanga, HD Mazigo, A Manjurano, D Morona, A Thomas and EJ Kweka. Measuring repellence and mortality effects of clove and cinnamon essential oils impregnated nets against *Anopheles gambiae* senso stricto using tunnel test. *Journal of Natural Pesticide Research* 2023; **5**, 100046.
- [23] Budiman, H Ishak, Stang, E Ibrahim, R Yudhastuti, A Maidin, F Naiem, I Wahid and A Mallongi. Effectiveness of clove oil (*Syzygium aromaticum*) as biolarvaside of *Aedes aegypti*. *Biomedical and Pharmacology Journal* 2022;

- 15(4)**, 2287-2292.
- [24] I Amin, D Setyorin and Indriani. Pengaruh variasi *Solvent* organik dalam proses maserasi karagenan *Eucheuma Cottonii* di PT biota laut ganggang pinrang (in Indonesian). *Prosiding Seminar Nasional Teknologi Industri IX 2022* 2022; **1**, 171-175.
- [25] CS Sani, MDR Oedjoe, W Turupadang. Analysis of yield and type of carrageenan from different strains at Kupang Bay seaweed production center, East Nusa Tenggara. In: *Proceedings of the 3<sup>rd</sup> International Symposium on Transdisciplinary Approach for Knowledge Co-Creation in Sustainability*, Bogor Tengah, Indonesia. 2023.
- [26] V Suprunchuk. Ultrasonic-treated fucoidan as a promising thearapeutic agent. *Polymers in Medicine* 2021; **51(2)**, 85-90.
- [27] BG Bhernama. Analisis karakteristik karagenan *Eucheuma cottoni* asal aceh jaya menggunakan pelarut alkali (KOH dan NaOH) (in Indonesian). *Journal Amina* 2019; **1(2)**, 63-65.
- [28] FG Winarno. *Teknologi pengolahan rumput laut (in Indonesain)*. Pustaka Sinar Harapan, Jakarta, Indonesia, 1990.
- [29] H Laksono, CK Dyah, RPG Putri and M Purwoto. Characteristic of rapid visco analyze carrageenan extract with enzymatic pretreatment of *Kappaphycus striatum*. *ASEAN Journal of Chemical Engineering* 2022; **22(2)**, 326-336.