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DEVELOPMENT OF ANTI MOSQUITO LOTION MADE FROM CLOVE (*SYSYGIUM AROMATICUM*), LANGUSEI (*FICUS MINAHASSAE*), AND APIS DORSATA BINGHAMI HONEYCOMB PLANTS

Abstrak

Nyamuk adalah serangga vektor utama penyebab berbagai penyakit tropis, termasuk malaria, filariasis, dan berbagai penyakit virus, seperti demam berdarah. Indonesia merupakan negara dengan iklim tropis yang karakteristik lingkungannya sangat mendukung untuk berkembangbiaknya nyamuk. Nyamuk merupakan vektor menular yang harus dikendalikan. Upaya untuk mengendalikan vektor nyamuk yaitu salah satunya dengan menggunakan repellent. Repellent adalah bahan kimia yang mempunyai kemampuan untuk menjauhkan serangga sehingga dapat dihindari dari gigitan serangga. Penelitian ini bertujuan untuk mengetahui formula lotion anti nyamuk berbahan baku tanaman cengkih, langusei, dan sarang lebah dan mengetahui hasil uji organoleptik, fisik dan kekuatan repellent lotion anti nyamuk berbahan baku cengkih, langusei, dan sarang lebah. Metode yang di gunakan dalam jenis penelitian ini adalah metode eksperimen laboratorium. Sampel yang digunakan dalam penelitian ini yaitu daun cengkih, langusei, dan sarang lebah. Analisis yang dilakukan yaitu uji organoleptik, uji pH, uji homogenitas, dan uji daya proteksi. Lotion dengan penambahan ekstrak tanaman cengkih, langusei, dan sarang lebah madu memenuhi uji organoleptik, uji syarat mutu fisik dan kekuatan repellent lotion, yaitu mempunyai massa lotion yang halus dan lembut, warna dan aroma lotion khas campuran tanaman cengkih, langusei, dan sarang lebah madu. Hasil penelitian menunjukkan bahwa dari variasi konsentrasi ekstrak daun cengkih, langusei, dan sarang lebah madu (0%, 0,25%, 5%, 10%) hasil terbaiknya adalah formula IV yaitu dengan konsentrasi 10% yang memiliki sifat fisik paling baik, aman untuk digunakan dan daya proteksi yang paling efektif untuk mengusir nyamuk.

Kata kunci: Nyamuk, Repellent, Lotion, Daun Cengkih, Langusei, Sarang Lebah.

Abstract

Mosquitoes are the main insect vectors that cause various tropical diseases, including malaria, filariasis, and various viral diseases, such as dengue fever. Indonesia is a country with a tropical climate whose environmental characteristics are very supportive of mosquito breeding. Mosquitoes are infectious vectors that must be controlled. Efforts to control mosquito vectors include using repellents. Repellent is a chemical that can keep insects away so that insect bites can be avoided. This research aims to determine the formula for anti-mosquito lotion made from cloves, langusei, and beehives and to determine the results of organoleptic, physical tests, and the strength of repellent anti-mosquito lotion made from cloves, langusei, and beehives. The method used in this type of research is the laboratory experimental method. The samples used in this research were clove leaves, langusei, and honeycomb. The analyses carried out were organoleptic tests, pH tests, homogeneity tests, and protection power tests. The lotion, with the addition of clove, langusei, and honey bee hive plant extracts, meets the organoleptic test, test requirements for physical quality and repellent lotion strength, namely having a smooth and soft lotion mass, the color and aroma of the lotion is typical of a mixture of clove, langusei, and honey bee hive plants. The results of the study showed that from variations in the

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concentration of cloveleaf extract, langusei, and honey bee nest (0%, 0.25%, 5%, 10%), the best results were formula IV, namely with a concentration of 10% which had the best physical properties, safe to use and the most effective protective power to repel mosquitoes.

Keywords: Mosquitoes, Repellent, Lotion, Clove Leaves, Langusei, Beehive

INTRODUCTION

Mosquitoes are the main vector insects that cause various tropical diseases, namely diseases that can be transmitted through mosquitoes, including: Dengue Hemorrhagic Fever (DHF) and Malaria. The vectors of dengue fever are the *Aedes aegypti* and *Aedes albopictus* mosquitoes, while the *Anopheles* sp. is a vector for malaria. Apart from these two diseases, mosquitoes can also transmit Filariasis (elephantiasis), Chikungunya and Encephalitis (Islamiyah et al, 2013). High rainfall throughout the year causes lots of puddles to emerge, supported by high population density, resulting in increased mosquito density; rich in sunlight so the average air temperature is 26-30°C which supports mosquito breeding; has a large water area so air humidity tends to be high, causing mosquitoes to survive longer and the spread of disease more widely (Adrianto & Yuwono, 2021). This case has a very fast infection time, and in a short time it can cause death if not treated immediately (Ruminem, et al., 2020).

Data on the DHF morbidity rate in North Sulawesi was recorded at 94.97 per 100,000 population and was the fifth highest in Indonesia with a Case Fatality Rate of 1.13% in 2019 (Indonesian Ministry of Health, 2020). One effort to prevent mosquito bites is to use repellent. DEET changes the psychological response of ORN (olfactory receptor neurons) in mosquito antennae which are sensitive to lactic acid, thus inhibiting the response of the mosquito's olfactory system which responds to chemical signals and making mosquitoes no longer interested in humans (DEET Technical Fact Sheet, n.d.). Currently, the dosage forms that are widely used are anti-mosquito spray, lotion and electric mosquito repellent which contain synthetic chemicals such as DEET or N,N-diethyle-m-toluamide (Suhud et al., 2018).

The clove plant, which is better known as a raw material for cigarettes, has the Latin name *Eugenia aromatica*. Its distinctive aroma makes traditional people use it as a mixture of mosquito repellent (Yanuar & Dinata, 2007). Clove leaves contain ethanol compounds that can repel insects (Boesri et al., 2015). Langusei (*Ficus minahassae*) is a wild plant in Minahasa, North Sulawesi which can be used as a mixture to repel mosquitoes. The *Ficus Minahassae* tree is small, the stem emits white sap, the trunk is often densely covered with long, dense fruit that resembles a hanging beard. Like the function of *Ficus* in general, langusei also has the ability to regularly store water reserves during the dry season (Pitopang et al., 2008). Langusei fruit is also often used as a mixture of traditional medicines and can be used as a repellent to repel insects. *Apis dorsata* beehive which is formulated into wax containing flavonoid compounds and phenolic acids and several other ingredients is also effective as an aromatherapy candle which can repel mosquitoes (Rettob et al., 2021). Honey bee hive extract contains alkaloids, flavonoids, saponins, tannins, steroids and tripenoids based on the results of phytochemical analysis (Mokosuli et al., 2019).

METHOD

This research was carried out for 4 months, at the Biopharmaceutical and Biomolecular Laboratory, Biology Department, Faculty of Mathematics and Natural and Earth Sciences, Manado State University.

The method used in this research is the experimental method. In this research, there was a treatment of giving concentrated extracts of clove leaves, langusei, and honey bee nest to lotion preparations that would be used as anti-mosquito lotion ingredients. The concentration ratio used is 1:1:1 in 100g of lotion. Where concentration I is used: 0% extract, concentration II: 0.8gr clove leaf extract, 0.8gr langusei extract, and 0.8gr honeycomb extract, concentration III: 1.6gr clove leaf extract, 1.6gr langusei extract, and 1.6g honey bee hive extract, concentration IV: 2.5g clove leaf extract, 2.5g langusei extract, and 2.5g mixed into lotion.

The tools and materials used are: Whatman filter paper, glass jar, knife, scissors, blender, metal spoon, plastic packaging clip, Stand Up Zipper, Pot, Erlenmayer phyrex, funnel, label paper, stirring rod, aluminum foil, balance scale, Eppendorf micropipette, orbital shaker, glass

object, glass cover, binocular microscope, rotary evaporator and UV-VIS spectrophotometry, clove leaves (*Syzygium aromaticum* L.), langusei fruit (*Ficus minahassae*), honey bee nest (*Apis dorsata binghami*), triethanolamine (TEA), stearic acid, cetyl alcohol, glycerin, nipagin, nipasol, distilled water, quercetin and 70% alcohol.

Extraction of Clove Leaves (*Syzygium aromaticum*), Langusei (*Ficus minahassae*), and Honey Bee Nest (*Apis dorsata*)

The extraction method used is maceration. The samples used were clove leaves (*Syzygium aromaticum*), Langusei (*Ficus minahassae*), and honey bee nests (*Apis dorsata*) taken from the Minahasa area.

Making clove and langusei leaf extract begins by mixing the sample with 70% alcohol in a ratio of 1:2. Macerate for 3 days while stirring for 1 hour. After 3 days the sample will be filtered using filter paper to obtain a filtrate, and for extracts of clove leaves and langusei using a rotary evaporator. Honey bee hive extract is made using the maceration method using 70% alcohol as a solvent. Fresh honey bee hives are cut into pieces and weighed 250 grams and put into a maceration container. The maceration process lasts for 24 hours and is then filtered using a filter and sterile funnel to separate the filtrate from the dregs. The remaining dregs are macerated again with fresh solvent. The solvent was replaced three times and the maceration filtrate was evaporated at low pressure with a temperature of 60-70°C using an evaporator until a thick extract was obtained (Yuliana et al., 2015).

From the extraction results of clove leaves, langusei fruit and honeycomb using the maceration method, it shows that the extraction results of clove leaves are dark greenish brown, langusei are dark brown, and the extraction results of honeycomb are brown.

Anti-Mosquito Lotion Formulation Made from Clove Plants (*Syzygium aromaticum*), Langusei (*Ficus minahassae*), and *Apis dorsata Binghami* Beehive

Anti-mosquito lotion is made according to the lotion formula with a total mass of 100 grams as follows: stearic acid, cetyl alcohol, nipasol and glycerin are put into an evaporating cup, melted in a water bath and stirred until homogeneous. Put TEA into a beaker glass, add hot water, stir until dissolved. Nipagin is dissolved in boiling water, stirred until dissolved. The melted product is put into a hot mortar, TEA is added, stirred until it forms an emulsion corpus. Nipagin was added little by little, stirred until homogeneous (Ulaen et al., 2012). The mixed ingredients are then divided into 4 equal parts. Extract clove leaves, langusei and honey bee nest in a ratio of 1:1:1 then add them to each preparation to form 4 lotions with concentrations of 0%, 2.5%, 5% and 10%. Then the preparation that had been mixed with the extract was added with distilled water until each volume reached 100 mL.

Table 1. Comparison of Extract Concentrations

Concentration	Comparison		
	Clove leaves	Langusei	Beehive
0%	-	-	-
2,5%	1	1	1
5%	1	1	1
10%	1	1	1

Lotion Quality Test

Organoleptic Test: Organoleptic examination of the lotion as an anti-mosquito repellent, clove leaf extract, langusei and honey bee nest was carried out by observing the texture, color and aroma sensorily (Ambari & Suena, 2019). The test was carried out by distributing questionnaires to 15 panelists to objectively assess the lotion preparation. Before testing the 15 panelists, all panelists must be equalized first, with the steps being that all panelists must wash their hands up to the upper arms with soap and running water.

pH test: pH testing is carried out using a universal pH stick. Repellent lotion is applied to the universal pH stick and then the results are compared with the color standard on the universal pH stick packaging (Ambari & Suena, 2019).

Homogeneity Test: Homogeneity testing of lotion is carried out by taking a small sample of the lotion formula preparation, then placing a small amount of lotion between two glass objects. Observed the arrangement of coarse particles or inhomogeneity (Mardikasari et al, 2016).

Mosquito Bite Protection Power Test

The test was carried out in a room measuring 1.5 m x 1.5 m x 2 m in dark conditions. In the room there were approximately 30 mosquitoes that had been bred in a place that had not sucked blood at all. Then, before being tested on mosquitoes, volunteers had to wash their hands up to the upper arms using soap and running water. Then the volunteer whose arms had been rubbed with lotion entered the testing area for 10 minutes. The protective power against mosquito disturbances can be determined using the formula:

$$Dp = \frac{(K - P)}{K} \times 100\%$$

Information:

DP : Protection Power.

K : Number perch with control arm lotion does not contain clove leaf extract, langusei, and honey bee hive.

P : The figure rests on the arm and is smeared with lotion containing clove leaf extract, langusei and honey bee hive.

RESULTS AND DISCUSSIONS

1. Extraction of Clove Leaves (*Syzygium aromaticum*), Langusei (*Ficus minahassae*), Honey Beehive (*Apis dorsata*)

From the extraction results of clove leaves, langusei seeds and honeycomb using the maceration method, it shows that the results of clove leaf extraction are dark greenish brown, the results of langusei extraction are dark brown, and the results of honeycomb extraction are brown.

Table 2. Yield of Extraction Results of Clove Leaves (*Syzygium aromaticum*), Langusei (*Ficus minahassae*), and Beehive (*Apis dorsata*)

Sample	Solvent Volume	Sample Weight	Extract Weight	Extract Yield
Clove leaves	1000ml	250g	11,5g	4,6%
Langusei	1000ml	250g	7,5g	3%
Honey bee hive	1000ml	250g	11,4g	4,56%

2. Organoleptic Test

The results of organoleptic test observations for 3 weeks in Formulation I produced a white color on a base that did not contain clove leaf extract, langusei and honey bee nest. Meanwhile, Formulation II produces a light brown color. Formulation III produces a light brown color which tends to be darker than Formulation II. In Formulation IV the dark brown color is more intense than in Formulation II and Formulation III. In terms of dosage form, all formulas (Formulations I, II, III and IV) are in the form of good lotions. Formulation I produces no odor and no aroma, while Formulations II, III and IV produce a distinctive odor with a combination of extracts. An assessment is carried out on a scale of 1-5 (1 really doesn't like it, 2 doesn't like it, 3 neutral, 4 likes it, 5 really likes it) regarding the aroma, color and texture and also the reaction during 10 minutes of use.

Table 3. Organoleptic Observation Results

Organoleptic	Formulation I	Formulation II	Formulation III	Formulation IV
Color	White	Light brown	Dark light brown	Dark brown
Dosage Form	Mass Lotion	Mass Lotion	Mass Lotion	Mass Lotion
Smell	No Scent	Extract Combination	Extract Combination	Extract Combination

		Aroma	Aroma	Aroma
Texture	Soft	Soft	Soft	Soft

Table 4. Organoleptic Test Results

Panelist Code	Aroma	Color	Texture	Reaction after 10 minutes
1	2	3	2	2
2	2	2	2	2
3	4	4	2	2
4	2	3	2	1
5	2	2	2	1
6	2	2	3	2
7	3	4	3	3
8	2	3	2	2
9	4	1	2	2
10	3	1	2	3
11	3	2	3	2
12	2	3	2	2
13	2	2	2	1
14	2	3	2	2
Total	35	35	31	27
Average	2,5	2,5	2,2	1,9

Information:

- 1 = Very like it
- 2 = Like
- 3 = Neutral
- 4 = Don't like it
- 5 = Very dislike it

3. pH Test

Test the pH of each lotion concentration using universal pH paper. From data based on the pH paper that has been used in the lotion, it is known that the lotion meets the requirements because it shows pH = 6-7 on the universal pH test litmus paper. Lotion with a pH value that is too acidic can irritate the skin, whereas if the pH is too alkaline it can irritate and dry the skin.

Table 5. pH test results

Lotion with extract concentration	pH value
Formulation I (0 %)	7
Formulation II (2.5%)	6
Formulation III (5%)	6
Formulation IV (10%)	6

4. Homogeneity Test

The results of the homogeneity test for 3 weeks showed that the four formulas were homogeneous and stable at room temperature from the first week to the third week. This is indicated by the absence of coarse particles on the glass object used during testing and no separation between the lotion base and clove leaf extract, langusei and honey bee hive. The aim of the homogeneity test is so that the active ingredients contained in the lotion preparation can be distributed evenly and do not irritate the skin when the lotion is used.

Table 6. Homogeneity Test Results for Lotion Preparations

Replication	Room Temperature Storage (25°C - 30°C)		
	Week 1	Week 2	Week 3

Formulation I	Homogeneous	Homogeneous	Homogeneous
Formulation II	Homogeneous	Homogeneous	Homogeneous
Formulation III	Homogeneous	Homogeneous	Homogeneous
Formulation IV	Homogeneous	Homogeneous	Homogeneous

5. Protection Power Test

Testing of the protective power with the lotion formula of clove leaves, langusei and honey bee nest was carried out by applying the lotion with the specified concentration to the human arm and then placing it in a testing room containing 30 mosquitoes for 5-10 minutes. The results obtained from testing the protective power of the lotions showed that there were differences in the protective power of the four lotions with extract concentrations of 0%, 2.5%, 5% and 10%. Autan branded commercial lotion was used as a positive control to see and calculate the difference in protective power of the four lotion formulations. From the results above, it can be seen that the lotion formula with a concentration of 0% is a negative control in the test because the lotion does not contain clove leaf extract, langusei and honey bee nest. Meanwhile, the lotion formula with an extract concentration of 2.5% has an average protective power of 0.33%. The lotion formula with an extract concentration of 5% has an average protective power of 0.55% and the lotion formula with an extract concentration of 10% has the highest protective power, namely 1%. From these results it can be seen that the higher the concentration of the extract used in the lotion, the higher the protective power provided against mosquito bites.

Table 7. Protection Power Test Results

Concentration	Experiment 1	Experiment 2	Experiment 3	Average
Autan	100%	100%	100%	100%
0%	0,25%	0,16%	0,25%	0,22%
2,5%	16,64%	13,78%	20,80%	17,07%
5%	46,91 %	42,95%	31,90%	40,58%
10%	78,96%	83,97%	88,97%	83,96%

To determine whether there is a significant difference between the protective power of the lotion for each extract concentration, a test was carried out using one-way analysis of variance (ANOVA) with $\alpha = 0.05$ as in Table 8.

Table 8. ANOVA Test Significance Data

	Sum of Squares	df	Mean Squares	F	Sig.
Between Groups	10250.250	3	3416.750	108468	<.001
Within Groups	252.000	8	31.500		
Total	10502.250	11			

It can be noted that the results in the sig section of table 8 anova $P < 0.05$ means H_0 is rejected and H_1 is accepted. So statistically there is a real difference between lotion formulas with 0%, 2.5%, 5% and 10% extracts on the protective power of mosquito stings. Because there are differences, a further Duncan Multiple Range Test will be carried out (Table 9).

Table 9. DMRT Advanced Test Results

	Experiment	N	Subset for alpha = 0.05			
			F1	F2	F3	F4
Duncan ^a	Experiment 1	3	10.67			
	Experiment 2	3		21.67		
	Experiment 3	3			43.67	
	Experiment 4	3				87.00
	Sig.		1.000	1.000	1.000	1.000

The results of the DMRT test showed that the protective power of the four extracts 0%, 2.5%, 5% and 10% had significant differences. The 0% concentration is significantly different from the 2.5%, 5% and 10% concentrations. The protective power of a 2.5% concentration is significantly different from a concentration of 0%, 5% and 10%. The protective power of a 5% concentration is significantly different from a concentration of 0%, 2.5% and 10%. And the protective power of a 10% concentration is significantly different from a concentration of 0%, 2.5% and 5%.

CONCLUSIONS

Clove, langusei and honey bee hive extracts when formulated into lotion can be an anti-mosquito repellent. Lotion with IV formulation extract is the most effective formula in repelling mosquitoes. The lotion preparation with the addition of clove, langusei and honey bee hive plant extracts meets the organoleptic test, test requirements for physical quality and strength of repellent lotion, namely having a smooth and soft lotion mass, the color and aroma of the lotion is typical of a mixture of clove, langusei and bee hive plants. honey.

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