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Formulation and Effectiveness of Coconut Shell Potassium-Based *Piper betle* L Essential Oil Soap Repellent Against *Aedes aegypti*

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ABSTRACT: Serious concerns include mosquito bites and the risk of mosquito-borne diseases. The search for safe, natural substitutes for chemical insecticides is motivated by the effectiveness of skin repellents as a preventative measure. Essential oils from the betel plant (*Piper betle*) produce a unique smell that mosquitoes find repulsive. A useful invention for avoiding bites during everyday activities or bathing is to formulate a repellent as a soap. The purpose of this study was to assess the mosquito-repelling effectiveness of a betel-based soap formulation using the human-arm-bait method. Five dosage levels and four observation intervals (30 minutes, 1 hour, 2 hours, and 3 hours) were used in the randomized design experiment, with six replicates for each condition. *Two-Way ANOVA*, *Tukey HSD* post hoc tests, and Probit regression were used to analyze data on the number of mosquitoes landing to calculate *Lethal Time* (LT₅₀ and LT₉₀) values. Mosquito landing intensity was significantly influenced ($p < 0.001$) by dosage, observation time, and their interaction, according to *two-way ANOVA* results, with an R-squared value of 94.2%. A consistent and significant decrease in repellent efficacy over time ($p < 0.05$) was confirmed by *Tukey HSD* post-hoc tests; the average number of mosquito landings increased from 9.12 (at 30 minutes) to 17.56 (at 3 hours). With a mean difference of 4.00 landings, the most significant decline in repellent efficacy happened between the 2- and 3-hour intervals. The formulation's LT₅₀ was estimated by Probit modeling to be 1.257 hours (roughly 1 hour and 15 minutes), while the theoretical LT₉₀ was determined to be 19.342 hours. In summary, the tested repellent formulation provides robust protection, but its effectiveness diminishes markedly with prolonged skin exposure

1. INTRODUCTION

The main mosquito that transmits dengue hemorrhagic fever (DHF) in Indonesia is *Aedes aegypti* (Soegijanto, 2004). The first line of defense has been chemical control, particularly the widespread use of insecticides; however, persistent use has led to the emergence of resistant vector populations (Aguirre-Obando et al., 2015). Additionally, it is advisable to take preventive measures to protect oneself from bites, such as removing or minimizing breeding grounds, using bed nets, and applying insect repellents (Uc-Puc et al., 2016).

Using repellents that are intended to reduce human-vector contact is one method of controlling *A. aegypti* (Galaviz et al., 2021). In various forms, including sprays, vaporizers, creams, gels, lotions, wristbands, and soaps, synthetic chemical repellents—most notably those containing DEET (N, N-diethyl-3-methylbenzamide)—can reduce mosquito bites by 30% to 80%, depending on concentration (Galaviz et al., 2021). They may, however, negatively impact the environment and the skin. This issue has led to studies on mosquito repellents made from natural sources. There are a lot of natural repellents; about 344 plant species have compounds that have insecticidal qualities and work to keep adult mosquitoes away (Galaviz et al., 2021). Citronella grass, betel, clove, zodia, and lavender are among the plants that are commonly used as natural insecticides (Zulaikha et al., 2019). Plants

that contain essential oils can be used to make natural insecticides because these oils have unique scents that mosquitoes dislike (Mufidah et al., 2021). *Piper betle* (betel leaf) is one such plant. Saponins, flavonoids, polyphenols, triterpenoids, essential oils (such as chavicol, chavibetol, carvacrol, eugenol, and estragole), sesquiterpenes, sugars, and starch are among the secondary metabolites found in *P. betle*, which has many health advantages (ENI, 2020),(Widiyastuti et al., 2016). Studies have shown that essential oils extracted from its leaves make *P. betle* has been shown in earlier studies to be an effective mosquito repellent (Gunawan & Kurniaty, 2021),(Handayani, 2013). Insecticidal properties are thought to be present in essential oil compounds. Adding them to soap is one way to use them. On the market, natural-based bath soaps are still uncommon (Khuzaimah, 2013).

One kind of insect repellent product is soap. Chemicals like KOH and NaOH are commonly used as alkalis in the soap-making process (Nurama et al., 2014); however, using natural alkali sources, such as plant-derived potassium, is safer for the skin. Coconut shells can be used to produce natural alkali (Sucipto et al., 2011). The essential oil of *Piper betle* (betel leaf) can be used to make soap. Fatty acids from vegetable oils are mixed with sodium or potassium compounds to create soap, which can be solid, soft, or liquid and acts as a cleansing agent by creating a lather. It is anticipated that using *P. betle* soap for bathing will leave the essential oil's scent on the body, keeping mosquitoes away. *P. betle*'s leaf essential oil is a good option for creating a repellent soap because of its considerable capacity to both kill and deter mosquitoes. Additionally, evaluating *P. betle*'s potential as a repellent requires evaluating the soap's protective effectiveness. In order to create safe repellent products, research using natural ingredients and waste products like coconut shells is beneficial.

This study stands out because no prior research has developed a mosquito-repellent soap by combining *P. betle* with alkali derived from coconut shells (Adjei et al., 2022). Neem extract and alkali from banana stems have been used in earlier research; in one such study, 47.5% of 40 respondents who used the soap formulation were protected from mosquito bites for four hours. Other studies have shown that essential oils can either kill adult mosquitoes or repel mosquito larvae. Furthermore, research has demonstrated the effectiveness of soaps—liquid or solid—infused with chemical or botanical extracts as mosquito repellents.

As the primary agent in saponification, which turns oils or fats into soap, caustic soda (NaOH) is an essential component in the manufacture of bath soap. The chemical reaction necessary to produce soap would not occur in the absence of NaOH (Maripa, 2016). The NaOH dissociates into its constituent elements once the soap is formed. NaOH is an alkali used in the production of soap that is highly soluble in water and 96% ethanol. It is described as a rod-shaped, granular, crystal mass or flakes that are dry, hard, brittle, and exhibit a white crystal structure. It is also highly alkaline, corrosive, and effective as an additive (Depkes RI, 2013).

Research on *P. betle* as a larvicide is becoming more widespread in Indonesia. *Ae. aegypti* mosquitoes can be killed by the essential oil, phenyl acetate, chavicol, and phenol content of *P. betle*, according to a study by **Made Oka et al. (2011)**, with an LC50 of 309.03 ppm (Parwata et al., 2011). With an LC50 of 1422.81 ppm, **Handayani et al. (2013)** also found that higher *P. betle* concentrations led to higher mosquito mortality rates (Handayani, 2013). *P. betle*'s essential oil has the power to harm mosquito eggs and prevent insect development (Rahmawati et al., 2020). A portion of the coconut (*Cocos nucifera* L.) that has not been fully utilized is called the coconut husk. Usually, households only extract the coconut milk, leaving the husk to be thrown away or occasionally used as fuel. Potassium (K) ions and carbonate (CO₃) compounds are found in high concentrations in the ash produced by burning coconut husks (47% and 27.7%, respectively). Since potassium makes up about 40% of the husk ash, coconuts have a lot of potential as a cheap source of caustic potassium through the extraction of potassium salts from their husks. Thus, by first burning them into ash, coconut husks can be used to make alkali (Muhammad Yusuf Ritonga et al., 2013). Researchers at Virginia Tech found in a May 2023 study that mosquitoes dislike the smell of certain soaps. Mosquitoes can be repelled and bites avoided by using soap with an unpleasant scent (Puspapertiwi et al., 2023).

The purpose of this study is to assess the effectiveness of repelling mosquito bites from *Ae. aegypti* mosquitoes. A study involving body wash formulations containing *P. betle* essential oil at different concentrations (0.75, 1, 1.25, and 1.75 ml) was conducted on *Ae. aegypti*. Zodia liquid soap showed a protection rate of 90.93% at the one-hour mark for the 1.0% concentration and 93.18% for the 1.5% concentration, according to research by Simaremare et al. (2018), which employed neem leaf essential oil soap as an *A. aegypti* repellent (Simaremare et al., 2018).

The study assessed user acceptance of scent and color as well as protection durations of 30 minutes and 1, 2, and 3 hours. All things considered, this creative research on developing a natural insecticide from *P. betle* essential oil to prevent mosquito bites

is essential for reducing the prevalence of dengue fever. This disease frequently reaches outbreak status when readily accessible botanical resources are used, and chemicals harmful to the environment and human health are avoided.

2. MATERIALS AND METHODS

This study employed a quasi experimental design specifically a Posttest-Only Control Design utilizing a Completely Randomized Design (CRD). The research was conducted at the Integrated Laboratory of Poltekkes Kemenkes Aceh.

Tools and Materials: Tools included a steam distillation apparatus, a pestle and mortar, an analytical balance, laboratory glassware, a mixer, a water bath, pH paper, a centrifuge, a rotary evaporator, a thermometer, a hot plate, vials, a refrigerator, an oven, mosquito cages, and soap molds. Materials included betel leaves (*P. betle*), *Ae. aegypti* mosquitoes, toluene, ethyl acetate, TLC plates, sulfuric acid (H₂SO₄), anhydrous sodium sulfate (Na₂SO₄), methanol, coconut oil, palm oil, sodium hydroxide (NaOH), pH paper, ice cubes, sample containers, and distilled water.

Collection and Preparation of Research Materials: *P. betle* from Indonesia's Aceh Besar Regency served as the study's raw material. The test subjects were IPB-bred *Ae. aegypti* mosquitoes. Essential oil was produced and characterized at the UII Laboratory. In accordance with the standard protocol of 25–30 larvae per treatment group, 25 larvae were used per treatment (WHO, 2005). In compliance with (Supranto, 2000), five replicates were conducted.

2.1. Alkali Preparation

The shells of coconuts are burned until they become a grayish-white ash, which is then sieved through an 80-mesh screen. The fine ash is mixed with distilled water (at a 1:3 ratio) in an HDPE plastic container, stirred until homogeneous, and left to stand for 72 hours to allow solid particles to settle and to maximize the extraction of potassium compounds (K_2CO_3 / KOH). The supernatant (the transparent upper layer) is then separated from the ash sludge sediment by decantation, which completes filtration and purification. The decanted filtrate is filtered again through filter paper (or layered filter cloth) to obtain a clear, natural alkali solution free of microparticles (Ramadhan & Sukeksi, 2018).

2.2. Soap Preparation

The equipment used includes a scale, thermometer, basin, spoon, blender, knife, graduated cylinder, hand mixer, soap mold, and pH indicator. The materials used in this study include distilled water, betel leaf essential oil, coconut oil, palm oil, olive oil, an alkali derived from coconut shell ash, and NaCl.

Olive oil, coconut oil, and palm oil should be dissolved and heated to 700°C in a *stainless steel* pot to prepare the mixture. Gradually pour the coconut ash alkali solution into the oils while stirring. The mixture will appear hazy and milky white at this point, signifying the beginning of emulsification. The mixture should then be heated in the pot over low to medium heat to begin the hot saponification process. Add NaCl (salt) as the mixture progressively thickens to a porridge-like consistency, stirring occasionally to prevent the oil and lye solution from separating, until it finally becomes a transparent gel or paste (Sukawaty et al., 2016). Essential oil and solid soap formulation composition (Nurcahyati & Herliningsih, 2019).

Table 1. Of essential oil and solid soap formulation composition

Materials	Formula (%)				
	0 (negative control)	I	II	III	IV
<i>P. betle</i> essential oil (ml)	0	0,75	1	1,25	1,75
Coconut oil (ml)	30	30	30	30	30
Palm oil (ml)	25	25	25	25	25
Olive oil	10	10	10	10	10
Coconut shell alkali (gr)	8,9	8,9	8,9	8,9	8,9
Aquadest (ml)	100	100	100	100	100
NaCL (gr)	60	60	60	60	60

2.3. Research Procedure

Twenty-five female *A. aegypti* mosquitoes, raised in a lab setting and without food, were divided into five different cages for the experiment. *P. betle* essential oil was used in the treatments at volumes of 0.75 ml, 1 ml, 1.25 ml, and 1.75 ml, as well as a negative control that contained no essential oil. Acknowledging that there was no risk of disease transmission from mosquito bites, five male volunteers willingly participated. Before the test, volunteers cleaned their arms under running water and dried them with a fresh towel. They then wetted the arm, applied the recommended amount of soap, left it on for a minute, rinsed it under running water, and dried it with paper towels. After this preparation, the volunteers put their right arm into the cage for fifteen minutes while wearing gloves to protect their hands. In order to assess the protective efficacy, the arm was then taken out and put back in after 30 minutes, as well as at the first, second, and third hours, with a 15-minute observation period at each interval. The efficacy of the essential oil was tested in a mosquito cage measuring 10 x 10 x 10 cm with glass walls and a nylon mesh top.

2.4. Data Analysis

In order to assess the *P. betle* essential oil soap formulation, observations were made over two weeks, specifically on days 1, 7, and 14, in order to ascertain whether any variations in outcomes occurred during storage.

2.4.1. GC-MS Test

Gas Chromatography-Mass Spectrometry (GC-MS) using a Shimadzu GCMS-QP2010S system was used for compound analysis (specifications: Rastek Rxi 5MS column, 30 m column length, helium as the carrier gas, and EI ionization at 70 eV).

2.4.2. Organoleptic Test

The organoleptic test is used to assess the preparation's physical stability by examining its shape, color, and odor.

2.4.3. pH Test

One gram of soap is dissolved in 10 mL of distilled water for the pH test, and the pH is then measured with universal indicator paper. The outcome must be between 8 and 11, the typical pH range for soap.

2.4.4. Foam Stability Test

The purpose of the foam height test, which uses distilled water, is to gauge the foam's stability. The height of the foam in a test tube is measured to perform this test. A test tube filled with 10 mL of distilled water and a 1 g sample of solid soap is shaken vigorously and steadily for 60 seconds. The height of the resulting foam is then measured. After letting the mixture stand for half an hour, the foam height is measured once more and compared to the 1.3–22 cm standard set by the Indonesian National Standard (SNI) (Nurcahyati & Herliningsih, 2019). The following formula is used to calculate foam stability:

$$\text{Foam Stability} = 100\% - \% \text{ Vanishing Foam}$$
$$\% \text{ Vanishing Foam} = \frac{\text{Initial foam height} - \text{Final foam height}}{\text{Initial foam height}} \times 100\%$$

Irritation Test

The skin may experience various reactions (side effects) when using bar soap. A skin irritation test was performed on volunteers to determine whether such side effects were present. Six volunteers between the ages of 18 and 25 participated in the experiment. After applying the soap formulation to the volunteers' skin behind their ears for a full day, any changes, including redness, itching, and skin roughness, were noted.

2.4.5. Protection Test as a Repell

The following formula can be used to determine protection against mosquito interference:

$$DP = \frac{K - P}{K} \times 100\%$$

Description:

Dp : Protection efficacy

K : Landing rate on the control arm (containing no *P. betle* essential oil)

P : The number of landings on the arm treated with betel leaf essential oil.

Data obtained from the *simplex lattice design* calculations (predictions) and the actual test results were analyzed using one-way *Analysis of Variance (ANOVA)* to determine whether there were significant differences between treatments. The analysis moved on to the *Least Significant Difference (LSD)* test if the ANOVA result was significant ($p < \alpha$).

3. RESULTS AND DISCUSSION

The first step in ensuring the product's safety and quality is to test the soap's physical properties. According to the test results, the addition of betel leaf extract affects the soap's organoleptic qualities while preserving the physical attributes that meet the requirements for solid soap, as indicated in the table.

Table 2. Evaluation results for *P. betle* soap

Testing	Characteristic	0,75ml	1 ml	1,25 ml	1,75 ml	Control (-)
Organoleptic	Form	solid	solid	solid	solid	solid
	Odor	Distinctive scent of betel leaf	Distinctive scent of betel leaf	Distinctive scent of betel leaf	Distinctive scent of betel leaf	No scent of betel leaf
	Color	Pink	Pink	Pink	Pink	White
Foaming Ability	Foam height in test tube (cm)	8	7	5	5	7
Irritation Test	Irritation present	No	No	No	No	No
PH	Ph (4 weeks)	8	8	8	8	

Table 3 presents the GC-MS results for the non-polar fraction extract, including chromatographic information and identified active compounds.

Table 3. GC-MS analysis results of the non-polar fraction (n-hexane) of *P. betle* essential oil.

R Time	Compound Name	Area %
3.642	(1S)-Sabinene	15,26
3.859	Sabinene	5,00
5.362	l-Limonene	2,85
17.697	2-Methyl-2,4-pentandiol	2,73
24.001	Undecane, 5-phenyl-	4,61
24.399	Undecane, 4-phenyl-	3,34
25.106	Benzene, (1-ethylnonyl)-	2,92
26.491	6-Phenyldodecane	3,55
26.700	Benzene, (1-butyloctyl)-	3,32
26.757	Chavicyl Acetate	7,43
27.142	4-Phenyldodecane	2,58
29.072	Tridecane, 6-phenyl-	3,00
32.596	Eugenol	4,70
34.255	Phenol, methoxy-4-(2-propenyl)-, acetate	3,52
-	Other minor compounds (26 compounds < 2,5%)	33,20

GC-MS analysis identified 14 active compounds, with the five highest-percentage compounds being (1S)-sabinene, chavicol acetate, sabinene, eugenol, and 5-phenylundecane. Sabinene, chavicol acetate, eugenol, and undecane are volatile organic compounds (essential oils) typically derived from aromatic plant extracts.

The findings about the soap's ability to prevent mosquito landings and the recorded rates of mosquito mortality are shown below:

Table 4. Percentage protective efficacy of *P. betle* soap against *Ae. aegypti*

Soap dosage variations	Percentage of Protection Power (%)			
	30 minutes	1 hour	2 hours	3 hours
Control -	0%	0%	0%	0%
0,75 ml	67,89%	37,39%	28,70%	7,27%
1 ml	70,64%	51,30%	36,11%	15,45%
1,25 ml	76,15%	66,09%	49,07%	24,55%
1,75 ml	91,74%	87,83%	72,22%	53,64%

A *two-way ANOVA* statistical test was performed to ascertain whether changes in dose concentration, observation time, and the interaction between the two had a significant impact on the quantity of mosquitoes landing on the human arm bait. Table 5 displays the test results.

Table 5. *Two-Way ANOVA* Test Results for the Number of Mosquitoes Landing

Source Variation (Source)	Sum Squares	Degrees of Freedom (df)	Mean Square	F-value	Significance (Sig.)
Corrected Model	4181,16	19	220,061	68,555	<0,001
Intercept	16952,04	1	16952,04	5281,01	<0,001
Observation Time	937,64	3	312,547	97,367	<0,001
Dosage	2823,46	4	705,865	219,896	<0,001
Time * Dosage	420,06	12	35,005	10,905	<0,001
Error	256,8	80	3,21		
Total	21390	100			
Corrected Total	4437,96	99			

R Squared = 0,942 (Adjusted R Squared = 0,928)

According to Table 5, the Observation Time factor has a value of *Sig.* < 0.001 ($p < 0.05$), suggesting that the length of observation has a significant impact on the number of mosquitoes that land. The Time $\times$ Dosage interaction is a value of *Sig.* < 0.001 ($p < 0.05$), and the Dosage factor displays a value of *Sig.* < 0.001 ($p < 0.05$), indicating that variations in repellent dosage concentrations significantly affect the number of landing mosquitoes. This indicates that the interaction between dosage level and observation duration significantly influences the intensity of mosquito landings on the arm.

A very robust test model is indicated by the *R-squared value* of 0.942, which shows that 94.2% of the variation in the number of landing mosquitoes is explained by dosage, time, and their interaction, with the remaining 5.8% explained by factors not included in the study. To specifically investigate variations in the mean number of landing mosquitoes across time intervals, post hoc tests using *Tukey's HSD* and *Duncan's* methods were conducted. Table 6 provides an overview of these post-hoc test results.

Table 6. Results of *Tukey HSD* and *Duncan* Post-hoc Tests for the Observation Time Factor

Observation Time	N	Average Number of Mosquitoes Landing $\pm$ SD	Subset 1	Subset 2	Subset 3	Subset 4	Letter Notation
30 Minutes	25	9,12 $\pm$ 0,51	9,12				a
1 hour	25	11,84 $\pm$ 0,51		11,84			b
2 hours	25	13,56 $\pm$ 0,51			13,56		c
3 hours	25	17,56 $\pm$ 0,51				17,56	d
<i>Signifikansi (Sig.)</i>			1	1	1	1	

Description: Number followed by different letters in the notation column indicate significant differences at the alpha significance level = 0,05.

Table 6 results demonstrate that the four observation time groups can be easily divided into four different subsets (Subset 1 through Subset 4). At the 30-minute mark, there were 9.12 mosquitoes, which was the lowest mean number of landing mosquitoes (notation a). This number rose dramatically to 11.84 mosquitoes at the one-hour mark (notation b) and 13.56 mosquitoes at the two-hour mark (notation c), peaking at the three-hour mark with a mean of 17.56 mosquitoes (notation d). This pattern shows that the effectiveness of the repellent steadily decreased with each hour. Table 7 shows a visualization of the mean differences based on pairwise testing to determine the rate of decline in repellent efficacy between observation phases.

Table 7. Pairwise Comparison of Mean Differences in the Number of Landing Mosquitoes

(I) Time	(J) Time	Mean Difference (I - J)	Std. Error	Significance (Sig.)
30 Minutes	1 Hour	-2,72*	0,507	<0,001
	2 Hours	-4,44*	0,507	<0,001
	3 Hours	-8,44*	0,507	<0,001
1 Hour	2 Hours	-1,72*	0,507	0,006
	3 Hours	-5,72*	0,507	<0,001
2 Hours	3 Hours	-4,00*	0,507	<0,001

* The mean difference is significant at the 0.05 level (Tukey HSD Method).

Table 7 provides more proof of the decreasing effectiveness of repellents. With only a 1.72 ($p = 0.006$) increase in the number of landing mosquitoes, the most gradual decrease in repellent activity happened between the first and second hours. On the other hand, there was a mean difference of 4.00 ($p < 0.001$) between the second and third hours, when the number of landing mosquitoes increased, and the sharpest decline took place. The total number of mosquito bites increased by 8.44 ($p < 0.001$) between the beginning of the test (30 minutes) and its conclusion (3 hours).

The *Lethal Time* 50% (LT_{50}) and 90% (LT_{90}) parameters were determined by modeling the mathematical relationship between exposure duration and repellent failure rate using probit analysis, as indicated in Table 4.4.

Table 8. Estimated LT_{50} and LT_{90} Values for Repellent Durability

Probability Parameter	Time Estimate (Hours)	95% Confidence Interval (Lower Bound)	95% Confidence Interval (Upper Bound)
LT_{50}	1,257	0,55	1,693
LT_{90}	19,342	7,323	1844,159

Note: Uses the heterogeneity factor (a) and base-10 logarithm (b).

According to Table 8, the estimated LT_{50} value is 1.257 hours, or roughly one hour and fifteen minutes, based on the results of probit modeling. This finding suggests that before its efficacy is cut in half, the repellent formulation can keep 50% of the test mosquito population from landing, offering complete protection, for this amount of time. The range of 0.550 to 1.693 hours is the consistent confidence interval for this value.

The theoretical time at which the repellent loses almost all of its protective power, enabling 90% of the mosquitoes to land on the arm once more, is represented by the estimated LT_{90} value of 19.342 hours. Due to variations in recorded mosquito landings during the last hours of laboratory observation before reaching a constant saturation point, the LT_{90} 's extremely large confidence interval (7.323 to 1844.159 hours) shows a high degree of data heterogeneity. Consequently, the LT_{50} parameter (1.257 hours) provides a more valid empirical basis for evaluating the skin repellent's reliability limit

3.4 Discussion

According to organoleptic testing, the soap's physical base did not change when the volume of betel leaf (*Piper betle* L.) essential oil was changed from 0.75 mL to 1.75 mL; the soap maintained its solid form and a consistent pink color. The successful retention of the volatile compounds of the essential oil within the soap matrix after saponification is indicated by the strong, distinctive betel leaf aroma seen in all treatment formulations—in contrast to the negative control. Nevertheless, the foaming capacity decreased linearly as the volume of essential oil increased, resulting in a reduction in foam height from 8 cm to 5 cm. This phenomenon occurs because the essential oil's hydrophobic properties prevent soap micelle formation and reduce the surface tension needed to trap air and produce foam (El-Sakhawy et al., 2026).

In terms of skin safety, the formulation is safe for topical application on human skin since there are no irritation reactions at any concentration. The steady pH of 8 noted over four weeks of storage conforms to the safety requirements for solid soap, which are between 8 and 10. This slightly alkaline pH is very beneficial; in addition to maximizing the effectiveness of skin cleansing, a pH of 8 helps preserve the stability of the phenolic compounds in the essential oil—preventing their quick degradation by acidic environmental factors—and may also extend the retention of active ingredients on the epidermal surface following washing (Tungadi, 2020), (Nurintan & Tuljannah, 2025). The use of alkali from coconut shell extract does not cause skin irritation. By controlling the free alkali content to 0% and utilizing essential oils as active superfatting ingredients, natural soap successfully maintains moisture in the stratum corneum while providing mosquito repellent activity, as demonstrated in research by Vander Giessen et al. (2023) that soap alters the voltaic (odor) profile of human skin. Some synthetic soaps actually attract mosquitoes, while soaps made from certain compounds (such as natural oils) have been shown to be effective in repelling mosquito bites. (VanderGiessen et al., 2023)

Water extraction from coconut shell ash produces potassium-based alkalis, primarily potassium carbonate (K_2CO_3) and potassium hydroxide (KOH), rather than sodium hydroxide. Saponification using natural potassium salts produces a milder soap matrix with higher rinsing power, which reduces alkali residue remaining on the skin surface and minimizes the risk of stratum corneum irritation (Tarun et al., 2014)

The phytochemical profile found by GC-MS analysis of the non-polar (n-hexane) fraction explains the mechanism underlying this soap's repellent effectiveness. According to screening results, the formulation is primarily composed of hydrocarbon monoterpenes, namely (1S)-sabinene (15.26%) and sabinene (5.00%). Important phenylpropanoid markers, including chavicol acetate (7.43%) and eugenol (4.70%), as well as l-limonene (2.85%), are next in line. These substances deter mosquitoes by interfering with their olfactory systems, or receptors (Govindarajan, 2010). Aromatic phenolic compounds like eugenol have a thermally stable structure and can keep mosquitoes away (Hasanudin et al., 2026).

Short-range repellent effects and contact toxicity are significantly influenced by the presence of phenolic compounds and their esters, such as eugenol and chavicol acetate. Eugenol's strong affinity for octopamine receptors in the nervous system of insects is well known. When eugenol interacts with octopamine receptors, a mild neurotoxic reaction is triggered, causing mosquitoes to flee as soon as they land on the skin (Aneesha et al., 2026). The findings of this study are in line with Aditama et al. (2021), which showed that plant extracts like citronella (*Cymbopogon nardus*) are effective because they contain active phytochemical compounds that could act as botanical insecticides to repel and kill *Aedes aegypti* mosquitoes (Aditama et al., 2021).

Essential oils that contain highly volatile monoterpenes, such as *limonene* and *sabinene*, function as natural *spatial repellents* by interfering with the mosquito's sense of smell. The following steps are involved in their *odorant blockers*: volatile molecules are

released. *Sabinene* and *limonene* evaporate more quickly when body heat is applied to the skin after the essential oil-infused soap is applied (Nematollahi et al., 2021). As a result, chemical vapor molecules are released into the air surrounding the treated area (Afify & Potter, 2020).

The chemical vapor molecules interact with the olfactory receptors of mosquitoes. On their antennae, mosquitoes have sensory structures called sensilla that are fitted with a specialized protein known as *Orco* (*odorant receptor co-receptor*). The vaporized substances function as inhibitors; molecules of *sabinene* and *limonene* attach to or obstruct the *Orco* proteins on the sensilla. This makes it more difficult for mosquitoes to detect lactic acid and the smell of human sweat, two cues that usually lead them to bite (Joshua I. Raji, Nadia Melo, John S. Castillo, Sheyla Gonzalez, Valeria Saldana & C. Stensmyr, 2017).

The low number of mosquitoes landing at the 30-minute mark (an average of 9.12) indicates that the combination of sabinene, chavicol acetate, and eugenol has a strong repellent effect upon initial exposure. However, kinetic analysis shows a notable decline in efficacy over time. A *Tukey HSD* post-hoc test confirms a significant increase in mosquito landings at the three-hour mark (reaching 17.56).

The Probit regression model, which produces an estimated LT_{50} of 1.257 hours (roughly one hour and fifteen minutes), directly correlates with this declining pattern. The physicochemical characteristics of the plant's essential oils naturally result in this comparatively brief period of effective protection. Major components such as sabinene and limonene possess high vapor pressure, causing them to evaporate rapidly from the warm surface of human skin. The skin's chemical barrier thins significantly as the concentration of these low-molecular-weight monoterpene compounds decreases by the second hour.

The intrinsic volatility of *P. betle* essential oil continues to be a major limiting factor for long-term protection, even though the solid soap formulation (pH 8) helps retain some heavier compounds (such as eugenol and chavicol acetate) during rinsing. For short-term protection, such as after bathing or during brief outdoor activities, this natural repellent soap is therefore very effective; however, for protection lasting longer than three hours, it must be reapplied or natural polymer fixatives must be added.

The number of mosquito landings is significantly influenced by soap dosage, observation duration, and the interaction between the two, according to *two-way ANOVA* results ($p < 0.001$). With an *Adjusted R-Squared* value of 92.8%, the statistical model is strong and shows that the combination of dosage and duration of use nearly entirely causes variations in the number of mosquitoes landing on volunteers' arms. The decline in the soap's effectiveness over time is evident from post-hoc *Tukey HSD* and *Duncan* tests; a consistent decrease in repellent activity was noted at each hourly interval ($p < 0.05$). With the fewest landings (an average of 9.12 mosquitoes), the 30-minute mark provided the highest level of protection; however, the first and second hours saw a decline in efficacy, and by the third hour, the protective effect had significantly decreased. Between the beginning and the end of the test, the overall number of mosquito landings increased by 8.44 ($p < 0.001$). This pattern of declining repellent effectiveness is consistent with research by Fradin and Day (2002), who observed that plant-based natural repellents usually offer a comparatively brief window of maximum protection right after skin application (Fradin & Day, 2002).

The extremely large *confidence limits* connected with the Probit analysis result for the LT_{90} value (19.342 hours) are also explained by the sharp drop in efficacy seen at the third hour. The computer model uses mathematics to predict the point of total exhaustion, or the point at which the repellent runs out entirely. The quantity of mosquitoes that land during the last hours, however, varies greatly in laboratory settings and has not yet stabilized. Rutledge et al. (1985) have also reported discrepancies between computer modeling and actual data in topical repellent studies, which are caused by the evaporative nature of the active ingredient (Sharpington et al., 2000)(Rutledge et al., 1985).

Therefore, the LT_{50} value (1.257 hours, or roughly 1 hour and 15 minutes) serves as a far more accurate and realistic commercial standard when establishing reliability benchmarks for this repellent soap. This duration is consistent with research on commercial essential oil repellents by Katiyar et al. (2023), which found that the best protective efficacy on human skin usually lasts between one and two hours before active molecules evaporate because of body heat (Ganesan et al., 2019).

4. CONCLUSION

The stable solid formulation demonstrates strong repellent efficacy against *Aedes aegypti* mosquitoes: a repellent soap containing *Piper betle* L. (betel leaf) essential oil, which is skin-safe (pH 8). The three main compounds eugenol, chavicol acetate, and (1S)-sabinene work together to offer the best post-application protection, resulting in an effective duration (LT_{50}) of 1.257 hours.

However, because of the essential oil's inherent volatility, the soap's performance drastically decreases by the third hour as the duration increases.

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AUTHOR CONTRIBUTIONS

Wiwit Aditama: Conceptualization, Methodology, Investigation.

Zulfikar: Investigation, Formal Analysis.

Yuni Nindia : rearing mosquito

Vonna A: Methodology, Investigation.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ETHICS STATEMENT

This study was approved by the Health Research Ethics Committee of Health Polytechnic of Aceh (Poltekkes Kemenkes Aceh) under Reference No. DP.04.03/12.7/ 34612026. Written informed consent was obtained from all volunteers prior to their participation in the mosquito repellent testing..

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