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Formulation and antibacterial evaluation of kombucha bar soap with varying concentrations of patchouli essential oil, *Pogostemon cablin* (Blanco) Benth. (Lamiaceae) against *Staphylococcus aureus* ATCC 25923 (Staphylococcaceae)

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ABSTRAK

Permintaan terhadap agen antimikroba alami dalam produk perawatan pribadi semakin meningkat seiring dengan kebutuhan konsumen akan alternatif yang lebih aman dan berkelanjutan dibandingkan formulasi sintetis. Penelitian ini mengkaji pengembangan sabun batang berbahan dasar kombucha dengan penambahan variasi konsentrasi minyak atsiri nilam (0–3 %) menggunakan Rancangan Acak Lengkap. Evaluasi dilakukan terhadap sifat fisikokimia, meliputi pH, kadar air, tinggi busa, dan stabilitas busa. Aktivitas antibakteri terhadap Staphylococcus aureus ATCC 25923 diuji melalui pengukuran zona hambat, sedangkan senyawa bioaktif diidentifikasi menggunakan Gas Chromatography–Mass Spectrometry (GC–MS). Seluruh formulasi memenuhi persyaratan Standar Nasional Indonesia (SNI). Sabun yang dihasilkan memiliki pH rata-rata 8,0, kadar air di bawah 1 %, tinggi busa di antara 5,80–6,50 cm dan stabilitas busa di antara 77,5–91,5 %. Aktivitas antibakteri tertinggi ditunjukkan oleh zona hambat sebesar 10,67 mm. Analisis GC–MS mengungkapkan bahwa patchouli alcohol *dan asam dodekanoat merupakan senyawa utama, yang diduga berinteraksi secara sinergis sehingga meningkatkan aktivitas antibakteri sekaligus memperkuat stabilitas dan keamanan produk. Kombinasi kombucha dan minyak nilam 3 % menghasilkan formulasi dengan karakteristik pembusaan yang baik, stabilitas yang terjaga, serta potensi yang menjanjikan untuk aplikasi komersial.*

Kata kunci: antibakteri, kombucha, minyak atsiri nilam, sabun padat, Staphylococcus aureus

ABSTRACT

The demand for natural antimicrobial agents in personal care products has grown as consumers seek safer and more sustainable alternatives to synthetic formulations. This study examined the development of kombucha-based bar soap incorporating varying concentrations of patchouli essential oil (0–3 %) using a Completely Randomised Design. Physicochemical properties, including pH, moisture content, foam height, and foam stability, were evaluated. Antibacterial activity against *Staphylococcus aureus* ATCC 25923 was assessed through inhibition zone measurements, while bioactive compounds were identified using Gas Chromatography–Mass Spectrometry (GC–MS). All formulations met the Indonesian National Standard (SNI). The resulting soap has an average pH of 8.0, a water content below 1 %, a foam height of between 5,80–6,50 cm and foam stability of between 77,5–91,5 %. The strongest antibacterial effect was observed with an inhibition zone of 10.67 mm. GC–MS analysis revealed patchouli alcohol and dodecanoic acid as the principal constituents, whose synergistic interaction is presumed to enhance antibacterial activity and product stability. The incorporation of kombucha and 3 % patchouli oil produced a formulation with favourable foaming properties, reliable stability, and promising potential for commercial application.

Keywords: antibacterial, kombucha, patchouli essential oil, bar soap, *Staphylococcus aureus*

INTRODUCTION

Bath soap is a daily necessity for cleanliness and skin health. This increase aligns with efforts to protect oneself against skin infections caused by *Staphylococcus aureus* bacteria using natural products (Lailiyah & Rahayu, 2019). However, the widespread use of synthetic antibacterial agents in commercial soaps carries a risk of triggering bacterial resistance, thereby driving the development of safe and effective natural alternatives in daily skin cleansers.

Commercial soaps generally use various methods to fight skin bacteria, ranging from the addition of synthetic ingredients to various natural, plant-based components such as plant extracts (Anindita et al., 2025) and essential oils (Rahmawati et al., 2025). Among these options, Kombucha has emerged as a potential alternative because this fermented beverage is rich in antioxidants and organic acids that can kill bacteria (Wahdaniar et al., 2023). Although traditionally made from real tea, modern biotechnology classifies any herbal infusion fermented with a Symbiotic Culture of Bacteria and Yeast (SCOBY) as an herbal variation of kombucha. While previous research has shown that a variation of kombucha made from butterfly pea flowers can effectively inhibit *S. aureus* bacteria on the skin , this study focused (Rezaldi et al., 2022), on the use of traditional black tea kombucha. However, because kombucha is water-based, it needs assistance to work optimally in a kombucha bar soap. Therefore, the addition of patchouli essential oil, *Pogostemon cablin* (Blanco) Benth., may be helpful.

Antibacterial activity in soap formulations can be enhanced by adding various essential oils containing natural antibacterial compounds. Examples are *Piper ornatum* and *Piper betle* with confirmed β-caryophyllene oil content exhibiting antibacterial activity against common acne bacteria (Anindita et al., 2025). Similarly, clove oil (*Syzygium aromaticum*) is known to be rich in eugenol, which acts as an antibacterial compound (Rahmawati et al., 2025). Apart from these two oils, patchouli essential oil also contains patchouli alcohol which determines the quality of its aroma and functions as an antibacterial agent (Ulandari et al., 2022). In addition to being proven to significantly inhibit the growth of *S. aureus*, patchouli oil also functions as a natural binder (Ginting et al., 2021). By combining 50% kombucha concentration with various concentrations of patchouli essential oil of 0%, 1%, 2%, and 3%, this study aims to test the potential synergy of these two ingredients in making solid soap that not only meets the physical quality standards of the Indonesian National Standard (SNI) but also optimally inhibits *S. aureus*.

Based on the urgency of developing this natural product, this study focuses on addressing the issue of the best formulation and the level of antibacterial strength of kombucha bar soap and patchouli essential oil in inhibiting *Staphylococcus aureus*. By measuring the inhibition zone diameter based on standards (*Clinical and Laboratory Standards Institute (CLSI)*, 2023), this study is expected to provide accurate scientific data regarding the effectiveness of the formulation. Practically, the results of this study are expected to serve as a reference for the development of environmentally friendly natural-based cosmetics and become an alternative solution for sustainable skin care for the public.

METHODOLOGY

Research location, bacterial culture and tools

The research was conducted at the Multifunction Laboratory of UIN Ar-Raniry, Banda Aceh, from February to April 2026. The kombucha solution was harvested on the

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14th day. Patchouli essential oil was obtained from the ARC-PUIPT Nilam Aceh, Universitas Syiah Kuala. The main content of this essential oil is patchouli alcohol (PA), which belongs to the sesquiterpene compound group and indicates its high quality. The essential oil was produced through the steam distillation method from the dried leaves of *Pogostemon cablin* (Blanco) Benth. *Staphylococcus aureus* ATCC 25923 bacteria samples were obtained from the Faculty of Mathematics and Natural Sciences Laboratory, Syiah Kuala University. Samples were analysed by Gas Chromatography-Mass Spectrometry (GC-MS) at the Biology Laboratory, Faculty of Mathematics and Natural Sciences, State University of Medan.

Experimental design

This research was conducted experimentally in a laboratory using a Completely Randomised Design. In this study, the concentration of kombucha was kept constant at 50% (equivalent to 50 mL) for all soap formulas. The main objective of the study was to determine the effect of varying the concentration of patchouli essential oil, namely: 0% (F1), 1% (F2), 2% (F3), and 3% (F4). The formulation with a concentration of 0% (F1) was specifically intended to see the extent of the inhibitory activity of the soap produced if it only contained kombucha, without the addition of patchouli essential oil. To ensure data accuracy, 4 replication was carried out with the number of duplicates following the standard Federer formula. Before formulating the soap, the required kombucha solution was prepared through a fermentation process of black tea and sugar using a kombucha starter culture in the form of a SCOBY (Symbiotic Culture of Bacteria and Yeast). This fermentation and maintenance process is carried out in a sterile glass container covered with a porous cloth and stored in a fixed position at room temperature without opening the container or mechanical disturbance (vibration/shifting) for 14 days before harvesting.

Research procedure

Kombucha fermentation production

A total of 11 g of black tea (1.1%) and 107 g of sugar (10.7%) were weighed and dissolved in 1000 mL of boiling water maintained at 95–100 °C. The solution was allowed to cool to 40 °C, after which a symbiotic culture of bacteria and yeast (SCOBY) and 100 mL of starter solution (10%) were introduced. Fermentation was conducted in a closed container at room temperature for 14 days, after which the kombucha was harvested (Sanggor et al., 2024).

Bar soap production

An alkaline solution was prepared by dissolving 83 g of NaOH in 195 g of distilled water and cooling it to 25–30°C, while a mixture of oils consisting of 236 g of olive oil, 236 g of coconut oil, and 118 g of palm oil was stirred until homogeneous. The alkaline solution was then mixed into the oils until thickened. After that, the mixture was divided into four groups by adding a constant 50% concentration of kombucha (equivalent to 50 mL) along with varying patchouli essential oils at 0% (F1), 1% (F2), 2% (F3), and 3% (F4). This 50% concentration of kombucha served as an effective concentration, while lower concentrations produced insufficient antibacterial benefits (Nabilah et al., 2026),

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while higher use would interfere with the saponification process. Structurally, the excess organic acids in kombucha at full concentration neutralise the excess NaOH, leaving insufficient alkali to harden the soap, while the intense exothermic heat of the reaction simultaneously burns off the remaining sugars (Qotrunnisa et al., 2025) Therefore, this balanced proportion safely optimises the bioactivity of kombucha without compromising the physical hardness of the soap. Finally, the mixture is poured into silicone molds and left to set for 30 days (Nurhajawarsi, 2023).

Bar soap acidity (pH) test

Acidity (pH) testing was conducted to maintain the acidity level of the formulated soaps while ensuring their safety when used on the skin. The procedure began by thinly slicing 1 gram of sample from each formula and dissolving it in 50 mL of distilled water until completely homogeneous. This dissolution process is crucial to facilitate complete ionization of the soap molecules, thus obtaining accurate and stable measurements. For comparison, a commercial solid bath soap (designated K+) also underwent the same preparation and testing procedures as a control reference. The pH value of each solution was then determined by dipping a calibrated universal pH indicator into the solution and recording stable readings. Based on SNI 3532:2021 (Akbar et al., 2025), the standard and safe pH value for solid bath soaps ranges from 6.0 to 11.0.

Foam characteristics (height and stability)

This test aims to determine the soap's ability to produce foam and maintain its structure during use. The procedure is carried out by dissolving 2 g of soap sample in 10 mL of distilled water, then shaking for 1 minute. The height of the foam that immediately forms is measured using a ruler as the initial height. After the solution is allowed to stand for 5 minutes, the foam height is measured again to obtain the final height. For quality comparison, commercial bar soaps on the market are tested using exactly the same method as positive control (K+). According to SNI 06-3532-1994, a good foam height for bar soap ranges from 1.3 to 22 cm, while the standard foam stability requirement ranges from 73 % to 95 % (Leny *et al*., 2023). The foam stability percentage is calculated using the following formula:

$$\text{Foam stability} = \frac{\text{Final foam height}}{\text{Initial foam height}} \times 100$$

Moisture content test

The water content test aims to determine the percentage of free water content in the kombucha-patchouli solid soap preparation to ensure product compliance with the SNI 3532:2021 quality standard (maximum 23 %), which is important to ensure a solid texture, prevent shrinkage, and extend the product's shelf life from microbial contamination. The test is carried out by weighing an empty cup as the initial weight (B0). A 5 g soap sample is placed into the cup, then weighed to obtain the total initial sample weight (B1). The sample is then dried in an oven at 105 °C for 1 hour to evaporate the water content. After drying, the cup is cooled and reweighed to obtain the dry sample weight (B2). The percentage of water content is calculated using the following formula:

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$$\text{Water level\%} = \frac{B_1 - B_2}{B_1 - B_0} \times 100\%$$

Description:
B_0: Weight of empty cup
B_1: Weight of initial sample
B_2: Weight of dry sample

Antibacterial testing

Bacterial suspension was prepared by taking one loop of *Staphylococcus aureus* culture that had been rejuvenated on *Nutrient Agar* (NA) media, transferring it to a tube containing 10 mL of *Nutrient Broth* (NB), homogenizing it and incubating it for 24 hours at 37°C. Then, 1 mL of *Staphylococcus aureus* culture was added to 9 mL of 0.9% NaCl solution in the test tube. The turbidity was adjusted to the McFarland 0.5 standard (if too turbid, add 0.9% NaCl; if not turbid enough, add bacteria from NB) (Jungjunan et al., 2023). Soap samples were dissolved in distilled water (2 g in 20 mL) to obtain test solutions with varying patchouli oil concentrations (0%, 1%, 2%, and 3%). The *S. aureus* bacterial suspension was swabbed on the MHA media using a sterile cotton swab. A standard sterile blank disc with a diameter of 6 mm (HiMedia) was utilised for the assay. The discs were then immersed in the test solution, a positive control (chloramphenicol), and a negative control (distilled water), then placed on MHA media that had been marked into six sections in one petri dish. After that, the samples were incubated at 37°C for 24 hours. After 24 hours, the clear zone was measured using a vernier calliper (Aulia et al., 2020). The average diameter of the inhibition zone was calculated using the formula :

$$Diameter\ of\ the\ inhibition\ zone : \frac{(Dv - Dc) + (Dh - Dc)}{2}$$

Description:
D_V: Vertical Diameter
D_H: Horizontal Diameter
D_C: Disc Diameter

Test results are interpreted by observing the diameter of the inhibition zone formed. According to the Clinical and Laboratory Standards Institute (CLSI), 2023, the inhibition zones are as follows: Very Strong: > 20 mm. Strong: 10-20 mm. Moderate: 5-10 mm. Weak: < 5 mm. All tests were conducted with a number of replications that met the Federer formula: $(n-1)(t-1) \geq 15$.

Description:
n: number of replications
t: number of treatment groups

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Chemical characteristics of soap (Gas Chromatography-Mass Spectrometry GC-MS)

Gas Chromatography-Mass Spectrometry (GC-MS) analysis was performed to identify the bioactive volatile compounds embedded within the formulated soap matrix. This step is crucial to confirm the retention and stability of key chemical components, particularly patchouli alcohol from the essential oil, after undergoing the alkaline saponification process. Samples were prepared using a solvent extraction method with a 10:1 solvent-to-sample ratio. One gram of sample was weighed and dissolved in 10 mL of n-hexane to extract the target compounds. Injection and Evaporation: The sample extract was vaporised in a high-temperature injector and carried by helium gas. Subsequently, compounds were separated in a glass column based on differences in retention times of each component. Ionisation (MS): The molecules were bombarded with electrons (70 eV) until they broke into ion fragments with characteristic patterns (fingerprints). Finally, detection and identification: Ions were filtered based on mass (m/z).

Data analysis techniques

Data were analysed descriptively and quantitatively by calculating the average values of physical parameters, water content, and inhibition zone diameters for comparison with SNI standards and CLSI criteria. This test was validated using the Federer Formula to ensure statistical accuracy in a Completely Randomised Design (CRD) design. Meanwhile, GC-MS data were analysed by comparing the peak areas of the compounds.

RESULT AND DISCUSSION

Kombucha fermentation results

Fermented kombucha in a 1-liter glass jar (FIGURE 1). This 1-liter liquid has the typical cloudy brown colour of fermented tea drinks, with a thick, white-brown SCOBY layer growing evenly on the surface. During the 14-day fermentation process, this kombucha naturally produces fine carbonation gas bubbles from yeast activity, as well as a distinctive flavour that is predominantly sour, fresh, and slightly bitter due to the formation of organic acids from the breakdown of sugars.

FIGURE 1. Results of kombucha fermentation for 14 days

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Solid bath soap production results

The solid kombucha bath soap (**FIGURE 2**) exhibits a clean, uniform, and solid creamy white physical form after undergoing a 4-week curing process. This drying process aims to optimise air evaporation, resulting in a final soap mass of 677 grams (0.677 kg). This mass is divided into 30 square-shaped soap bars with dimensions of 4 cm long and 1 cm wide. The resulting soap has a smooth surface and a stable edge structure, indicating that the saponification process has taken place completely and maturely.

FIGURE 2. Physical appearance of the bar soaps after curing process for 30 days

Physical characteristics of solid body soap

Physical characteristic testing was conducted to ensure that the kombucha solid body soap with patchouli essential oil met quality and safety standards. Tested parameters included pH, foam height and stability, and water content. In general, all formulas demonstrated stable results and met industry standards. A summary of the test results is presented in **TABLE 1.**

TABEL 1. Physical characteristics test results of kombucha bar soap with patchouli essential oil

Test Parameters	F1 (0 %)	F2 (1 %)	F3 (2 %)	F4 (3 %)	K+	Quality Standards
pH (Average)	8.0	8.0	8.0	8.0	7.0	6.0 – 11.0 (SNI 3532:2021)
Foam Height (cm)	6.50	6.25	5.80	6.10	4.38	1.3 – 22 cm (SNI 06-3532-1994)
Foam Stability (%)	77.5	83.5	87.5	91.5	92.5	73 – 95 % (Akbar et al., 2025)
Water Content (%)	0.30	0.31	0.33	0.30	0.26	≤ 23 % (SNI 3532:2021)

Analysis of soap acidity (pH)

Based on the test results (**TABLE 1**), kombucha solid body wash with various concentrations of patchouli essential oil had an average pH value of 8.0. This value indicates that all formulations meet the SNI 3532:2021 standard, which stipulates a safe pH range between 6.0 and 11.0. If the soap's pH is too high, the excessive alkaline properties will damage the acid mantle, which functions as the skin's natural protective layer, causing moisture loss, excessive skin dryness, and increasing the risk of pathogen infection due to a weakened skin defense system (Akbar et al., 2025). Conversely, if the soap's pH is too low, the formulation can cause skin irritation (Sari et al., 2024).

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Analysis of foam height and stability

Foam formation is an important parameter for consumer comfort and soap's cleansing power. The test results in **TABLE 1** show that all kombucha patchouli soap formulas (F1–F4) produced an initial foam height between 5.80 cm and 6.50 cm. This value meets the SNI 06-3532-1994 standard (safe range 1.3–22 cm), making it safe and less likely to cause skin irritation due to excessive foam formation (Akbar et al., 2025). Compared with commercial soap (K+), which only produced a foam height of 4.38 cm, kombucha patchouli soap had a significantly higher initial foam height. This is due to the use of coconut oil in the kombucha soap formula. Coconut oil is rich in lauric and myristic acids, which naturally act as excellent foaming agents (Oktiani et al., 2025). Commercial soaps (K+) produce lower foam because the manufacturer's formulation limits the foaming agent to maintain gentleness on sensitive skin.

For foam stability, the formulated soaps ranged from 77.5% to 91.5%, meeting the 73–95% standard (Leny et al., 2023), commercial soap (K+) produces a much more stable foam because it contains synthetic surfactants, which are artificial foam stabilisers. Kombucha soap, on the other hand, produces thinner foam because its chemical structure consists entirely of natural oils that interact with the organic acids in the kombucha itself. Although it produces less foam, kombucha soap is much gentler and more moisturising for sensitive skin.

Analysis of water content

Based on the test results, the water content of all kombucha and patchouli essential oil soap formulas (F1–F4) ranged from 0.30% to 0.33%, which means they met the quality requirements of the Indonesian National Standard (SNI 3532:2021) with a maximum limit of 23%. When compared to commercial soap (K+) which has a water content of 0.26%, the characteristics of the soap formulated in this study showed a very competitive quality profile and were within the equivalent low range. This low water content was achieved through a natural drying process (curing), where water freely evaporated optimally through the pores of the soap matrix. This minimal water content characteristic is very advantageous because it is able to keep the soap solid, does not easily shrink, and does not soften quickly when exposed to water during use (Wuryandari & Pamella, 2025), if the water content in the soap is too high, exceeding the SNI threshold, the soap will have an overly soft structure, dissolve quickly, and is prone to extreme shrinkage after drying. Furthermore, the low free water content in this formulation synergistically inhibits the growth of contaminating microorganisms (Ilsan et al., 2025), thus naturally extending the product's shelf life without relying on synthetic preservatives. Despite its very low percentage, this 0.3% water content is considered ideal because it maintains the soap's physical structure without becoming too brittle or prone to cracking.

Antibacterial test results

This test was conducted to determine the ability of kombucha solid bath soap with varying amounts of patchouli oil to inhibit *Staphylococcus aureus*. Data regarding the

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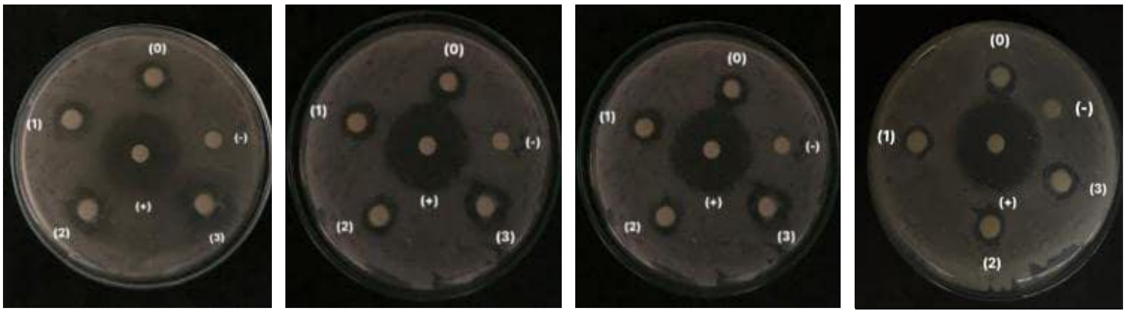
diameter of the inhibition zone (the clear area around the soap) from four replicates, along with the average, can be seen in **TABLE 2**.

TABLE 2. Antibacterial test results of kombucha bar soap with patchouli essential oil

Sample	**Inhibition zone diameter Repetition (mm)**				**Average (mm)**	**Inhibition zone criteria**
	1	**2**	**3**	**4**		
0 %	8.60	6.35	7.75	6.60	7.32	Moderate
1 %	8.60	6.60	8.25	6.65	7.52	Moderate
2 %	9.00	6.70	8.60	6.50	7.70	Moderate
3 %	11.50	10.65	10.10	10.45	10.67	Strong
K+ Chloramphenicol 150 µg	23.15	23.55	23.30	22.65	23.16	Very strong
K- Distilled water	0	0	0	0	0	None

The clear zones indicating antibacterial inhibition of soap at each repetition are presented in (**FIGURE 3**).



(a) (b) (c) (d)

FIGURE 3. (a) F1: repetition 1 (b) F2: repetition 2 (c) F3: repetition 3 (d) F4: repetition 4

According to the Clinical and Laboratory Standards Institute (CLSI), 2023, antibacterial inhibition strength is divided into four categories: 5 mm or less (weak), 6–10 mm (moderate), 11–20 mm (strong), and 21 mm or more (very strong). Based on the available data, the kombucha solid bath soap produced was proven to be able to significantly inhibit the growth of *Staphylococcus aureus*. According to CLSI standards, the formula without patchouli oil (F1, 0%) and the formula with low patchouli oil content (F2, 1% and F3, 2%) had moderate inhibition, with average diameters of 7.32 mm, 7.52 mm, and 7.70 mm, respectively. The greatest increase in inhibition was seen in formula F4 (3% patchouli oil), which produced an inhibition zone of 10.67 mm and was categorised as strong. As a comparison, the positive control (K+) using chloramphenicol provided very strong inhibition of 23.16 mm, while the negative control (K-) using distilled water did not provide any inhibition at all.

The inhibitory effect of the formula without patchouli essential oil proves that the use of 50 mL of fermented kombucha in this soap works well as a natural antimicrobial agent. This bioactivity directly correlates with its chemical profile, which is heavily

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dominated by dodecanoic acid (lauric acid) at 17.37% as identified from the highest peak resolved in the corresponding chromatogram spectrum. Dodecanoic acid acts as a primary structural fatty acid that disrupts bacterial cellular integrity while serving as a foam supporter (Kumalasari & Septiawan, 2025). Characteristically, the inhibitory activity of this kombucha base formulation on Gram-positive bacteria is higher than Gram-negative bacteria. This susceptibility occurs because Gram-positive bacteria possess a porous, thick peptidoglycan layer lacking an outer lipopolysaccharide membrane, rendering their cell walls highly vulnerable to the penetration of organic acids and phenolics (Anggraini et al., 2023).

A profound chemical rearrangement occurs when the formulation is fortified with 3% patchouli essential oil. Upon the addition of the essential oil, the concentration of dodecanoic acid drops drastically to approximately one-fifth of its initial value, falling from 17.37% down to 3.70%. Interestingly, despite this substantial decrease in the primary fatty acid driver, the antibacterial inhibition zone expands significantly from a moderate 7.32 mm to a strong 10.67 mm. This phenomenon implies that the enhanced bioactivity is no longer driven solely by dodecanoic acid but is explicitly accelerated by the emergence of patchouli alcohol, which registers at 3.35% in the fortified matrix as identified from the prominent peak in the modified chromatogram.

The presence of patchouli alcohol in the soap matrix is a direct outcome of the sesquiterpene biosynthesis pathways within *Pogostemon cablin* leaves. In the plant's trichomes, farnesyl pyrophosphate undergoes a complex enzymatic cyclisation mediated by patchouli alcohol synthase, a specialised sesquiterpene cyclase, to form the tricyclic tertiary alcohol skeleton of patchouli alcohol. Once embedded into the soap, patchouli alcohol acts as a potent hydrophobic pharmacophore. Its principal target of inhibition is the fluidic arrangement of the bacterial cytoplasmic membrane. Due to its high lipophilicity, patchouli alcohol easily partitions into and across the thick peptidoglycan layer, directly intercalating into the double lipid layer (lipid bilayer) of *S. aureus* (Pratiwi et al., 2026). This structural intercalation causes severe membrane fluidisation, compromises membrane permeability, and induces cellular leakage.

This mechanism works in a tight chemical synergy with the low molecular weight organic acids (primarily acetic acid) provided by kombucha fermentation. The acetic acid decreases the trans-membrane electrochemical potential by dropping the extracellular pH gradient. This surrounding acidity forces un-dissociated acid molecules to passively cross the compromised lipid bilayer into the neutral cytoplasm. Once inside, the acid undergoes intracellular dissociation, releasing excess protons (H^+) that aggressively disrupt the internal ion balance and deactivate vital enzymatic machinery (Rezaldi et al., 2026). Thus, the combination of patchouli alcohol and kombucha creates a lethal dual-action target system: the essential oil disrupts the structural boundary of the lipid membrane, while the organic acids collapse the internal homeostasis of the pathogen.

This dual mechanism combination offers significant functional advantages over commercial antiseptic soaps on the market, which generally rely on synthetic surfactants and antimicrobials. The overly aggressive use of synthetic chemicals in commercial products carries a high risk of triggering dermal irritation and dry skin due to the erosion of the skin's natural lipid layer (Leny et al., 2023). In contrast, the kombucha-patchouli

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soap preparation is dominated by a plant-based lipid base that is biocompatible with human epidermal tissue. This component has a dual function, namely as a natural amphiphilic surfactant that gently cleanses and as an emollient agent that maintains hydration in the *stratum corneum* layer. Thus, the formula with a 3% patchouli oil concentration is not only effective in degrading the cellular defense system of microorganisms by disrupting ionic balance (Rezaldi et al., 2026). In addition, this preparation also triggers disruption of lipid membrane permeability (Pratiwi et al., 2026). This synergistic effect is able to maintain the integrity and moisture of the skin's defense barrier continuously.

Chemical characteristics of soap using gas chromatography-mass spectrometry (GC-MS)

Samples were prepared using a solvent extraction method with a 10:1 solvent-to-sample ratio. One gram of sample was carefully weighed and dissolved in 10 mL of n-hexane. The n-hexane serves as a non-polar solvent to extract target compounds from the sample matrix. The mixture was then homogenised (through sonication or shaking) and filtered using a membrane filter (0.45 µm PTFE) to ensure no solid particles were carried into the GC-MS injection system.

Chemical compound profile analysis using Gas Chromatography-Mass Spectrometry (GC-MS) was conducted to identify the active components in kombucha solid bath soap. Based on the chromatogram results, 37 compound peaks were detected in the soap sample without patchouli essential oil (0%) and 34 compound peaks in the sample with patchouli essential oil concentration (3%). For efficiency and focus of the discussion, further analysis was limited to the 10 compounds with the highest area percentage of each sample. The profile of the 10 main compounds in the soap sample without patchouli essential oil (0%) is described as follows:

TABLE 3. GC-MS results of kombucha bar soap without patchouli essential oil

Number	Retention Time (minutes)	Compound Name	Area (%)
1.	2.614	Cyclopentane, methyl-	2.45
2.	21.038	Dodecanoic acid	17.37
3.	23.711	Tetradecanoic acid	4.76
4.	26.272	Hexadecanoic acid	9.58
5.	27.853	9-Octadecenoic acid (*Z*)-, methyl ester	5.10
6.	28.316	*cis*-13-Octadecenoic acid	12.44
7.	29.894	Glycidyl palmitate	6.62
8.	31.700	9-Octadecenoic acid (*Z*)-, oxiranylmethyl ester	14.13
9.	41.526	Avocadenol D	4.05
10.	44.753	*E,E,Z*-1,3,12-nonadecatriene-5,14-diol	4.71

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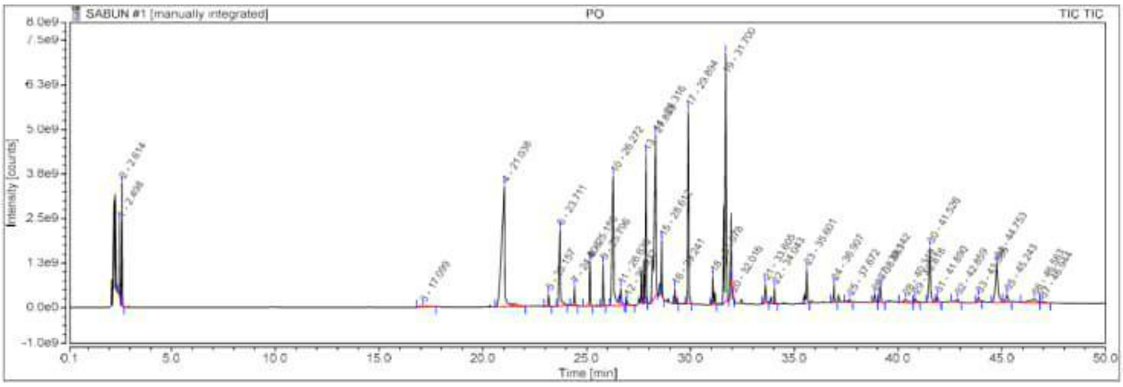

FIGURE 4. GC-MS results of kombucha bar soap without patchouli essential oil

Based on the results of gas chromatography-mass spectrometry (GC-MS) analysis, the chemical content profile of the kombucha solid soap sample without patchouli essential oil is dominated by dodecanoic acid (17.37%). The quantitative data presented in (**TABLE 3)** directly correspond to the highest peaks resolved in the chromatogram spectrum of (**FIGURE 4**). This compound functions primarily as a basic antibacterial agent and foam supporter (Kumalasari & Septiawan, 2025). Furthermore, the emollient or moisturising compound group provided a large contribution consisting of 9-octadecenoic acid (z)-, oxiranylmethyl ester (14.13%), cis-13-octadecenoic acid (12.44%), Glycidyl palmitate (6.62%) and 9-octadecenoic acid (z)-, methyl ester (5.10%), which collectively function to coat the skin and prevent dehydration. The skin protection activity is strengthened by hexadecanoic acid (9.58%) as an antioxidant (Sari et al., 2025). As well as tetradecanoic acid (4.76%) which complements the antibacterial properties and cleaning (Kumalasari & Septiawan, 2025). In addition, there are compounds with special metabolic activities, namely E,e,z-1,3,12-nonadecatriene-5,14-diol (4.71%) and Avocadenol D (4.05%) which act as antidiabetic and antilipidemic agents (Safitri et al., 2024). Finally, Cyclopentane, methyl- (2.45%) is present as a carrier compound that maintains the stability of the soap base mixture.

TABLE 4. GC-MS results of kombucha bar soap with 3% patchouli essential oil

Number	Retention Time (minutes)	Compound Name	Area (%)
1.	2.624	Cyclopentane, methyl-	23.03
2.	20.861	Dodecanoic acid	3.70
3.	22.395	Patchouli alcohol	3.35
4.	26.184	*n*-Hexadecanoic acid	3.15
5.	28.218	*trans*-13-Octadecenoic acid	4.35
6.	31.670	9-Octadecenoic acid (*Z*)-, oxiranylmethyl ester	8.24
7.	36.288	9-Octadecenoic acid (*Z*)-, octadecyl ester	2.26
8.	40.356	9-Octadecenoic acid (*Z*)-, oxiranylmethyl ester	2.90
9.	44.107	9-Octadecenoic acid (*Z*)-, oxiranylmethyl ester	11.86
10.	44.750	9-Octadecenoic acid (*Z*)-, oxiranylmethyl ester	5.04

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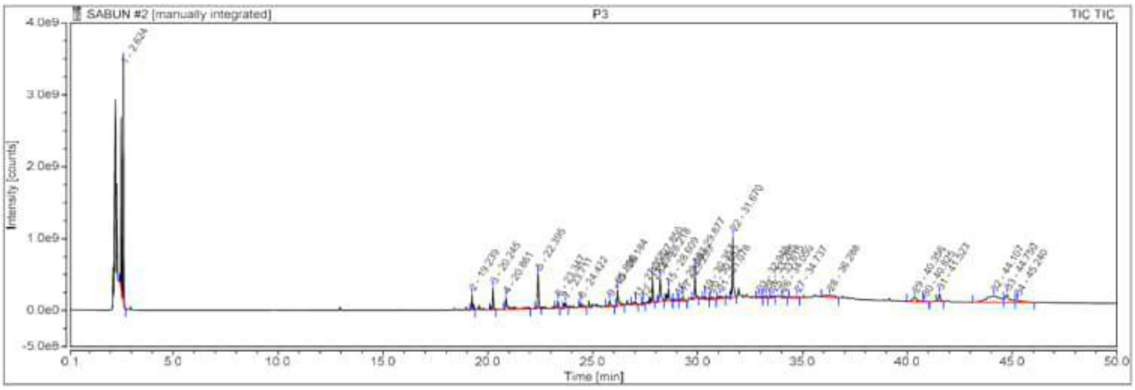

FIGURE 5. GC-MS Results of Kombucha Bar Soap with 3% Patchouli Essential Oil

In samples with the addition of 3% patchouli oil, the compound profile shifted significantly, reflecting the hybrid matrix of the saponified Kombucha base and the integrated essential oils. Comparative GC-MS diagnostics revealed that this integration caused a proportional reduction in dodecanoic acid (from 17.37% in the control base without oil to 3.70% in the 3% formulation). This shift does not represent a form of fatty acid degradation but rather represents a dilution of the volumetric peak area caused by the inclusion of highly volatile phytogenic compounds and the stabilised carrier introduced by the essential oils.

This chemical rearrangement is quantitatively summarised in (**TABLE 4**), identifying the major bioactive from the peaks visualised in (**FIGURE 5**). This profile shift is marked by the dominance of Cyclopentane, methyl- (23.03%, as shown in **TABLE 4**), which serves to maintain the stability of aroma, texture, and provide additional antioxidant activity (Jemi et al., 2023). Concurrently, the retention of 3.35% patchouli alcohol (comprising the peak in **FIGURE 5**) directly demonstrates the post-saponification stability of the essential oil's primary bioactive tracer, uniquely mapping its therapeutic concentration.

Conversely, the core saponified Kombucha base details detailed in (**TABLE 4**) are represented by the abundance of ester-based emollient and antioxidant compounds, primarily the accumulation of 9-octadecenoic acid (Z)-, oxiranylmethyl ester. This compound appeared at four different retention times (11.86%; 8.24%; 5.04%; and 2.90%) with a total area reaching 28.04%. This moisturising effect is supported by trans-13-octadecenoic acid (4.35%), which also possesses anti-inflammatory properties (Muhtadi et al., 2025), as well as 9-octadecenoic acid (Z)-, octadecyl ester (2.26%) which acts as a skin conditioning agent. Meanwhile, for the health protection aspect, the bioactive potential of this soap is significantly enhanced by the presence of patchouli alcohol (3.35%). Originating from the added essential oil in the 3% formulation, patchouli alcohol plays a key role through a strategic synergy with dodecanoic acid (3.70%) to provide strong and responsive antibacterial activity (Ulandari et al., 2022; Kumalasari & Septiawan, 2025). This series of components is concluded by n-hexadecanoic acid (3.15%), which acts as an antioxidant (Sari et al., 2025) and stabilises the soap foam structure.

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Conclusion

This study concludes that the solid soap formulation combining 50% kombucha fermentation solution and 3% patchouli essential oil is the most optimal variation that meets SNI. Physically, this soap shows an ideal pH of 8.0, an initial foam height ranging from 5.80-6.50 cm, and excellent foam stability of up to 91.5%. The resulting minimum water content (0.30%-0.33%) is also far below the maximum limit of standard provisions. In terms of bioactivity, the addition of 3% patchouli oil significantly increases the antibacterial efficacy against *Staphylococcus aureus* ATCC 25923 with a strong category inhibition zone of 10.67 mm. GC-MS analysis confirmed the post-saponification retention of patchouli alcohol (3.35%) and dodecanoic acid (3.70%) as the main active components that underscore the dual-action antibacterial mechanism, so this formulation has great potential as a safe, stable, and effective natural skin cleanser.

Author contributions

MK: Project conception, research methodology, implementation of laboratory experiments, data analysis of physical and antibacterial test results, and drafting the original manuscript. DH: Project conception, methodology supervision, critical review of data analysis results, and editing and finalisation of the manuscript. All authors have read and approved the final version of this manuscript for publication.

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Conflicts of interest statement

The authors declare that they have no conflicts of interest related to the research, authorship, and/or publication of this article. The entire research process was conducted independently for the purpose of advancing scientific knowledge.

Ethical compliance

No human subjects or animal testing were involved in this study, and all procedures were performed in a laboratory environment without risk to health.

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