

Phytochemical content of Maluku nutmeg (*Myristica fragrans* Houtt) in Inhibiting Skin Pathogenic Bacteria

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Abstract

Banda nutmeg (*Myristica fragrans* Houtt) is one of the native species of Maluku that has various benefits other than as a cooking spice. Nutmeg, especially its fruit, offers significant advantages in the medical field as a natural dermatological product, effectively treating skin infections caused by pathogens. One such bacterial infection is pyoderma, characterized by the presence of pus-producing bacteria, leading to the formation of pustules or greenish-yellow crusts upon examination. Among the bacteria responsible for skin infections, *Pseudomonas aeruginosa* and Methicillin Resistant *Staphylococcus aureus* (MRSA) are notable bacterial species that cause skin infections. The active compounds found in nutmeg act as antibacterial agents by interfering with the peptidoglycan components within bacterial cells. This interference results in the formation of intact cell walls, ultimately leading to bacterial cell death. This study aimed to identify the bioactive profile of Maluku nutmeg extract metabolites and assess their inhibitory effects on pathogenic bacteria, specifically *P. aeruginosa* and MRSA, responsible for skin infections. The study revealed the presence of various bioactive compounds in nutmeg extract, including alkaloids, flavonoids, terpenoids, phenols, and tannins. Notably, the application of nutmeg pulp at an 80% concentration proved highly effective in inhibiting bacterial growth, surpassing the efficacy of the positive control. The enhanced effectiveness against MRSA, compared to *P. aeruginosa*, can be attributed to the higher concentration of secondary metabolites present in nutmeg extracts.

Keywords: nutmeg, phytochemical screening, antibacterial, regeneration

Introduction

The potential of nutmeg as a high-value export

commodity from Maluku is often overshadowed by the focus on its seeds, leaving the fruit's flesh as mere waste. The lack of awareness among the public, particularly in Maluku, regarding nutmeg's medical benefits hampers its production. While Maluku locals traditionally process nutmeg flesh into sweets, they remain unaware that it holds significant potential as a natural dermatological product for treating various diseases. In dermatology, infectious diseases are a key area of study, encompassing bacterial infections like pyoderma, fungal infections (dermatomycosis), viral infections, and manifestations of parasites. Pyoderma, caused by pus-producing bacteria, is identifiable through pustules or greenish-yellow crusts. Common causes, including *Staphylococcus aureus*, *Streptococcus pyogenes*, *Acinetobacter* sp., and *Pseudomonas* sp., can lead to diverse skin infections.

Recent research, such as the study conducted by Rashidian et al. in 2022, highlights nutmeg's metabolite content's effectiveness in inhibiting both gram-positive and gram-negative bacteria, including *Pseudomonas aeruginosa* (gram-negative) and Methicillin Resistant *Staphylococcus aureus* (gram-positive), as proven through the disc diffusion method in vitro. Understanding the skin's inflammatory response to bacteria is crucial; localized inflammation and suppuration are common in bacterial skin infections. Some bacteria can cause bacteremia or lesions in skin tissue. While antibiotics can mitigate pathogenic bacteria, overuse can lead to resistance or genetic changes that enable bacteria to survive. Hence, exploring alternative solutions, particularly from natural sources, is vital. Natural ingredients, like nutmeg, offer a sustainable approach to healthcare. Traditional medicine, derived from plants, not only provides affordability and accessibility but also boasts minimal side effects. In essence, recognizing the untapped potential of nutmeg's flesh in dermatology and promoting its use as a natural remedy can pave the way for a more sustainable and effective approach to combating skin infections.

The essential oils derived from nutmeg have long been recognized for their potent antimicrobial properties, as reported by Nurhasanah (2014). Nutmeg (*Myristica fragrans* Houtt), a native species of Maluku, has a plethora of benefits that have been well-documented over the years. Its efficacy in the medical field, particularly as a potent antibacterial agent, has been extensively studied (Matulyte et al., 2020). Studies have demonstrated that nutmeg essential oil possesses antioxidant effects, effectively inhibiting bacterial growth (Kamelia and Silalahi, 2018). A study by the World Health Organization has identified nutmeg as one of the medicinal plants with exceptional antibacterial properties, surpassing even certain antibiotics (Green, 2005 as cited in Arrizqiyani et al., 2017). Phytochemical screening has revealed the presence of alkaloids in nutmeg extract, which function as antibacterial agents by disrupting the peptidoglycan components in bacterial cells, leading to intact cell wall formation and subsequent bacterial cell death (Siegers et al., 2022). Nutmeg essential oil extracted from nutmeg pulp also finds utility in treating various health issues, including bladder and urinary tract inflammation, halitosis, dyspepsia, flatulence, impotence, insomnia, and a myriad of skin problems (Simamora et al., 2018).

The pulp of nutmeg contains flavonoid compounds that inhibit bacterial growth, including phenolic compounds capable of denaturing proteins and damaging bacterial cell membranes. Additionally, saponin, found in nutmeg, acts as an antibacterial compound by interfering with the permeability of bacterial membranes. The essential oil extracted from nutmeg contains hydrocarbon monoterpenes, monoterpene acids, and aromatic ethers, all functioning as potent antibacterial substances. Myristicin, a key compound found in nutmeg oil, possesses functional groups such as allyl, phenyl, and ether. Moreover, chalcone-derived compounds, secondary metabolites of the flavonoid group, are known to be effective in inhibiting bacterial growth (Woriwun et al., 2021). The flavonoid compounds present in nutmeg flesh extract impact antioxidant activity, neutralizing the toxic effects of free radicals by donating hydrogen ions or capturing reactive oxygen species (ROS) directly, thereby preventing ROS regeneration (Wally, 2022).

Given the diverse and potent properties of nutmeg extract compounds in inhibiting the growth of pathogenic bacteria, this research was conducted to unveil the comprehensive bioactive profile of Maluku nutmeg extract (*Myristica fragrans* Houtt) and explore its inhibitory activity against skin infections caused by *Pseudomonas aeruginosa* and methicillin-resistant *Staphylococcus aureus* (MRSA). Our study can potentially develop natural antimicrobial solutions.

Material and Methods

This research is a pure experimental study in a laboratory (*True Experimental*) because this study was given an intervention with all external variables affecting controlled. The study was organized in a completely randomized design with 6 treatments and 3 replications on each test bacteria including control, namely P1 (20% nutmeg pulp), P2 (40% nutmeg pulp extract), P3 (60% nutmeg pulp extract), P4 (80% nutmeg pulp extract), positive control (*chloramphenicol*), negative control (aquadest). This research was conducted at the Laboratory of Basic Chemistry, Pattimura University, Ambon and the Laboratory of the Center for Environmental Health Engineering and Disease Control in Ambon. The time of this research was carried out in August 2022. The research targets include the bioactive content of secondary metabolites based on phytochemical analysis of the flesh extract of nutmeg (*Myristica fragrans* Houtt) with various concentrations and antibacterial activity against *P. aeruginosa* and Methicillin Resistant *Staphylococcus aureus* (MRSA) bacteria.

The research materials included nutmeg obtained from the community garden of Hitu Village, Central Maluku Regency, while the test bacteria were *Pseudomonas aeruginosa* ATCC 9027 and methicillin-resistant *Staphylococcus aureus* ATCC 6538 from Laboratory BTKL Ambon, *Myristica fragrans*, Sabouraud dextrose broth, nutrient agar, aquadest, Carbopol 940, CMC-Na, propyl paraben, methyl paraben, propylene glycol, H₂SO₄, alcohol 70%, ethanol 96%, ethanol 70%, concentrated HCl, Mg powder, FeCl₂ 1%, NaCl 0.9%, Wagner's solution, aquadest, Buchardat and Mayer reagent (Mujipradhana et al., 2018). All tools and media were sterilized at temperature of 121°C for 15 minutes prior to use,

The flesh of nutmeg was collected from the same tree by selecting 5 mature fruits and then washing them until clean. The nutmeg flesh that has been washed clean was dried in the sun, ground using a blender, then sieved using a 60-mesh sieve. Two-hundred grams of nutmeg powder was weighed and dissolved in 96% absolute EtOH (800 ml) while stirring, then left for 5 days with 2 filtrations. The filtrate product was evaporated using a rotary evaporator (Saharuddin and Kondolele, 2020). The concentrated extract was diluted according to the desired concentration using distilled water to become extracts of concentration 20%, 40%, 60% and 80%.

Phytochemical Screening Test

Phytochemical screening of the fresh drying extract

was carried out to determine the content of secondary metabolites contained in it (Sirait and Enriyani, 2021). The content of these metabolites includes alkaloids, saponins, triterpenoids, flavonoids, and tannins.

The steps for the alkaloid test are to drop 3 drops of Wagner's solution on 0.5 ml of the extract, observe if the bottom of the reaction tube changes color to brown or yellowish orange, then the extract contains alkaloids. The saponin test stage is to add 2 ml of hot water to 0.5 ml of the extract, shaken quickly, if there are bubbles such as foam that can be held for more than 10 seconds, the extract contains saponins. The triterpenoid test stage is to add 3 drops of Buchardate solution to 0.5 ml of extract then add 1 ml of H_2SO_4 and 0.25 anhydrous acetic acid, observe if the color of the solution formed is red-orange or brownish purple then it is a triterpenoid, but if the solution is bluish green, then steroids are formed.

The terpenoid test step is to add 2 drops of concentrated sulfuric acid to 0.5 ml of the extract and shake it quickly, if it turns yellow it indicates that there is a terpenoid content (Bakhriansyah et al., 2021).

The flavonoid test stage begins by adding 3 drops of HCl and 0.2 g of magnesium powder to the 0.5 mL extract; if the solution turns pink or brownish red, the extract contains flavonoids. The tannin test begins by adding 2 drops of 1% $FeCl_2$ solution to 0.5 ml the extract; the solution will turn blackish green, blue, dark blue blackish or purple when it contains tannin (Bhernama, 2020).

Preparation of basic and germination media

The basic media consists of 2.8 g of nutrient agar then mixed with 100 ml of distilled water (28 g.L^{-1}) in an Erlenmeyer flask. The seed medium used 7 grams of nutrient agar was dissolved in 250 ml of distilled water in an Erlenmeyer flask.

Each medium was homogenized with a stirrer over a water bath until it boiled. The homogenized media was sterilized in an autoclave at 121°C for ± 15 minutes, waiting for the temperature to reach $450\text{--}500^\circ\text{C}$. The base media and the seedling media were used as test media for the base and second layers (Muljono et al., 2016).

Bacterial culture and inoculation

Pseudomonas aeruginosa and MRSA was taken using a sterile needle, then inoculated onto the agar slanted media with a scraping technique and incubated in an incubator at 37°C for 24 hours (Muljono et al., 2016).

Bacterial solution test (Mc.Farland Solution)

Pseudomonas aeruginosa and MRSA was taken using a sterile ossicle needle, then inoculated onto the agar slanted media with a scraping technique and incubated in an incubator at 37°C for 24 hours. Inoculation was conducted by taking *P. aeruginosa* and MRSA with sterile ossicle and suspended in a tube containing 2 ml of 0.9% NaCl solution until the turbidity was the same as the standard turbidity of 0.5 strength Mc. Farland solution (Gansareng et al., 2018; Muljono et al., 2016).

Media testing

The nutrient agar (NA) medium was weighed as much as 5.04 g, then dissolved with 180 ml aquadest (28 g.L^{-1}) in an Erlenmeyer flask, homogenized using a magnetic stirrer and sterilized by autoclave at $121^\circ\text{C} \pm 15$ minutes and wait until it is cool down. About 30 mL liquid media is poured into a petri dish, leveled, and allowed to half solidify, followed by placing the blanks which had been soaked in the tested extract for 30 minutes and incubated for 1 x 24 hours at 37°C .

Cream manufacturing and testing

Cream was made by melting *Cera alba* as a cream emulsifier using a porcelain cup over a water bath so that the mixture can melt quickly. The extract was placed in a warm mortar so that the melt does not form a crust, does not stick to the container, and maintains the stability of the cream mass, then added to the mixture previously stirred ad homogeneous.

The next test is cream of nutmeg flesh extract with the addition of honey to be applied to patients with bacterial infections and then measuring the scale of wound closure/skin infection using a measuring device.

Creation of positive control and negative control

The positive control used was a 30 mg chloramphenicol disc, placed on the media and incubated at 37°C (Fredella et al., 2022) while the negative control was 20 mL of distilled water. The negative control was used as a comparison of the positive control.

Antibacterial tests

The Nutrient Agar (NA) medium was weighed as much as 5.04 g, then dissolved with 180 ml quads (28 g.L^{-1}) in an Erlenmeyer flask. Homogenization was conducted using a magnetic stirrer followed by sterilization using autoclave at 121°C for 15 minutes. The media that is still liquid is poured into

a petri dish of about 30 ml, leveled, and allowed to half solidify (Wahyuni and Karim, 2020). The blanks that had been soaked in the tested extract for 30 minutes were incubated for 1 x 24 hours at 37°C.

Bacterial Inhibitory Zone Tests

After 24 hours of incubation, the bacteria inhibition zone was observed and measured in the form of a clear zone (Fredella et al., 2022). Bacterial susceptibility was indicated by the presence of a clear zone around the paper disk. Measurements were made using a caliper in millimeters (mm). The mechanism for measuring the horizontal diameter is added to the vertical diameter and then divided by two. The formula for calculating the zone of inhibition used is as follows (Fiana et al., 2020).

$$(DV - DC) + (DH - DC) / 2 \dots\dots (1)$$

Information:

DV: Vertical Dimeters

DH: Horizontal diameters

DC: Disc Diameters

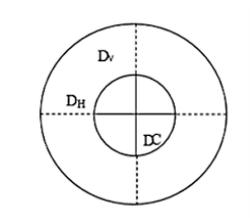


Figure 1. Measurement of the inhibition zone (Toy et al., 2015).

Inhibition data was interpreted based on Davis and Stoud (1971) and Fredella et al., (2022), summarized in Table 1.

Data Analysis

The antibacterial activity data was collected in the form of antibacterial inhibition zones, which were subsequently analyzed using ANOVA. Normality of the data distribution was confirmed if the p-value was greater than 0.05; otherwise, if $p < 0.05$, the distribution was considered non-normal. For normally distributed inhibition zone data, the analysis proceeded with the Levene Test to assess the homogeneity of variance among the data groups. Subsequently, the Tukey test was conducted to identify significant differences

within each treatment group. In cases where the data did not follow a normal distribution but exhibited homogeneity, the analysis continued with the Kruskal-Wallis test. Post hoc comparisons were then made using the Mann-Whitney test to determine specific differences between groups.

Result and Discussion

Phytochemical Screening Test

Phytochemical tests were conducted at the Basic Chemistry Laboratory of Pattimura University, Ambon, to identify secondary metabolites present in fruit pulp extracts. Parameters such as color changes were observed and compared with existing literature references. The presence of flavonoid compounds in an extract was indicated by a change in color to greenish yellow. Terpenoid compounds manifested as a red or orange color change. Tannin compounds were identified by a bluish color change, and steroid compounds resulted in a clear color after shaking. Phenolic compounds were distinguished by a bluish-brown color, and the presence of alkaloid compounds led to the formation of a brownish precipitate on the lower surface of the extract. The results of the phytochemical tests on nutmeg flesh extract are in Table 2.

Bacterial Inhibitory Zone Tests

The results of the antibacterial activity of nutmeg flesh extract showed different inhibition zones in each treatment group for both *P. aeruginosa* and *MRSA* bacteria (Table 3)

Based on the data in Table 3, the best dose of nutmeg pulp extract was at a concentration of 80% for both *P. aeruginosa* and *MRSA*. The inhibitory power for *P. aeruginosa* bacteria was in the strong category, while for *MRSA* bacteria had a very strong category and did not differ much from chloramphenicol antibiotics. The differences in the inhibition zones are described in Figure 2.

Based on Figure 2, nutmeg pulp extract can inhibit the growth of *P. aeruginosa* and *MRSA*. The comparison of the mean zone of inhibition was higher for *MRSA* bacteria when compared to inhibition on *P. aeruginosa*

Table 1. Bacterial inhibition criteria (Toy et al., 2015)

Criteria level	Criteria inhibition (mm)
Weak	< 5 mm
Rather strong	5-10 mm
Strong	10-20 mm
Very Strong	>20 mm

Table 2. Phytochemical screening test results

Compound	Colour	Results
Alkaloid	Orange-yellow	+
Flavonoid	Yellow-green	+
Terpenoid	Red	+
Steroid	Clear	-
Phenolics	Blue-brown	+
Tannin	Blue	+

Note: + indicates that the compound was detected; - indicates that the compound was not detected

Table 3. Average inhibition zone of *P. aeruginosa* and MRSA (mm)

Pathogenic bacteria	Average inhibition zone (mm)	Inhibitory category
<i>P. aeruginosa</i>		
20%	10.6	Rather strong
40%	10	Rather strong
60%	10.3	Rather strong
80%	17.3	Strong
K+	0.67	Weak
K-	0	-
MRSA		
20%	14.6	Strong
40%	12.3	Strong
60%	12	Strong
80%	19.6	Very strong
K+	21.3	Very strong
K-	0	-

bacteria, likely because the secondary metabolite content in nutmeg extract was more effective for MRSA than *P. aeruginosa*.

After measuring the clear zone or the zone of inhibition of bacterial growth, a comparison test was conducted to determine the effects of nutmeg pulp extract on the inhibition of growth of *P. aureginosa* and MRSA bacteria (Table 4).

The experiments involved various treatments with different concentrations of nutmeg pulp extract (20%, 40%, 60%, and 80%) to assess their effectiveness in inhibiting the growth of *P. aeruginosa* and MRSA. The results, as summarized in Table 4, revealed significant differences in the inhibition of *P. aeruginosa* growth only with the 80% concentration, while for MRSA, significant differences were noted with the 20% and 80% concentrations, compared to the control groups (K+ and K-). Interestingly, the 40% and 60% concentrations did not show significant differences in inhibiting MRSA growth.

Specifically, the highest inhibition of *P. aeruginosa* growth (17.3 mm) was achieved with an 80% concentration of nutmeg pulp extract. Similarly, the highest inhibition of MRSA growth (19.6 mm) occurred with the 80% concentration of nutmeg pulp extract. However, these inhibitory effects were not as potent as those observed with chloramphenicol, the standard antibiotic used in the study.

The Effectiveness of Nutmeg Cream on Skin Regeneration

The results of the cream effectiveness test for skin tissue regeneration were carried out on people who had ulcers with 3 varied treatments: nutmeg cream with the addition of 5% honey (P1), nutmeg cream with the addition of 10% honey (P2), and nutmeg cream with the addition of 15% honey (P3) which was tested for 7 days of observation. The results found can be seen in Figure 4.

According to Figure 4, the nutmeg flesh cream

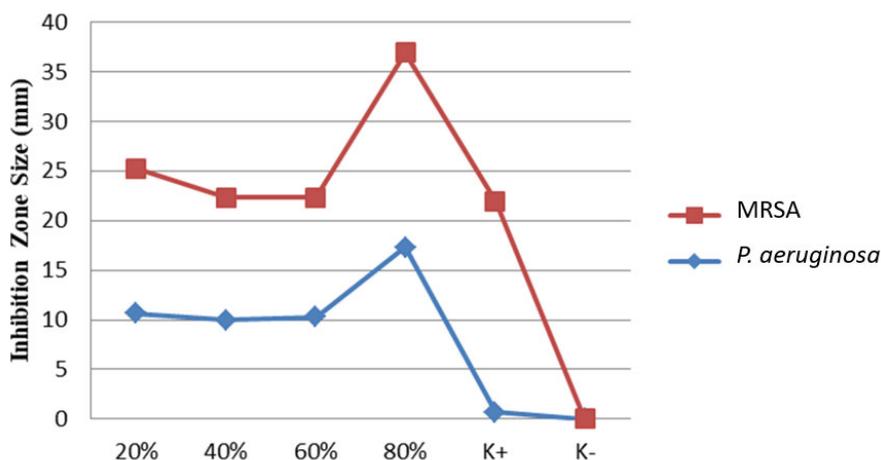


Figure 2. Differences in the inhibition zone of *MRSA* bacteria and *P. aeruginosa*

Table 4. Comparison concentration test

Test bacteria	Treatment	N	Subset for $\alpha= 0.05$	
			1	2
<i>P. aeruginosa</i>				
	Concentration -	3	.000	
	Concentration +	3	6.667	6.667
	60%	3	10.333	10.333
	40%	3		16.000
	20%	3		16.667
	80%	3		17.333
<i>MRSA</i>				
	Concentration -	3	.000	
	60%	3	8.100	8.100
	40%	3	8.433	8.433
	80%	3	13.667	13.667
	20%	3	14.667	14.667
	Concentration +	3		21.333

treatment supplemented with 15% honey (P3) exhibited the most potent regenerative ability in healing boils caused by skin pathogenic bacteria infections. Nutmeg flesh extract contains a variety of secondary metabolite compounds, such as alkaloids, flavonoids, terpenoids, phenolics, and tannins. This composition was also verified by Hoda et al. (2020), who found that nutmeg extract comprises active components like saponins, tannins, terpenoids, carbohydrates, steroids, and flavonoids, all of which act as antimicrobials.

In the comparison results, the 80% concentration significantly outperformed other treatment groups in inhibiting the growth of *P. aeruginosa*. Additionally, the 80% concentration's inhibition of *MRSA* growth significantly differed from the effects observed with

the 20% concentration and the positive control (K+). *MRSA* bacteria, a strain of gram-positive bacteria, are only resistant to penicillin antibiotics, not chloramphenicol. As noted by Faisal and Permana (2020), chloramphenicol antibiotics remain effective against *Staphylococcus aureus* bacteria, but not against *P. aeruginosa*. This is evident from the large inhibition zone formed by the positive control against *MRSA* bacteria (>18 mm in diameter). *P. aeruginosa* possesses an outer membrane rich in lipids, separated from the cell wall by the periplasmic space, making antibiotic activity less effective. The substantial inhibition zone observed in *MRSA* bacteria results from the presence of the antibacterial substance chloramphenicol 30 mg (Utami et al., 2015).

Our study demonstrated that an 80% concentration

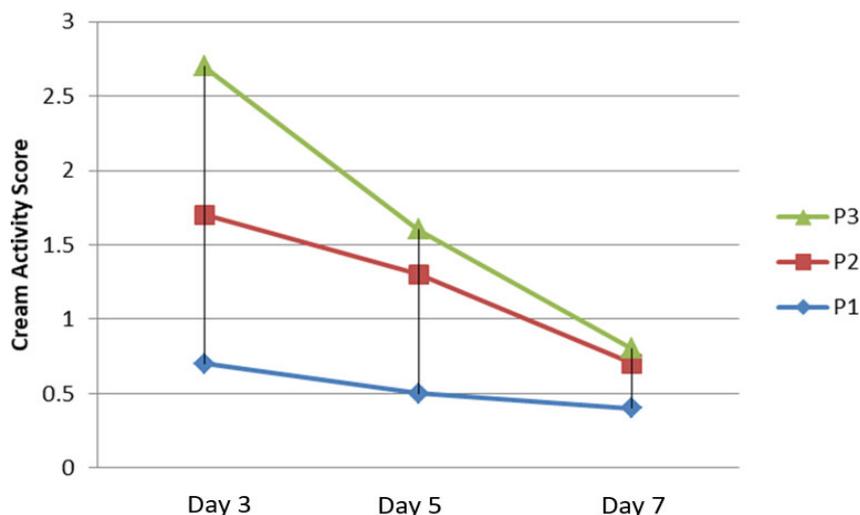


Figure 4. Cream effectiveness for skin regeneration. P1: nutmeg cream + 5% honey; P2: nutmeg cream + 10% honey; P3: nutmeg cream + 15% honey.

of nutmeg pulp extract effectively inhibited MRSA, aligning with Singh et al.'s research (2017) indicating that nutmeg pulp extract is more effective against gram-positive bacteria than gram-negative ones. The thin cell wall of gram-positive bacteria increases the permeability, allowing antibacterial components to penetrate the cell wall more easily and destroy the peptidoglycan within, leading to bacterial cell death.

The effectiveness of the 80% nutmeg pulp extract suggests that higher concentrations contain greater amounts of active substances. The size of the inhibition zone formed directly correlates with the concentration used. Phytochemical tests revealed the presence of alkaloids in nutmeg plants, which possess antibacterial properties by disrupting peptidoglycan components in bacterial cells. This interference leads to the formation of an intact cell wall layer and subsequent bacterial cell death (Siegers et al., 2022; Kaawoan et al., 2016). Gram-positive bacteria, being polar, allow easy penetration of polar substances like saponins and alkaloids through their cell walls (Anastasia et al., 2022).

Nutmeg pulp extracts are rich in flavonoids and tannins, as highlighted by Robinson (1995) in Gansareng et al.'s study (2018). Flavonoids act as antibacterials by binding to bacterial proteins, inhibiting enzyme activity, and disrupting bacterial metabolic processes. Their lipophilic nature enables them to damage bacterial cell membranes, which contain lipids, allowing these compounds to pass through the membrane. Flavonoids, being the largest group of phenolic compounds, effectively inhibit bacterial growth (Isromarina et al., 2020).

At a cellular level, flavonoids inhibit DNA synthesis,

preventing pathogenic bacteria from replicating. They also have the potential to interact with plasmids (circular DNA in bacteria) in the nucleus, further interfering with bacterial replication (Rezaldi et al., 2022). The difference in polarity between the lipids constituting DNA and the alcohol groups in flavonoids damages the DNA lipid structure in bacteria, leading to bacterial cell lysis or death. Additionally, flavonoid compounds hinder bacterial oxygen utilization. Inhibition of macromolecular biosynthesis, a process requiring energy, prevents the development of bacterial molecules into more complex forms (Rinanti et al., 2020).

Furthermore, steroid compounds and terpenoids present in nutmeg pulp extracts inhibit bacterial growth by disrupting protein synthesis. These compounds accumulate and induce changes in bacterial cell components. Terpenoid compounds, soluble in lipids, easily penetrate the cell walls of both gram-positive and gram-negative bacteria. Steroids, acting as antibacterials, associate with membrane lipids, leading to sensitivity to steroid components. This sensitivity causes leakage in liposomes, decreasing membrane integrity and altering cell membrane morphology. These changes result in increased cell brittleness and lysis (Wahyuni and Karim, 2020).

Based on these findings, it can be inferred that the application of nutmeg flesh cream is effective in rejuvenating skin cells infected with bacteria. This skin cell regeneration is believed to be closely linked to the antioxidant properties present in nutmeg. Gansareng et al. (2018) demonstrated that nutmeg contains robust antioxidants, primarily from the flavonoids, tannins, saponins, triterpenoids, phenols, and myristicin classes. Myristicin is specifically known

to target cells with abnormal growth, indicating its selective action. Flavonoids, being potent antioxidants, play a crucial role in repairing and safeguarding cell structures. Their effectiveness is further enhanced by α -tocopherol, tannins, polyphenols, saponins, and minerals like magnesium found in nutmeg leaves (Kaawoan et al., 2016).

Furthermore, flavonoids possess the ability to inhibit oncogenesis through various mechanisms. Firstly, they induce apoptosis and halt the cell cycle by inhibiting topoisomerase enzymes. Additionally, they suppress cytochrome P-450, rendering carcinogenic compounds inactive, and enhance the expression of the enzyme glutathione S-transferase, facilitating the detoxification of carcinogens for swift elimination from the body. At a cellular level, flavonoids inhibit DNA synthesis, preventing pathogenic bacteria from replicating. These findings underscore the multifaceted benefits of nutmeg and its potential in combating bacterial infections and promoting skin health. Flavonoids possess the potential to interact with plasmids (circular DNA in bacteria) found in the nucleus. The disparity in polarity between the lipids constituting DNA and the alcohol groups in flavonoid compounds is a contributing factor to the damage inflicted upon the DNA structure in bacterial lipids. This damage leads to bacterial cell lysis or death (Rezaldi et al., 2022).

In addition to the beneficial compounds found in nutmeg, honey also exhibits potent antimicrobial properties. As noted by Yuliati (2017), honey has long been utilized in traditional medicine due to its antibacterial qualities, attributed to its high osmolarity, hydrogen peroxide content, low pH, and water activity. The acidic pH of honey, ranging from 3.2–4.5, inhibits the metabolism of both gram-negative and gram-positive bacteria, making them susceptible to lysis. Another significant aspect contributing to honey's ability to inhibit bacterial activity is its rich content of vitamins B2, B3, B6, C, K, carotene, and biotin. These vitamins enhance the body's resistance to bacterial infections, fortifying the immune response against microbial threats.

Conclusion

Our study establishes that nutmeg flesh extract comprises various secondary metabolite compounds, including alkaloids, flavonoids, terpenoids, phenolics, and tannins. These findings indicate a significant impact of nutmeg pulp extract on the activity of both *Pseudomonas aeruginosa* and methicillin-resistant *Staphylococcus aureus* (MRSA). Consequently, the nutmeg flesh cream demonstrates efficacy in skin cell

regeneration. This effectiveness is attributed to the higher efficacy of the secondary metabolite content in nutmeg extract against MRSA compared to *P. aeruginosa*.

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