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ATSIRI OIL POTENTIAL OF PEEL OF KAFFIR LIME, LIME, AND CALAMANSI ORANGE AS ANTIOXIDANT AND ANTIBACTERIA

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ABSTRACT: Orange is a member of the Rutaceae family which is known as one of the horticultural crops that is widely cultivated in the tropical and subtropical regions of southern Asia. Citrus plants such as fruit peels are used as a source of essential oil-producing ingredients that have antioxidant and antimicrobial activity. This study aims to determine the potential of kaffir lime, lime, and calamansi orange peel essential oils as antioxidants and antibacterials. Scoping review uses the PEOS framework and several databases such as the electronic database Atlantis Press, PubMed, ScienceDirect, and the Google Scholar search engine. Search results that meet the criteria are then analyzed using PRISMA Flowchart, Data Extraction, and Theme Mapping. The results of the scoping review study of 74 articles related to titles and abstracts, 8 articles met the inclusion and exclusion criteria. Essential oils from kaffir lime peel (*Citrus hystrix*), lime (*Citrus aurantifolia*), and calamansi orange (*Citrus microcarpa* Bunge.) have antioxidant and antibacterial activities. The higher the concentration of essential oils from the peel of the fruit, the greater the antioxidant and antibacterial activity produced. Fresh orange peel essential oil has the strongest antioxidant and antibacterial activity compared to orange twig and leaf oil.

Keywords: Essential Oil, Kaffir Lime, Lime, Calamansi Oranges, Antioxidant, Antibacterial.

INTRODUCTION

Medicinal plants have long been used for the alternative treatment of illness, recovery, health maintenance, and disease promotion (Jeffrey et al., 2019). This is because plants contain many compounds that have medicinal properties, a class of organic molecules used to preserve health and treat disease is referred to as phytochemical substances. Plant phytochemical compounds that provide pharmacological effects are secondary metabolites, including essential oil groups, flavanoids, alkaloids, steroids, and triterpenoids which will give a very specific aroma, taste, and smell to the plant of origin (Bączek et al., 2017). Citrus is a member of the Rutaceae family which is known as one of the horticultural crops that is widely cultivated in the tropical and southern subtropical regions of Asia. Furthermore, citrus plant parts, such as leaves, fruit peels, and twigs are used as a source of essential oil-producing ingredients. In general, essential oils are composed of a complex mixture of terpenoids, oxygenated terpenoids and sesquiterpenes, phenylpropanoids, and oxygenated benzenoids. These compounds are categorized as plant secondary metabolites and are responsible for the characteristic aroma. In several reports it is stated that essential oils have various health benefits and are used in aromatherapy, the pharmaceutical industry as well as for the aroma and taste of food. It was even reported that essential oils from citrus peels have antioxidant and antimicrobial activity (Indrayani, 2019).

The essential oils of several plants have been known to have antibacterial activity. The antibacterial activity of essential oils is due to the fact that essential oils contain compounds that can inhibit or kill bacterial growth. Essential oil components that contain phenol groups such as carvacrol have the potential to be antibacterial (Jirna et al., 2017). Orange peel contains a variety of volatile compounds that are very beneficial. Processing orange peel waste into essential oils is expected to increase added value (Indrayani, 2019). Orange peel oil contains active components that are beneficial, including terpene compounds, flavonoids, coumarins, linalol, and others. Additionally, orange peel oil has antibacterial and antioxidant properties and acts as a larvicide (Liew et al., 2020). Orange peel oil is stored in oil glands on the outer skin (flavedo) and develops with the maturity level of the fruit (Oe et al., 2019). Orange peel contains complex carbohydrates such as cellulose, hemicellulose, pectin and other components. Removal of oil from the material can be done by damaging the outer layer that covers the sac/oil gland. Degradation by living things like bacteria and fungi is anticipated to improve oil production yields (Mohideen et al., 2022).

The main content of the peel of the calamansi orange is essential oil. The main content of citrus fruit peel is pectin and essential oils. The pectin content in citrus peels ranges from 15-25% of the dry weight. The content of essential oils in citrus peels is around 70-92%. The peel part of the citrus fruit contains essential oils which consist of various components such as terpenes, sesquiterpenes, aldehydes, esters, and sterols (Nabilah et al., 2020). Meanwhile, kaffir saponins, tannins, and essential oils are present in lime peel. Kaffir lime leaves are also used as the main ingredient in traditional medicines. Kaffir lime peel has a pharmacological effect as an antiseptic and antioxidant. Compounds contained in kaffir lime peels that function as antibacterials are tannins (Arfania, 2018). The essential oil content of lime peel is effective as an antimicrobial in killing gram-negative bacteria, such as *Escherichia coli* and *Staphylococcus aureus* bacteria with a concentration of 0.0625% (Husni et al., 2021). *Escherichia coli* is a bacteria that dwells in both human and animal intestines. These bacteria are generally beneficial and necessary for a healthy human digestive tract. *Escherichia coli* is a pathogen that can cause diseases such as diarrhea and other intestinal tract diseases, transmission via contaminated water or food, or contact with animals or people (Nurisyah et al., 2020).

The benefits of oranges apart from being antibacterial also function as antioxidants. Currently, the utilization of natural ingredients from citrus sources which contain vitamin C as a source of antioxidants has been developed. Antioxidants are used to protect the skin from oxidative damage so as to prevent premature aging. Natural ingredients are preferable to synthetic ingredients when creating cosmetics. Synthetic materials can have negative side effects and even harm the skin's natural form (Wulandari & Idiawati, 2013). Antioxidant intake is obtained orally or topically by rubbing it on the skin. One way to produce products with high selling value as well as good creativity and low capital is by processing waste in the surrounding environment, besides being able to help reduce environmental pollution, also in this way, products that have high selling value and are environmentally friendly. Environment can be produced while maintaining environmental sustainability. One of the environmental wastes that will be used as a product that has a fairly high selling value and is friendly to the environment is orange peel waste (Sari et al., 2021). Orange peels are abundant, easy to obtain and their utilization is not maximized, making this waste suitable as an active ingredient in anti-aging cosmetics. Based on previous research, the peels of lime, kaffir lime, and calamansi oranges are rich in components of flavonoids, tannins, and coumarins. Plants produce a wide class of secondary metabolites known as flavonoids, which are a subclass of polyphenols. All plant parts, including the leaves, roots, wood, bark, pollen, nectar, flowers, fruits, and seeds, contain these substances. Free radicals can be neutralized by flavonoids, which also prevent lipid oxidation (Warsito et al., 2017). This supports previous research which showed that the ethyl acetate extract of lime peel waste had an IC50 value of 0.492 mg/mL (Shakya et al., 2019).

Determination of antioxidant capacity is an important indicator in classifying how much antioxidant activity there is in the sources tested. The antioxidant activity of a compound can be evaluated from the antioxidant effect to control the degree of oxidation. Antioxidants can reduce free radicals through two mechanisms, namely, Single Electron Transfer (SET) and Hydrogen Atom Transfer (HAT) (SET). Antioxidants contribute hydrogen atoms to the HAT process, which stabilizes free radical speciesso they don't multiply. In the SET mechanism, free radicals are reduced through electron donation from compounds. Determination of antioxidant capacity is an important indicator in classifying how much antioxidant activity there is in the sources tested. The antioxidant activity of a compound can be evaluated from the antioxidant effect to control the degree of oxidation. Antioxidants can reduce free radicals through two mechanisms, namely, Single Electron Transfer (SET) and Hydrogen Atom Transfer (HAT) (SET). Antioxidants contribute hydrogen atoms to the HAT process, which stabilizes free radical species so they don't multiply. In the SET mechanism, free radicals are reduced by donating electrons from compounds (Yuliani, 2015).

The antioxidant and essential oil from orange peel has antimicrobial properties is influenced by the composition of the oil. Differences in plant parts used as raw materials show differences in antioxidant and antibacterial activity. Essential oil from fresh orange peel has the strongest antioxidant and antibacterial activity compared to twig and leaf oil from orange. This is due to the higher monoterpene hydrocarbon content (pinene, sabinene, myrcene, limonene) in fruit peel oil compared to leaf and twig oil (Ulya et al., 2019). Therefore, this scoping review study aims to determine the potential of the essential oils of kaffir lime, lime, and calamansi orange peels as antioxidants and antibacterials.

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METHOD

Research Design

This study uses the type of scoping review research. The literature search uses articles from 2012 to 2022, using restrictions on articles in English and Indonesian, free full text, data from the last 10 years. The articles obtained were taken from the electronic database Atlantis Press, PubMed, ScienceDirect, and the Google Scholar search engine.

Population and Samples

In the search for articles identified 2 articles from the Atlantis Press database, 5 articles from the PubMed database, 12 articles from the ScienceDirect database, and 55 articles from the Google Scholar search engine, after being filtered for relevance, 65 articles were obtained. Then further article screening was carried out to find the right and complete reference regarding the potential of the essential oils of Purut, Lime, and Calamansi Orange peels as antioxidants and antibacterials and obtained 8 articles that would be used for the scoping review. The authors screened the titles and abstracts of all articles to serve as inclusion criteria. Full text studies were retrieved and reviewed based on these criteria. Thus leaving 8 articles for the final review.

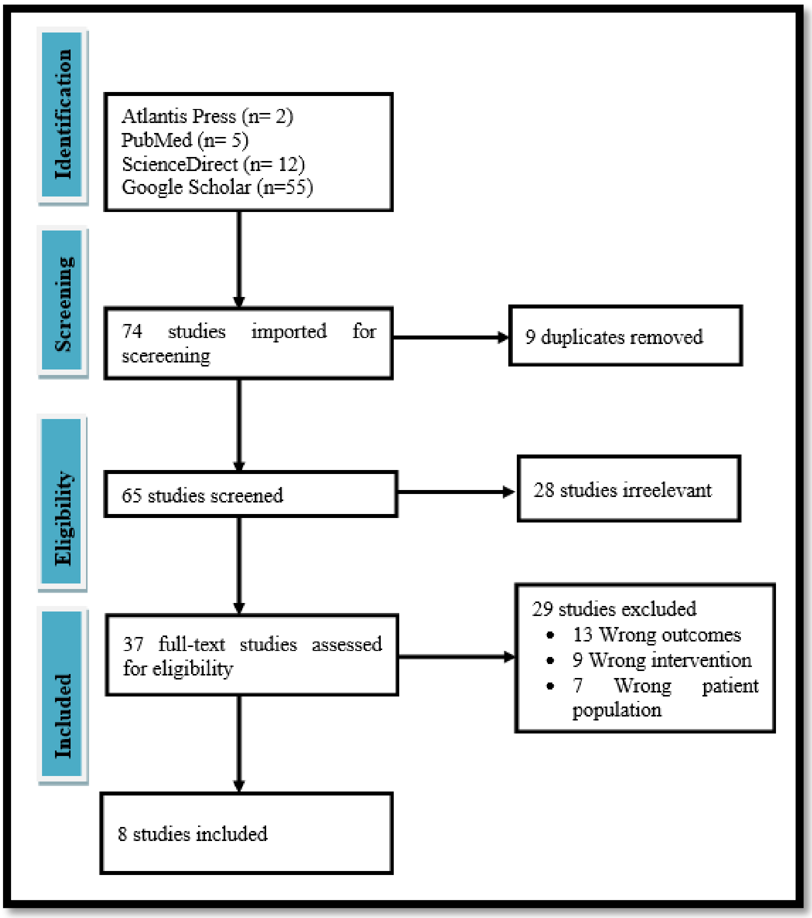

Figure 1. PRISMA Flowchart Preparation Steps.

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Instruments

Table 1. Framework Research Question.

P	E	O	S
-	Kaffir lime peel Lime peel Calamansi orange peel	Antioxidant Antibacterial	Original research, published from 2012-2022

Procedures

Article inclusion criteria used: 1) articles published in English and Indonesian; 2) articles published between 2012-2022; 3) articles that discuss some of the potential of the essential oils of kaffir lime, lime, and calamansi orange peels as antioxidants and antibacterials; and 4) there are no specific target country criteria. Article exclusion criteria used: 1) opinion articles, review articles (systematic reviews and literature reviews), reports and commentaries; and 2) letters and book reviews.

The search was carried out using keywords in English (“Potential”) or (“potency”) and (“Essential oils”) or (“essential oil”) and (“Kaffir lime peel”) and (“lime peel”) and (“Calamansi orange peel”) and (“Antioxidants”) and (“antioxidants”) or (“Antibacterials”) and (“antibacterial”). While the search for keywords in Indonesian is (“*Potensi*”) *atau* (“*Efektivitas*”) *dan* (“*Minyak atsiri*”) *dan* (“*Kulit jeruk purut*”) *dan* (“*Kulit Jeruk Nipis*”) *dan* (“*Kulit Jeruk Calamansi*”) *dan* ("*Antioksidan*") *dan* ("*Antibakteri*"), then the articles were screened according to the criteria set by the researchers and in accordance with the research questions. After that, the articles are sorted to ensure that no articles with the same title are discovered. The content was then arranged according to preset inclusion and exclusion criteria. Articles that include only the abstract will be eliminated. In order to obtain articles that will be analyzed.

Data Analysis

After that, the retrieved articles are obtained. Articles are extracted based on the article's author, nation, publication year, the number of samples utilized, the measuring instruments employed, the findings of the research done, and the article database.

RESULTS AND DISCUSSION

Extraction Data

Table 2. Extraction Data.

No.	Author/Year	Sample	Result
1	(Mohideen et al., 2022).	Kaffir lime peel is used to extract essential oil. Two gram-positive bacteria (*Staphylococcus aureus* ATCC 25923 and *Staphylococcus epidermidis* ATCC 12228) and two gram-negative bacteria (*Staphylococcus epidermidis* ATCC 12228), (*Escherichia*	Gram-positive bacteria were more sensitive to essential oils (mean diameter of the inhibition zone; *S. aureus* = 19.3 ± 1.5 mm and *S. epidermidis* = 19.3.0 ± 0.6 mm). Gentamicin was found to be most effective against all bacteria tested (inhibition zone diameter greater than 15 mm). Streptomycin resistance was only found in *S. epidermidis*. Kaffir lime

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No.	Author/Year	Sample	Result
		coli ATCC 25922 and Shigella dysenteriae ATCC 13313).	essential oil was found to have antibacterial activity. Kaffir lime essential oil has the potential to be used as an antibacterial agent in various applications.
2	(Nurisyah et al., 2020).	Lime peel with extraction is done via fractionation with ethyl acetate solvent after maceration with a 96% ethanol solvent. Extract of ethyl acetate which is a cream preparation with an extract concentration of 3%, 6%, and 9%.	The results showed that the physical properties of the cream complied with the physical stability requirements based on organoleptic test parameters, homogeneity, pH and viscosity. Likewise, the antioxidant activity test of the cream against DPPH free radicals showed that formula 1 with an extract concentration of 3% gave an average IC50 value of 28.24 mg/ml; formula 2 with an extract concentration of 6% gave an average IC50 value of 22.97 mg/ml; and formula 3 with an extract concentration of 9% gave an average IC50 value of 14.80 mg/ml. Lime peel extract is formulated into a cream preparation with type M/A. Formula 3 with an extract concentration of 9% is the best formula with the greatest antioxidant activity.
3	(Kindangen et al., 2018).	Calamansi orange peel against *Staphylococcus aureus* and *Escherichia coli* extract concentrations of 5%, 25%, 50%, and 100%.	The outcomes demonstrated that the volatile oil of calamansi orange peel has an average diameter of 11.16 mm in *Staphylococcus aureus* and 13.33 in *Escherichia coli*.
4	(Mirah et al., 2020).	Ethanol extract from lime peel and seeds.	According to the study's findings, lime peel (20.16 ± 0.23 g/mL) has a higher IC50 value than lime seeds (14.09 ± 0.88 g/mL). The tyrosine enzyme's IC50 value was lower in lime peel (34.23 ± 0.93 g/mL) than in lime seeds (67.88 ± 3.73 g/mL). Therefore, lime peel has the potential to become an inhibitor of the tyrosinase enzyme and has antioxidant potential.
5	(Husni et al., 2021).	Calamansi fruit peel and leaf essential oil. Isolated by water distillation and analyzed for chemical compounds by GC-MS.	The GC-MS results showed that the main components of essential oil in Calamansi rind were D-limonene (29.52%), (R)-(+)-citronellal (13.76%), 3-isopropenyl-5,5-dimethyl -citronellol (6.90%), cyclopentene (8.88%), -terpinene (7.30%), and -terpineol (4.61%) citronellol (25.74%), citronellol (12.94%), 3-carene (8.43), and -phellandrene (4.89%) are the main compounds in calamansi leaves. MRSA, *S. aureus*, *S. mutans*, *P. aeruginosa*, and *E. coli* bacteria were all susceptible to the antibacterial effects of the calamansi fruit peel and leaf essential oil. The diffusion

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No.	Author/Year	Sample	Result
			technique revealed that the fruit peel essential oil passed the antibacterial activity testhad moderate to strong inhibition. The dilution antibacterial activity test revealed that fruit peel essential oil has the lowest MIC was a concentration of 0.39%.
6	(Warsito et al., 2017).	Twigs, leaves and peel of kaffir lime are the main components of citronellol. Antioxidants were by measuring how well they can snare DPPH radicals, they are examined. *E. coli*, a gram-negative bacteria, is used to gauge an antimicrobial agent's effectiveness. The chemical makeup of kaffir lime oil was examined using Gas Chromatography-Mass Spectrometry (GC-MS).	The IC50 values for kaffir lime oil from peel, leaves, a mixture of twigs, and citronella were 6.43 (L/mL), 6.83 (L/mL), 9.48 (L/mL), and 10.1 (L/mL), respectively. Kaffir lime oil from fruit peels shown the highest effectiveness in preventing the development of *Escherichia coli* bacteria, with a MIC value of 12.5 (L/mL), when compared to other kaffir lime oils and their primary component citronellal. The primary constituents of kaffir lime oil from peel were -pinene, limonene, citronellal, and terpinen-4-ol, according to GC-MS analysis, while citronellal and citronellol were the main constituents of kaffir lime oil from a mixture of leaves and twigs.
7	(Nabilah et al., 2020).	The peel of the calamansi orange (*Citrus microcarpa* Bunge) obtained 500 mg 200 grams of simplicia were weighed for the distillation process after the drying process.	The essential oil of the peel of the calamansi orange (*Citrus microcarpa* Bunge) is an effective antibacterial for mouthwash which can prevent the occurrence of flak and dental caries.
8	(Liew et al., 2020).	Nanoemulsion of lime essential oil from using the spontaneous emulsification method, lime (*Citrus aurantifolia*), kaffir lime (*Citrus hystrix*), and lime (*Citrofortunella microcarpa*) were created. Particle size, Polydispersity Index (PDI), turbidity, morphology, and antibacterial characteristics were also measured.	Transmission electron microscopy micrographs reveal spherical particles of oil with various sizes. By displaying the biggest diameter of the inhibitory zone, freshly made nanoemulsion calamansi orange essential oil was the most efficient against *Escherichia coli*, *Salmonella* spp., and *Staphylococcus aureus* (8.34, 7.71, and 9.98 mm, respectively). After one month of storage at room temperature, the antibacterial activity of an essential oil nanoemulsion decreased the least. Essential oil nanoemulsions have enormous potential for use as flavoring and antibacterial components in water-based food and beverage products.

Mapping Tematis

In this step, where the author classifies an interesting theme study findings reviewed in the article, the theme mapping is as follows:

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Tabel 3. Mapping Tema.

Theme	Sub Themes/Researchers
Essential oils of Kaffir lime, Lime, and Calamansi oranges as antioxidants and antibacterials.	1. Orange essential oil antioxidant (Warsito et al., 2017). 2. Lime essential oil antioxidant (Mirah et al., 2020; Nurisyah et al., 2020). 3. Calamansi orange essential oil antioxidant (Husni et al., 2021). 4. Antibacterial kaffir lime essential oil (Liew et al., 2020; Mohideen et al., 2022; Warsito et al., 2017). 5. Lemon essential oil antibacterial (Liew et al., 2020). 6. Calamansi orange essential oil antibacterial (Husni et al., 2021; Kindangen et al., 2018; Nabilah et al., 2020).

Essential Oil of Kaffir Lime Peel as Antioxidant and Antibacterial

An antioxidant can be defined as various types of substances that are able to complete against oxidized substrates at a relatively low concentration level and inhibit or prevent the oxidation of the substrate. Antioxidant activity test of four types of kaffir lime oil samples, kaffir lime oil fraction, major components and BHT control was characterized by a decrease in activity (absorbance) of DPPH radicals (Warsito et al., 2017).

The peel of kaffir lime (*Citrus hystrix* DC.) is underutilized because it is often considered as waste, even though the kaffir lime peel contains many bioactive compounds such as phenolics, flavonoids, limonins, furanocoumarins, and pectin which are even higher than the juice and pulp. These compounds are antioxidants that can inhibit free radicals, prevent oxidative damage, cell death, so that they have a preventive effect on various diseases. This study aims to determine the effect of the type of solvent on the antioxidant and anti-inflammatory activity extracted from kaffir lime peel (*Citrus hystrix* DC.) and the effect of the concentration of kaffir lime peel extract (*Citrus hystrix* DC.) extracted from the selected solvent on antioxidant and anti-inflammatory activity in vitro. In this study, kaffir lime peels were dried and then ground and sieved using an 80 mesh sieve to become flour (Herman & Jaya, 2020).

The analyzes carried out included water content and total phenol for flour and fresh samples, as well as antioxidant and anti-inflammatory activity for flour samples which included solvent optimization and testing with the best solvent. The method used to test the water content is thermogravimetry, total phenol with Folin-Ciocalteu, antioxidant activity with DPPH, and anti-inflammatory with protein denaturation. Based on the results of the analysis, the water content of fresh kaffir lime peels was 77.14Â ± 0.30% and kaffir lime peel powder was 13.16Â ± 0.06. The total phenol content on a dry basis in fresh samples was 30.05Â ± 2.09 mg GAE/g sample and 19.41Â ± 0.41 mg GAE/g sample in flour samples. The type of solvent and the resulting concentration of the selected solvent affect the antioxidant and anti-inflammatory activity of kaffir lime peel extract. The best solvent for determining antioxidant and anti-inflammatory activity in kaffir lime peels is methanol which produces % RSA at a concentration of 100 mg/ml of 89.58Â ± 0.95% and % inhibition of protein denaturation at a concentration of 50 mg/ml of 60.81Â ± 1.01%. The results of concentration

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variations showed that the concentration of kaffir lime peel of 50 mg/ml was able to act as an antioxidant and anti-inflammatory because it had a percentage of RSA and protein denaturation inhibition above 50%. The percentage of RSA in kaffir lime peel at a concentration of 50 mg/ml was 68.65Â ± 0.32% and the percentage of inhibition of protein denaturation was 62.45Â ± 7.04% (Yuliani, 2015).

Based on the results of the study, it was shown that the methanol extract was classified as a strong antioxidant compound, the ethyl acetate extract was classified as an antioxidant compound in the moderate category, while the n-hexane extract was classified as a weak antioxidant category. The methanol extract and ethyl acetate extract had stronger antioxidant activity than the n-hexane extract, presumably due to the presence of flavonoids and polyphenolic compounds according to the results of phytochemical testing (Muzuka et al., 2018).

One of the most frequent causes of common bacterial infections, such as cholecystitis, bacteremia, cholangitis, urinary tract infections, traveler's diarrhea, and other clinical illnesses like meningitis in young children and pneumonia, is *E. coli* bacteria. Infections in the urinary tract are primarily caused by *E. coli* obtained from hospitals and the community. The cause of infection in 50% of women with urinary tract infections, 4% of cases of diarrhea, and 12-50% of nosocomial infections is *E. coli*. Meningitis in infants caused by *E. coli* is as much as 8%, while the mortality rate and incidence rate related to bacteremia by *E. coli* is the same as the mortality rate and incidence rate of aerobic Gram-negative bacilli (Chandra, 2022). Infections caused by bacteria are usually treated with antibiotics which can be obtained from chemical synthesis or microorganisms. To obtain other sources of antibacterial activity, many plants have been studied in vitro including kaffir lime. Several researchers have tested kaffir lime's ability to fight off a variety of germs (Dewi, 2019).

Methanol extract of kaffir lime peel and several Gram-positive and Gram-negative bacteria are resistant to some of its fractions' moderate to potent antibacterial activities. *S. aureus* is more susceptible to ethyl acetate extract and kaffir lime peel essential oil than *E. coli*. Kaffir lime peel and leaf essential oils and ethanol extracts have antibacterial action against several enterobacterial and Salmonella species (Yuliani, 2015). The results indicated that the essential oils of kaffir lime peel and leaves were able to inhibit the growth of 5 strains of Propionibacterium acne. This study aims to measure the antibacterial activity of kaffir lime leaf essential oil against *Staphylococcus aureus* and *Escherichia coli* and determine the class of compounds in the essential oil that have antibacterial activity (Siregar et al., 2020).

Kaffir lime peel (*Citrus hystrix* DC.) contains essential oils, besides that the leaves, fruit peels and stem bark of the kaffir lime plant contain active substances of flavonoids, phenolics, and terpenoids. The kaffir lime peel contains many compounds from the class of flavonoids, steroids, saponins, and tannins. The peelpart is usually used as a medicine for boils, heartburn, inflammation of the peel, scaly peel and peeling peel. Kaffir lime peel has a content that shows antimicrobial activity and is also used as an antibacterial and can inhibit the growth of *Staphylococcus aureus* infection (Sari & Asri, 2022).

Lime Peel Essential Oil as Antioxidant and Antibacterial

Lime peel is rich in flavonoids, tannins, and coumarins. Plants produce a wide class of secondary metabolites known as flavonoids, which are a subclass of polyphenols. All plant parts, including the leaves, roots, wood, bark, pollen, nectar, flowers, fruits, and seeds, contain these substances. Free radicals can be neutralized by flavonoids, which also prevent lipid oxidation. This supports research conducted by Nurisyah & Asmawiyah (2018) showing that the ethyl acetate extract of lime peel waste has an IC50 value of 0.492 mg/mL. Another study conducted by Hindun et al. (2017) stated that the ethanol extract of lime peel has potential as a tyrosinase inhibitor with an IC50 value of 42.11 mg/mL. This is the reason why the formula made in this study uses ethyl acetate extract from lime peels (Kurniandari et al., 2015).

Antioxidants are compounds that stop free radicals from oxidizing while also delaying, slowing down, and preventing the oxidation process. Free radicals' lack of electrons is compensated for by antioxidants, which stabilize free radicals. The DPPH (1,1 Diphenyl-2-picrylhydrazil) test method is the most popular in vitro test method due to its ease, accuracy, efficiency, simplicity, and speed in determining antioxidant activity. The IC50 value for DPPH free radicals, which tries to identify the concentration of preparations that may inhibit 50% of DPPH free radicals, is used to determine antioxidant activity (Ulya et al., 2019). A stable free radical at room temperature is called DPPH and is often used to evaluate the activity of antioxidant compounds or natural products. The DPPH solution is violet in color which gives a strong absorption at a wavelength of 517 nm. Antioxidant compounds cause the electrons of the DPPH free radical to become a pair which is marked by a loss of color that correlates with the number of electrons absorbed (Sari & Asri, 2022).

Topical formulations containing antioxidants are expected to be able to protect the skin from the unwanted effects of free radicals from the sun, whereas cream dosage forms are more practical. Cream is a semisolid preparation, in the form of an emulsion with an air content of not less than 60% and is intended for external use. Cream preparations are preferred because they are more pleasant, easy to spread evenly, and practical to use (Kurniandari et al., 2015).

According to studies on the antioxidant activity of lime peel extract cream (*Citrus aurantifolia*) against DPPH free radicals, formulas 1 and 3 with 3%, 6%, and 9% extract concentrations, respectively, gave an average IC50 value of 28.24 mg/ml, 22.97 mg/ml, and 14.80 mg/ml, respectively. The results of testing the activity of antioxidant creams against DPPH free radicals for each formula F1, F2, and F3 showed that the higher the concentration of lime peel extract, the stronger the antioxidant activity of the formula. This is evidenced by the smaller IC50 value, which indicates that the concentration of formula 3 cream which can bind 50% DPPH is lower than formula 1 and formula 2. Based on the IC50 F3 value, it shows that the resulting cream preparation still has antioxidant activity that is not active, so to increase the antioxidant activity of cream preparations it is necessary to combine it with extracts of other ingredients that have strong antioxidant activity. Based on the results of this study indicate that lime peel extract cream can be developed as an antioxidant cream. Considering that lime peel is a large amount of waste in the environment, its use is only to obtain essential oil as a flavor enhancer but also provides more benefits (Parhusip, 2017).

The use of lime peel extract (*C. aurantifolia*) has been shown to inhibit the growth of *S. dysenteriae* as indicated by the formation of an inhibition zone. The inhibition zone formed against *S. dysenteriae* is due to the presence of secondary metabolites that have antibacterial activity in lime peel. Secondary metabolites are compounds that have the ability to inhibit bacterial growth. Lime peel contains secondary metabolites including saponins, flavonoids, alkaloids, and tannins (Farida et al., 2021). The content of flavonoids in lime peel namely hesperidin, naringin, hesperetin, and naringenin. Lime peel also contains secondary metabolites in the form of essential oils consisting of limonene, linnalool, citronellal and citronellol which have antibacterial properties. Phenolic group compounds such as saponins, flavonoids, tannins, and alkaloids have bacteriostatic properties. Each secondary metabolite compound has a different mechanism in inhibiting bacterial growth (Parhusip, 2017).

Secondary metabolite compounds in inhibiting bacterial growth begins with the destruction of the cell wall. Compounds that play a role in damaging the cell wall include flavonoids, phenols, and alkaloids. Saponins, flavonoids, and terpenoids work by changing the permeability of the cytoplasmic membrane of bacterial cells. The next mechanism is protein denaturation by phenolic compounds. Further destruction is carried out by tannins, alkaloids, and phenolic compounds by inhibiting enzymatic processes in cells causing bacterial cells to die (Siregar et al., 2020).

Flavonoid compounds are able to penetrate bacterial cells' peptidoglycan prevents the cell layer from forming fully. Additionally, flavonoids have the capacity to combine with extracellular proteins to produce complex chemicals that harm the bacterial cell membrane. Another mechanism of flavonoids is that they inhibit the work of the DNA gyrase enzyme causing the process of protein synthesis to be inhibited and bacterial cells unable to replicate (Jirna et al., 2017).

Tannin secondary metabolites are able to prevent the availability of substrates needed by bacterial cells. Tannin compounds also interfere with cell permeability, cell membrane integrity, and protein synthesis. Tannins also have an antimicrobial effect by interfering with the metabolism of bacterial cells causing the bacteria to die. Alkaloids are also contained in lime peels. The mechanism of alkaloid inhibition is by interfering with the synthesis of nucleic acids and proteins, modifying damaging the cell membrane and bacterial cell wall, increasing the permeability of the bacterial cell membrane, and interfering with the metabolism of the bacterial cell. Alkaloids also work by preventing the creation of proteins and nucleic acidsby inhibiting the process of DNA replication causing bacteria to be unable to carry out cell division. Lime peel also contains essential oil compounds. The presence of essential oils can increase the permeability of cell membranes causing the release of enzymes and inhibition of

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bacterial respiration processes. Essential oils are also able to disrupt the structure of bacterial proteins causing denaturation of proteins (Sari & Asri, 2022).

Calamansi Orange Peel Essential Oil as Antioxidant and Antibacterial

One of the many groups of compounds found in calamansi oranges are flavonoid compounds. Flavonoid compounds have antioxidant activity. Flavonoids that have antioxidant properties include flavonols, kaatexins, flavones, and chalcones. In addition, Calamansi Orange (*Citrofortunella microcarpa*) contains vitamin C which can act as an antioxidant. The content of vitamin C has a good effect in counteracting free radicals that can damage cells or tissues (Kindangen et al., 2018).

The DPPH radical absorption method is used to measure the antioxidant activity of the Calamansi orange peel (*Citrus microcarpa*) since it is a quick, easy procedure that only requires a small amount of sample. Antioxidant activity is measured by the proportion of DPPH that is inhibited. The difference in absorbance between the sample's absorbance and the DPPH absorbance, as determined by a UV-Vis spectrophotometer, was used to calculate the percentage of inhibition. This shows that the percentage of inhibition in gallic acid is greater than that of the calamansi orange extract. However, both have the same correlation, namely the greater the concentration of the sample, the % inhibition increases. Meanwhile, the measurement of percent (%) inhibition was carried out on samples of Gerga Oranges. When compared with the results of calamansi oranges, the percentage value of inhibition in gerga oranges showed the same correlation with increasing concentrations. Calamansi orange peel is rich in essential oil and 0.15% ascorbic acid (Tutuarima, 2020).

The skin of the calamansi orange (*Citrus microcarpa*) contains flavonoids. Flavonoids act as antibacterial and antioxidants which are effective in overcoming free radicals that have a bad effect on the skin. Flavonoid compounds have antioxidant activity. The antioxidant class of flavonoids includes flavones, flavonols, kaatexins, and chalcones. Numerous additional substances, including naringin, neohesperidin, neoeriocitrin, and poncirin, contribute to the bitterness of oranges. Flavanones are highly significant citrus flavonoids (Veranita et al., 2021).

Antioxidant compounds in calamansi orange peel can inhibit by reacting with reactive free radicals to create relatively stable unreactive free radicals, oxidation is accomplished. Phenolic compounds and flavonoids are sources of natural antioxidants which are usually found in plants. In addition, antioxidants have the ability to donate electrons, bind and end deadly free radical chain reactions. The antioxidants used are then recycled by other antioxidants to prevent them from becoming free radicals (for themselves) or remaining in that form but with a different structure (Nabilah et al., 2020).

The essential oil the primary terpenoid group constituents are found in the peel of the calamansi orange. These substances have been demonstrated to have antibacterial effects by blocking the production of nucleic acids, polysaccharide walls, and ergosterol in cell membranes (Palma et al., 2019). Antibacterial activity in the crude extract and essential oil of the calamansi orange peel is due to the

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presence of phytochemical compounds in the calamansi orange peel, such as flavonoids, alkaloids, tannins, saponins, and terpenoids. These secondary metabolites will interfere with the cellular membrane's permeability or its component parts in such a way that shrinkage occurs or the components in the cell come out, can even inhibit the formation of the cell membrane and cause inhibition of bacterial growth. By altering the permeability of the bacterial cell wall and preventing bacterial motility, flavonoid chemicals can prevent bacterial growth. H+ ions from phenolic chemicals and their derivatives (flavonoids) attack the polar group (phosphate group) during the disintegration of the cytoplasmic membrane, causing the phospholipid molecule to disintegrate into glycerol, carboxylic acid, and phosphoric acid. Because of this, phospholipids are unable to keep the cytoplasmic membrane in its proper form, which causes the membrane to leak and cause the bacteria to develop slowly or even die (College et al., 2017).

Alkaloids work as antibacterials by interfering with the peptidoglycan's constituent parts in bacterial cells, which prevents the cell wall layer from forming properly and results in cell death. Tannins have the ability to inhibit bacterial growth because they have antibacterial properties that precipitate proteins and cause the membrane of the bacterial cell to contract, decreasing cell permeability. The mechanism of action of saponins causes the release of numerous significant components of the bacterial cell, including proteins, nucleic acids, and nucleotides. Saponins function as an antibacterial by interfering with the stability of the bacterial cell membrane, leading to bacterial cell lysis. Terpenoid compounds are also known to be active against bacteria, the mechanism that occurs involves the breakdown of the cytoplasmic membrane from the role of hydrophobic components. The mechanism of inhibition of bacterial growth by terpenoid compounds is that terpenoids porins (transmembrane proteins) on the bacterial cell wall's outer membrane react with these substances to produce robust polymeric bonds, which harm the porin. Porin damage reduces the permeability of the bacterial cell wall, which will leave the cells lacking in nutrients and prevent or cause the death of bacterial growth. Porin damage is the entry and exit site for chemicals (Nabilah et al., 2020).

There are two main components found in the peel of the calamansi orange, namely monoterpenes and sesquiterpenes. These two components are known to have antimicrobial properties, especially as antibacterial. The content of monoterpenes in essential oils can inhibit the growth of bacteria. Most monoterpenes are lipophilic. Monoterpenes have been shown to be more likely to diffuse into the structural phase of the bacterial membrane than the aqueous phase. The accumulation of monoterpene molecules into the membrane phase will cause the membrane to expand (swell), increase membrane permeability, thereby damaging the membrane that binds to bacterial cell proteins (Husni et al., 2021).

CONCLUSION

Based on the scoping review that has been done, it can be concluded that the essential oils from the rind of kaffir lime (*Citrus hystrix* DC.), lime peel (*Citrus aurantifolia*), and calamansi orange (*Citrus microcarpa* Bunge.) have antioxidant and antibacterial activity. The higher the concentration of essential oils from kaffir lime peel, lime peel, and calamansi orange peel, the greater the antioxidant and antibacterial activity produced.

SUGGESTION

Further research is needed to optimize the antioxidant and antibacterial activity of extracts of kaffir lime peel, kaffir lime peel, and calamansi orange peel by increasing the concentration or by combining them with antioxidant compounds from other natural extracts.

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