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Update on Non-Communicable Disease: Global Perspective on Health Challenges and Innovation

Phytochemical Screening and Potency of Mango Peel Extract (*Mangifera indica* L.) var. Gedong Gincu in Inhibiting the Growth of *Staphylococcus aureus*

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ABSTRACT

Background: *Staphylococcus aureus* is a pathogen most of which develop into *Methicillin-resistant Staphylococcus aureus*. To prevent bacterial resistance, herbal medicine is needed. Mango plants have secondary metabolite compounds that can inhibit bacterial growth. Gedong gincu mango is a specific mango variety that grows widely in Cirebon district. There has been no research that knows the secondary metabolite content and its potential as an antibacterial, especially the peel part which only becomes waste.

Aims: To find out the chemical compounds contained and determine the potential of mango peel extract (*Mangifera indica* L.) var. gedong gincu in inhibiting the growth of *Staphylococcus aureus*.

Methods: This research is an experimental with a posttest only control group design. Phytochemical screening test employed a qualitative method. The extract was made using the maceration method with 70% ethanol solvent. Antibacterial testing with well diffusion method, and given four treatment concentrations (W/V), namely 25%, 50%, 75%, 100%. The measurement on the inhibitory zone after 24 hours was done at temperature of 37°C.

Results: Gedong gincu mango peel extract contains secondary metabolite compounds flavonoids, tannins, saponins, and steroids. The inhibitory activity of gedong gincu mango peel extract with a concentration of 25% gedong gincu mango peel extract has an average inhibition zone of 11,55 mm, 50% average inhibition zone 13,55 mm, 75% average inhibition zone 14,88 mm, and 100% average inhibition zone 16,22 mm in inhibiting the growth of *Staphylococcus aureus* p(<0.05).

Conclusion: Mango peel extract var. gedong gincu with a concentration of 25% has the potential to inhibit the growth of *Staphylococcus aureus* bacteria.

Keywords: *Mango peel gedong gincu; Phytochemical screening; Inhibition potency; Staphylococcus aureus.*

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1. Introduction

Staphylococcus aureus is a Gram-positive bacterium that often causes infectious diseases in humans and animals, including skin infections, endocarditis, pneumonia and causes sepsis (Wu *et al.*, 2022). The use of antibiotics, both bactericidal and bacteriostatic, has long been used as a drug to overcome *Staphylococcus aureus* infection which is known to have a very good success rate (Rossiter *et al.*, 2017). The inappropriate use of antibiotics leads to a high prevalence of antibiotic-resistant *Staphylococcus aureus* (MRSA) and thus becomes a particular concern for global health (Chen & Huang, 2014). Based on data from the Centers of Disease Control and Prevention, the incidence of antibiotic resistance has increased to 2.8 million infections per year, resulting in 35,000 deaths every year in the United States (Jain *et al.*, 2022) and in Indonesia the resistance of *Staphylococcus aureus* bacteria increased by 77.20% in 2023 (Fitrandi *et al.*, 2023).

The use of plants as medicinal ingredients has been carried out by the people in Indonesia for a long time, because of the abundant natural wealth so that traditional medicine has become part of the cultural heritage and knowledge in descending ways that continue to be preserved until now, one of which is the mango fruit (Noviyanty, 2020). Special of phytochemical content and nutritional components belonging to mango (*Mangifera Indica* L.) is known to have effects as antioxidants, antidiabetics, anti-inflammatory, antimicrobial, antifungal, and dyslipidemia (Nadhifah 2021). Other research explain the activity of secondary metabolite compounds such as flavonoids, tannins, steroids, triterpenoids, alkaloids, and phenols can significantly inhibit bacterial growth (Lobiuc *et al.*, 2022; Mahizan *et al.*, 2019).

In West Java, there is a mango plant with a typical mango varieties, namely the mango (*Mangifera Indica* L.) var. gedong gincu which is still included in the Anacardiaceae family (Yadav *et al.*, 2018), with a highest production center in Cirebon district (Badan Pusat Statistik, 2021). Based on the results of previous research, it is known that the mango plant (*Mangifera Indica* L.) contains antioxidants found in stems, leaves and fruits, especially the pulp and skin (Suwardike *et al.*, 2018). Research on the potential of mango (*Mangifera Indica* L.) var. gedong gincu which has been carried out by Aqyun 2019 obtained the results of Gedong Gincu mango leaf extract has an anti- hyperglycemic effect. Mango plants also have effects as a lowering of uric acid levels, anti-inflammatory, and antibacterial (Ifmaily *et al.*, 2023; Brajawikalpa *et al.*, 2024). Another potential of Gedong Gincu mango, both on the peel and leaves, is in the wound healing process which was tested on white rats (Fathunnisa *et al.*, 2024 and Loren *et al.*, 2024).

Research on the content and potential of gedong gincu mango fruit has not been widely carried out, one of which is the potential of mango skin (*Mangifera Indica* L.) var. gedong gincu especially as an antibacterial. The management of mango skin waste that has not been optimal should be utilized as added value and income for the community (Mardhatilla *et al.*, 2021). Apart from the economic aspect, the utilization of mango skin waste can also be used as herbal therapy where the secondary metabolite content contained in it can be used as an alternative therapy in treating infections caused by *Staphylococcus aureus* bacteria.

Based on the explanation above, this study was conducted to determine other potentials of gedong gincu mango, with the aim of analyzing the secondary metabolite compounds contained and active as antibacterials from mango skin (*Mangifera Indica* L.) var. gedong gincu so that it can then be used as herbal therapy in the community.

2. Methods

Study design/ Research procedures

This research was conducted at the Research Laboratory of the Faculty of Medicine, Swadaya Gunung Jati University. This type of research is true experimental research using *a post-test only control group design*. Using 6 groups, consisting of 4 treatment groups that were given mango peel extract (*Mangifera indica* L.) var. gedong gincu with concentrations of 25%, 50%, 75%, 100%, and also 2 control groups, namely, positive control group using Amoxicillin because it has broad spectrum antibacterial activity, is bactericidal and effective against Gram positive bacteria and is a standard antibiotic (Clinical Laboratory Standart Institute, 2024) and negative control group using Dimethyl Sulfoxide 10% because it does not affect the test results for antibacterial activity. The number of repetitions of each group is 4 times obtained based on the calculation of Federer's formula.

Mango plant samples (*Mangifera indica* L.) var. gedong gincu were obtained from mango farmers in Sedong Lor Village, Sedong District, Cirebon Regency. The sampling method was carried out using simple random sampling. The inclusion criteria were *Staphylococcus aureus* bacteria that had been equalized to the Mc Farland standard of 0.5 and grew on Mueller Hinton Agar media. *Staphylococcus aureus* is a clinical isolate that is used as a standard laboratory testing with a well diffusion method (Clinical Laboratory Standart Institute, 2024). The exclusion criteria were *Staphylococcus aureus* bacteria contaminated with other microorganisms.

Measurements

The procedure of this research includes:

a. Plant Determination

Plant determination was carried out at the Biology Laboratory of the Faculty of Mathematics and Sciences, State University of Semarang with number 55/UN.37/SHP/Lab. Plant Taxonomy/IV/2024.

b. Extraction

Mango peel simplicia powder (*Mangifera indica* L.) var. gedong gincu then macerated using 70% ethanol in a ratio of 1:5. The maceration process is carried out for 3 days in a closed container. After 3 days, it is filtered using filter paper and filter buchner. Macerate is evaporated with a rotary evaporator, then thickened on a water bath at a temperature of 40° Celsius until a thick extract is obtained

c. Calculation of % yield

The percentage of rendemen was obtained from the comparison between the weight of the extract obtained and the weight of the initial simplicia. The larger the percentage value of the rendemen obtained, the better the value of the extract produced (Depkes RI, 2000). The calculation of the percentage of rendemen can be used with the following formula:

$$\% \text{ yield} = \frac{\text{weight of extract obtained}}{\text{weight of simplicia used}} \times 100\%$$

d. Phytochemical Screening

Phytochemical screening of mango peel extract (*Mangifera indica* L.) var. gedong gincu using a qualitative method by adding reagents from each test compound and seeing the color change that occurred (Harbone, 1987). Phytochemical screening tests were carried out at the Laboratory of the Center for Biopharmaceutical Studies, LPPM – IPB University with number 405.043/LPSB IPB/V/24.

1. Flavonoids

Mango peel extract of 0.5 grams is dissolved in 2 mL of methanol p.a, heated then cooled and filtered. Filtrate is added to the H₂SO₄ reagent then in the vortex and placed on the test tube rack waiting for the reaction to be completed. The formation of red, yellow or orange color indicates the presence of flavonoid group compounds.

2. Alkaloids

Mango peel extract is 0.5 grams of extract added 1 mL of HCl 2N and 9 mL of hot and cooled aquadest. Then strain the filtrate divided 3 into tubes A, B, and C equally. Tube A is added Wagner reagent 2 drops of positive alkaloid results will form brown-black deposits. Tube B is added with *Dragendorff* reagent 2 drops of positive alkaloid results will form an orange precipitate. Tube C is added with *Mayer* reagent 2 drops of positive alkaloid results will form a white precipitate.

3. Tannins

Mango peel extract as much as 0.5 grams of extract is added to 10 mL of hot water and cooled. Filter the filtrate, drip 2 drops of FeCl₃ 1%. Tannin positive result if dark blue or greenish-black color is formed.

4. Saponins

Mango peel extract as much as 0.5 grams of extract is added to 10 mL of hot water, cooled and homogenized until foam/foam appears. Let it sit for 2 minutes then add 1 drop of HCl 2 N, homogenize again until a foam is formed that settles for 10 minutes showing positive results for saponins.

5. Quinone

Mango peel extract as much as 0.5 grams of extract added NaOH 1N is homogenized, then observe the color change. Positive results are indicated by the formation of a yellow color.

6. Steroids and Triterpenoids

Mango peel extract as much as 0.5 grams of extract is added 2 mL of ethanol then in vortex and soaked in warmed aquades then cooled and filtered. The filtrate is evaporated until thick and ether is added, 3 drops of anhydrous acid and 1 drop of concentrated H₂SO₄, observe until there is a red tinge that indicates a positive triterpenoid. Meanwhile, the positive results of steroids are formed in green.

e. Antibacterial Test

The antibacterial test process begins with sterilization using an oven for 45 minutes at a temperature of 160°C. MHA as a medium in sterilization using an autoclave at a temperature of 121°C within 15 minutes. Antibacterial testing of mango peel extract (*Mangifera indica* L.) var. gedong gincu was carried out using the diffusion method on MHA media with the addition of 50 µL of suspension of *Staphylococcus aureus* bacteria which had been adjusted to the Mc Farland standard of 0.5 and each concentration of mango peel extract (*Mangifera indica* L.) var. gedong gincu as much as 35 µL into the well diffusion. Petri dishes are wrapped in paper and incubated at 37°C for 24 hours. Bacterial growth was observed and zoned using a caliper. The bacteria were obtained from cultures available in the Research Laboratory of the Faculty of Medicine, Swadaya Gunung Jati University, which had previously been confirmed using a coagulase test.

Statistical techniques

The data obtained were statistically analyzed using IBM SPSS Statistics 22. The data was tested for normality with Shapiro Wilk was used to determine the distribution of inhibitory power from all data treatments due to the small number of samples the data distribution is normal if $p > 0.05$, then continued with the One-Way Anova test to determine the differences in the inhibitory power of each treatment group, significant results if the p value < 0.05 .

Ethical Clearance

This research has been ethically approved by the Ethic Committee of Faculty Medicine, Swadaya Gunung Jati University, with number 11/EC/FKUGJ/IV/2024, on April 8th, 2024.

3. Results**Plant Determination**

The results of the taxonomy test that has been carried out at the Biology Laboratory of the Faculty of Mathematics and Sciences, Semarang State University number 55/UN.37/SHP/Lab. Plant Taxonomy/IV/2024. showed that the sample used was indeed the mango plant (*Mangifera Indica* L.) var. gedong gincu.

Results of Calculation % Yield

The result of the large calculation of the percentage of rendemen is based on the weight of the sample obtained after the maceration process of 183 grams with the weight of simplicia obtained from 20kg of gedong gincu mango fruit of 475 grams. So that the percentage of mango peel extract (*Mangifera Indica* L.) var. gedong gincu which was obtained by 38.52%.

Phytochemical Screening

Qualitative phytochemical screening tests were carried out on mango peel extract (*Mangifera indica* L.) var. gedong gincu to show the presence of several classes of secondary metabolite compounds as seen in Table 1.

Tabel 1. Results of Phytochemical Screening Mango Peel Extract (*Mangifera indica* L.) var. Gedong Gincu

Compounds	Results	Color Change Indication
Flavonoids	Positive	Yellow – orange
Wagner		Brown-black deposits
Alkaloids	Negative	White deposits
Mayer		Orange deposits
Dragendorff		
Tannins	Positive	Blue – blackish
Saponins	Positive	Foaming
Quinone	Negative	Yellow
Steroids	Positive	Blue – greenish
Triterpenoids	Negative	Red – orange

In the phytochemical test of mango peel extract (*Mangifera Indica* L.) var. Gedong Gincu found that mango peel extract positively contained flavonoids, tannins, saponins, and steroids, but it was seen that mango peel extract (*Mangifera Indica* L.) var. Gedong Gincu did not contain quinone and triterpenoid compounds.



Figure 1. Positive flavonoids with yellow color change, positive tannins with blackish color change, positive saponins with foaming, and quinone is negative because there is a precipitate

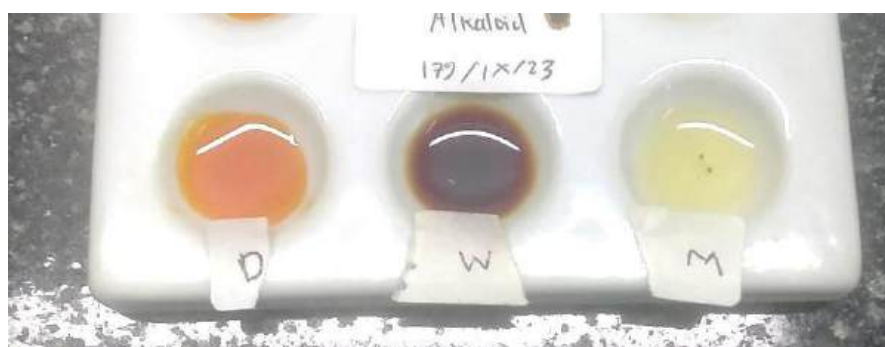


Figure 2. Alkaloid is negative in all reagents (wagner, mayer, dragendorff)



Figure 3. Positive for steroids because it is greenish in color

Antibacterial Test Results

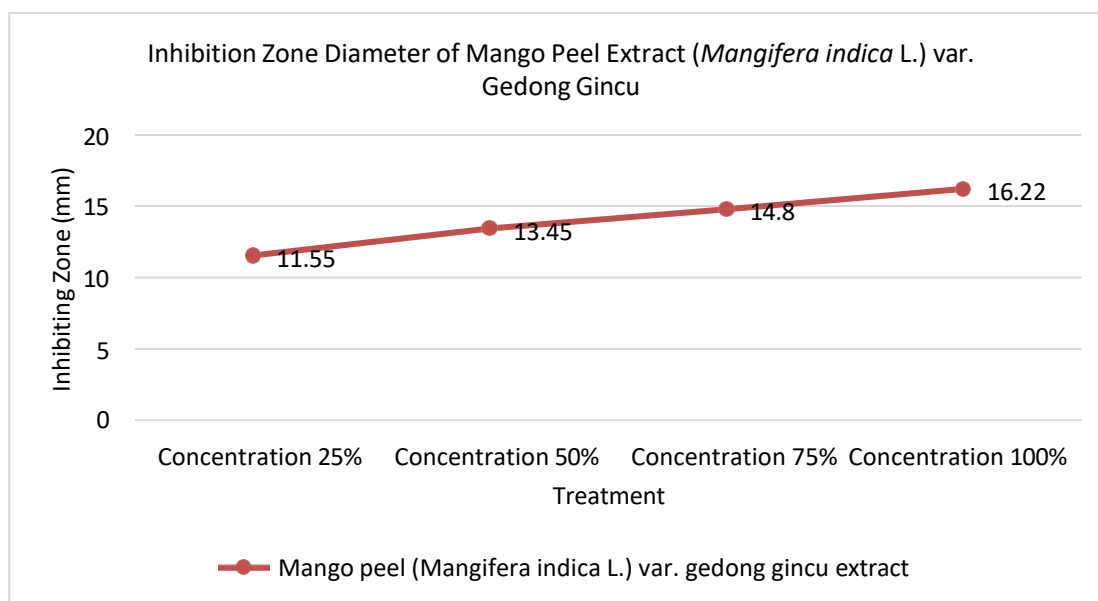


Figure 4. Average Inhibition Zone Diameter of Mango Peel Extract (*Mangifera indica* L.) var. Gedong Gincu

Based on figure 3, the 100% concentration extract group had the highest inhibition with an average of 16.22 mm, the 25% concentration extract group had the lowest inhibition with an average of 11.55 mm. The negative control uses a 10% DMSO and does not indicate any inhibition.

Bivariate analysis

Table 2. Results of One-Way Anova Test Analysis of Mango Peel Extract (*Mangifera indica* L.) var. Gedong Gincu

Group	Inhibition Zone Diameter (mm)				Average (mm)	F	Sig.
	I	II	III	IV			
25% Concentration	11.85	11.40	11.20	11.75	11.55	3281.187	0.000*
50% Concentration	12.85	13.15	14.15	13.65	13.45		
75% Concentration	14.65	14.10	14.90	15.58	14.80		
100% Concentration	16.35	16.00	16.25	16.30	16.22		
Amoxicillin (+)	35.90	36.10	35.80	36.00	35.90		
DMSO 10% (-)	0	0	0	0	0		

Significant p<0.05

The results of the One-Way Anova test obtained a significant p value (<0.05) so that it can be concluded that mango peel extract (*Mangifera indica* L.) var. gedong gincu can inhibit the growth of *Staphylococcus aureus*.

4. Discussion

This research discusses the content of secondary metabolite compounds contained in gedong gincu mango peel extract and its potential to inhibit the growth of *Staphylococcus aureus* bacteria using different concentrations. Mango peel (*Mangifera indica* L.) var. gedong gincu was determined and identified. Furthermore, extraction was carried out by the maceration method. This method was chosen to avoid damage to thermolabile compounds (Badaring et al., 2020). The solvent used for maceration is 70% ethanol which is known to produce high rendemen and produce extracts with the highest secondary metabolite content with a maceration time of 3 days (Afifah et al., 2023).

From the research results obtained from phytochemical screening, it is known that gedong gincu mango peel extract contains secondary metabolite compounds including flavonoids, tannins, saponins and steroids based on color changes when reacted with certain solvents. These results are different from previous research conducted by Fathunnisa et al. (2024) where the phytochemical screening carried out obtained flavonoids, tannins, phenols, triterpenoids, steroids, alkaloids, and saponins. This difference can occur due to differences in the level of ripeness of the mangoes used in the test (Maldonado et al., 2019). However, the content of the compounds in the gedong gincu mango peel is more complete compared to the cengkir mango skin which only contains secondary metabolite compounds such as tannins, flavonoids and phenols (Artauli S et al., 2025). The diversity of mango varieties causes differences in the secondary metabolite compounds they contain. Based on previous studies, it is known that the Arum Manis mango skin contains more complete compounds, namely flavonoids, tannins, steroids, alkaloids, saponins (Mandike et al., 2022). contained. Based on previous research, it is known that Arum Manis mango skin contains more complete compounds, namely flavonoids, tannins, steroids, alkaloids, saponins (Mandike et al., 2022).

The results of this research also show that the inhibitory activity of bacterial growth can be seen from each concentration tested, the results are in the form of an inhibition zone which is observed and measured based on its diameter. The average zone of inhibition obtained from each concentration was 11.55mm at an extract concentration of 25%; 13.45mm at 50% concentration, 14.80mm at 75% concentration; and 16.22mm at 100% concentration. The comparison of the ability of gedong gincu mango peel extract to inhibit the growth of bacteria can be said to be better than other mango varieties, it can be seen that based on previous research it is known that madu mango peel extract does not have antibacterial activity against *Escherichia coli* (Paulus KA et al., 2024), besides that based on research conducted by (Mandike et al., 2022) the results showed a strong anti-bacterial effect with the largest inhibitory zone diameter of 13.91mm against the growth of *E.coli* and 13.40mm in *S. typhi* bacteria. The secondary metabolite compounds contained in mango skin have the potential to inhibit bacterial growth (Artauli S et al., 2025).

The mango plant has pharmacological effects as an antioxidant, antidiabetic, anti-inflammatory, hepatoprotective, antihyperlipidemic, antihyperglycemic, antimicrobial, antiobesity, antifungal, antidiarrheal, etc. (Kurrota *et al.*, 2023). Based on the results of the phytochemical test, it can be known that the mango peel extract (*Mangifera Indica* L.) var gedong gincu has the potential as antibacterial (Handarni *et al.*, 2020; Kunti & Zulfa, 2020). Flavonoids have a mechanism for destroying *Staphylococcus aureus* cell membranes by inhibiting the processes of osmoregulation, respiration, and transport of peptidoglycan biosynthesis either directly or indirectly. In addition, flavonoids are able to bind to lipid layers by inactivating or inhibiting the synthesis of intracellular and extracellular enzymes (Hartmann *et al.*, 2010; Gorniak *et al.*, 2019; Reygaert, 2014). Tannins can interfere with bacterial growth by inhibiting iron uptake from the bacterial environment. In addition, tannins can increase the release of K⁺ which can affect permeability and cause damage to bacterial cell membranes (Farha *et al.*, 2020). This is supported by previous research that explains that tannins are able to inhibit the growth of *Staphylococcus aureus* and *Streptococcus sanguis* bacteria by binding to proteins that form the bacterial cell wall (Hepziba *et al.*, 2023).

Saponins have a mechanism by lowering the surface tension of the wall and damaging the permeability of bacterial cell membranes. Cytoplasmic leakage is caused by saponins diffusing through the outer membrane and vulnerable cell walls (Akbar *et al.*, 2016). Vulnerable bacterial cell walls can result in intracellular compound leakage and disrupted bacterial metabolism (Norhayati *et al.*, 2019). Antimicrobial compounds that interfere with bacterial cytoplasm are bactericidal (Cavalieri *et al.*, 2005).

In this study, it is not yet known which compounds work to inhibit bacterial growth, so it is necessary to purify the compounds from the extract which are then tested for further research to obtain the minimum inhibitory concentration and maximum bactericidal concentration.

5. Conclusion

Mango peel extract varietas gedong gincu has the potential to inhibit the growth of *Staphylococcus aureus* bacteria with an effect that depends on the concentration of the extract. The higher the concentration of the extract, the larger the inhibition zone produced. At a concentration of 25%, the inhibition zone recorded was 11.55 mm, while at a concentration of 100% the inhibition zone increased to 16.22 mm.

Conflict of Interest

The research is no conflict from the researchers and the organization and also the funders for the research.

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