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Antibacterial Activity of 70% Ethanol Extract of Jackfruit Leaves (*Artocarpus heterophyllus* Lam.) against *Bacillus subtilis* and *Pseudomonas aeruginosa*

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Abstract

This study evaluated the antibacterial activity of 70% ethanol extract of jackfruit leaves (*Artocarpus heterophyllus* Lam.) against *Bacillus subtilis* and *Pseudomonas aeruginosa*. Jackfruit leaves collected from Jambi, Indonesia, were processed into simplicia and extracted by maceration using 70% ethanol, producing an extract yield of 7.2%. Phytochemical screening revealed the presence of alkaloids, flavonoids, tannins, saponins, steroids, and triterpenoids. Fourier-transform infrared spectroscopy (FTIR) analysis indicated the presence of hydroxyl, aliphatic C–H, carbonyl/aromatic C=C, C–O, and aromatic C–H functional groups, supporting the presence of bioactive secondary metabolites. Antibacterial activity was evaluated using the disc diffusion method at extract concentrations of 20%, 40%, 60%, 80%, and 100%, with chloramphenicol as the positive control and 10% dimethyl sulfoxide as the negative control. The extract produced inhibition zones against both test bacteria, with the highest inhibition observed at 100% concentration. The minimum bactericidal concentration (MBC) assay showed no bacterial growth at 100% extract concentration in all replications for both bacteria, indicating bactericidal activity at the highest concentration tested. However, the minimum inhibitory concentration (MIC) could not be conclusively determined from the current optical density data because the inhibition pattern was not consistently concentration-dependent. These findings suggest that jackfruit leaf extract has potential as a natural antibacterial agent, although further studies are required to confirm MIC values, isolate active compounds, standardize extract quality, evaluate safety, and develop suitable formulations.

Keywords: antibacterial activity; *Artocarpus heterophyllus*; *Bacillus subtilis*; disc diffusion; jackfruit leaves; *Pseudomonas aeruginosa*.

Abbreviations: DMSO: dimethyl sulfoxide; FTIR: Fourier-transform infrared spectroscopy; MBC: minimum bactericidal concentration; MIC: minimum inhibitory concentration; NA: nutrient agar; OD: optical density.

INTRODUCTION

Infectious diseases remain a major health problem in developing countries, including Indonesia. Bacterial infections are of particular concern because bacterial cells can multiply rapidly and may require effective antimicrobial control. Antibacterial agents are compounds that suppress bacterial growth or kill bacterial cells by interfering with essential physiological processes, including cell-wall formation, membrane integrity, enzymatic activity, and nucleic acid or protein synthesis (Tilarso et al., 2021; Nurhamidin et al., 2022). However, the increasing occurrence of bacterial resistance to synthetic antibiotics has encouraged the exploration of plant-derived antibacterial agents that are safer, more accessible, and potentially more sustainable.

Bacillus subtilis and *Pseudomonas aeruginosa* are relevant test organisms for evaluating antibacterial activity because they represent different bacterial cell-wall structures. *B. subtilis* is a Gram-positive, rod-shaped bacterium that can occur in soil, water, and the human body, whereas *P. aeruginosa* is a Gram-negative opportunistic bacterium frequently associated with wound, respiratory, urinary tract, and postoperative infections (Suriani & Muis, 2016; Suyono & Farid, 2011). The structural difference between Gram-positive and Gram-negative bacteria is important because the outer membrane of Gram-negative bacteria can limit the penetration of antibacterial compounds.

Jackfruit (*Artocarpus heterophyllus* Lam.), a member of the Moraceae family, is widely available in Indonesia and has been reported to contain bioactive secondary metabolites. Jackfruit leaves have been associated with antimicrobial properties and are known to contain flavonoids, saponins, tannins, alkaloids, steroids, and triterpenoids (HK, 2016; Ranasinghe et al., 2019; Aulia

Debby Pelu et al., 2022). These compounds may contribute to antibacterial activity through protein denaturation, enzyme inhibition, membrane disruption, and changes in cell permeability. Previous work also reported that 70% ethanol extract of jackfruit leaves contains antibacterial secondary metabolites and can inhibit bacterial growth (Mawardika et al., 2023).

On this basis, the present study aimed to evaluate the antibacterial activity of 70% ethanol extract of jackfruit leaves against *B. subtilis* and *P. aeruginosa*, to identify major phytochemical groups and functional groups in the extract, and to determine the extract concentration with inhibitory and bactericidal effects. The results are expected to provide scientific evidence for the further development of jackfruit leaves as a source of natural antibacterial agents.

MATERIALS AND METHODS

Study area and sample collection

This experimental study was conducted at Biomedical Laboratory II, Universitas Adiwangsa Jambi, Indonesia. Jackfruit leaves were collected from Kenali Besar Subdistrict, Alam Barajo District, Jambi City. The plant material was taxonomically identified at the Plant Taxonomy Laboratory, Department of Biology, Faculty of Mathematics and Natural Sciences, Universitas Padjadjaran. The sample was confirmed as jackfruit (*Artocarpus heterophyllus* Lam.).

Materials

The materials used in this study included jackfruit leaves, 70% ethanol, distilled water, nutrient agar, bacterial suspensions of *B. subtilis* and *P. aeruginosa*, 10% DMSO, chloramphenicol 30 µg, hydrochloric acid, Mayer reagent, amyl alcohol, magnesium powder, ferric chloride, acetic acid, sulfuric acid, barium chloride, and McFarland standard solution. The equipment included a grinder, sieves, filter paper, rotary evaporator, analytical balance, glassware, autoclave, water bath, sterile Petri dishes, inoculating loop, incubator, caliper, oven, and aluminum foil.

Preparation of simplicia and extract

Fresh mature jackfruit leaves were washed, dried under sunlight for three days, ground into powder, and sieved through a 40-mesh sieve to obtain uniform particle size. A total of 365.3 g of simplicia powder was macerated using 70% ethanol for 3 × 24 hours. The filtrate was separated using filter paper and concentrated with a rotary vacuum evaporator at 45 °C until a viscous extract was obtained. The extract was stored in a brown glass bottle until analysis (Dwiyanti & Suarya, 2023). Extract yield was calculated as the ratio of extract weight to the initial simplicia weight multiplied by 100%.

Phytochemical screening

Phytochemical screening was performed to detect major secondary metabolite groups. Flavonoids were tested by adding water to the extract, heating for 10 minutes, and adding magnesium powder, hydrochloric acid, and amyl alcohol; a red to yellow coloration indicated a positive reaction. Saponins were detected by shaking the sample with hot water and adding hydrochloric acid; stable foam indicated saponins. Alkaloids were detected using Mayer reagent after dissolution in 70% ethanol. Tannins were detected using ferric chloride, whereas steroids and triterpenoids were tested using acetic acid and concentrated sulfuric acid (Aulia Debby Pelu et al., 2022; Kusumawati et al., 2017; Wahyuni et al., 2024).

FTIR analysis

Fourier-transform infrared spectroscopy (FTIR) was used to identify functional groups in the jackfruit leaf extract. Approximately 0.1 g of concentrated extract was analyzed using an FTIR spectrophotometer in the wavenumber range of 4000–400 cm^{-1}. The detected absorption bands were interpreted based on their characteristic functional-group regions.

Preparation of bacterial cultures

Pure cultures of *B. subtilis* and *P. aeruginosa* were rejuvenated by streaking one loopful of bacterial culture onto nutrient agar and incubating at 37 °C for 24 h (Angelina, 2015). Bacterial suspensions were prepared by transferring colonies into sterile solution and adjusting the turbidity to the 0.5 McFarland standard (Amanah et al., 2018; Alouw et al., 2022).

Disc diffusion antibacterial assay

Antibacterial activity was evaluated using the paper disc diffusion method because it is rapid, simple, and suitable for preliminary screening (Katrin et al., 2015). Nutrient agar plates were inoculated with standardized bacterial suspension. Sterile paper discs were treated with jackfruit leaf extract at concentrations of 20%, 40%, 60%, 80%, and 100%. Chloramphenicol was used as the positive control, whereas 10% DMSO was used as the negative control. Plates were incubated at 37 °C for 24 h. The inhibition zone around each disc was measured using a caliper in millimeters by calculating the vertical and horizontal diameters after subtracting the disc diameter (Toy et al., 2015).

Determination of MIC and MBC

The minimum inhibitory concentration (MIC) was determined by liquid dilution using extract concentrations of 20%, 40%, 60%, 80%, and 100% prepared in 10% DMSO. Each concentration was inoculated with standardized bacterial suspension and incubated at 37 °C for 24 hours. Optical density was measured before and after incubation, and the change in OD was used to indicate bacterial growth inhibition

(Balouiri et al., 2016; CLSI, 2020). For minimum bactericidal concentration (MBC) determination, one loopful from the MIC assay was streaked onto sterile nutrient agar and incubated at 37 °C for 24 h. The lowest extract concentration showing no bacterial colony growth was recorded as the MBC (Balouiri et al., 2016; CLSI, 2020).

Data analysis

Organoleptic and phytochemical results were analyzed descriptively. Inhibition-zone data and OD values were summarized as mean ± standard deviation. The antibacterial data were analyzed using one-way analysis of variance in SPSS, with $p < 0.05$ considered to indicate a statistically significant difference among treatments.

RESULTS AND DISCUSSION

Extract yield and phytochemical profile

The preparation of jackfruit leaf simplicia produced 365.3 g of dried powder from the collected plant material. Maceration with 70% ethanol yielded 26.2 g of concentrated extract, corresponding to an extract yield of 7.2% (Table 1). The use of 70% ethanol was appropriate because this solvent can extract both polar and semi-polar compounds and is commonly applied in natural-product extraction. Smaller and more uniform particle size may also improve solvent contact with plant tissues and enhance extraction efficiency (Maulidah et al., 2022).

Table 1. Yield of 70% ethanol extract of jackfruit leaves.

Material	Simplicia weight (g)	Extract weight (g)	Yield (%)
Jackfruit leaf extract	365.3	26.2	7.2

Phytochemical screening showed that the extract contained alkaloids, flavonoids, tannins, saponins, steroids, and triterpenoids (Table 2). These results support previous reports that jackfruit leaves contain antibacterial secondary metabolites (Aulia Debby Pelu et al., 2022; Mawardika et al., 2023). Phenolic compounds such as flavonoids and tannins may interact with bacterial proteins and cell-wall components, whereas saponins and terpenoid/steroid compounds may disturb membrane integrity. The simultaneous presence of these metabolites suggests that the antibacterial activity of the extract may result from multiple complementary mechanisms.

Table 2. Phytochemical screening of jackfruit leaf extract.

Secondary metabolite	Reagent	Observation	Interpretation
Alkaloids	HCl 2N + Mayer reagent	White precipitate	Positive
Flavonoids	HCl 2N + amyl alcohol + Mg powder	Yellow-green solution	Positive
Tannins	FeCl3 1%	Dark green solution	Positive
Steroids	Acetic acid + H2SO4	Blue-green solution	Positive
Triterpenoids	Acetic acid + H2SO4	Red solution	Positive
Saponins	HCl 2N	Foam height 1 cm	Positive

FTIR functional-group analysis

FTIR analysis detected several functional-group regions in the extract (Table 3). A broad absorption band at 3400–3300 cm^{-1} indicated O–H stretching, which is commonly associated with hydroxyl groups in phenolic compounds. Absorption at 2920–2850 cm^{-1} indicated aliphatic C–H stretching, suggesting hydrocarbon chains that may be associated with terpenoid or steroid compounds. Bands in the 1700–1600 cm^{-1} and 1510–1450 cm^{-1} regions indicated carbonyl and aromatic C=C structures, whereas absorption at 1250–1050 cm^{-1} suggested C–O stretching from alcohols, phenols, esters, or glycosidic compounds. These spectra are consistent with the phytochemical findings and support the presence of compounds that may contribute to antibacterial activity.

Table 3. FTIR functional groups detected in jackfruit leaf extract.

Wavenumber (cm^{-1})	Functional group	Intensity	Interpretation
3400–3300	O–H stretching	Strong, broad	Phenolic hydroxyl groups
2920–2850	Aliphatic C–H	Moderate	Saturated organic chains
1700–1600	C=O / aromatic C=C	Sharp	Carbonyl and conjugated aromatic structures
1510–1450	Aromatic C=C	Moderate	Benzene ring structure of phenolic compounds
1250–1050	C–O stretching	Strong	Alcohols, esters, and phenols
900–700	Aromatic C–H	Sharp	Substituted aromatic compounds
600–700	Fingerprint region	Weak	Complex organic compound pattern

Antibacterial activity by disc diffusion

The disc diffusion assay showed that the 70% ethanol extract of jackfruit leaves produced inhibition zones against both *Bacillus subtilis* and *Pseudomonas aeruginosa* at all tested concentrations. The negative control did not produce any inhibition zone, indicating that 10% DMSO did not contribute to antibacterial activity under the experimental conditions. Against *B. subtilis*, the inhibition zone increased from 4.33 ± 1.52 mm at 20% extract concentration to 7.91 ± 0.89 mm at 100% concentration, indicating a concentration-dependent increase in antibacterial activity. However, the inhibition zones produced by the extract were lower than that of the positive control, which showed an inhibition zone of 22.13 ± 0.90 mm. Against *P. aeruginosa*, the extract also showed increasing inhibition zones from 2.142 ± 0.894 mm at 20% to 7.907 ± 1.949 mm at 100%. These findings suggest that jackfruit leaf extract has antibacterial activity against both Gram-positive and Gram-negative bacteria, although its activity was weaker than the standard antibiotic control. The antibacterial effect may be associated with the presence of flavonoids, tannins, saponins, alkaloids, steroids, and triterpenoids, which may interfere with bacterial cell-wall integrity, membrane permeability, and metabolic processes.

Table 4. Inhibition zones of jackfruit leaf extract against Bacillus subtilis.

Treatment	Replication 1 (mm)	Replication 2 (mm)	Replication 3 (mm)	Mean ± SD (mm)
Negative control	0.00	0.00	0.00	0.00 ± 0.00
Positive control	21.09	22.65	22.66	22.13 ± 0.90
20%	2.66	4.70	5.62	4.33 ± 1.52
40%	5.66	6.11	6.69	6.15 ± 0.52
60%	6.78	7.01	6.33	6.71 ± 0.35
80%	6.62	7.82	6.68	7.04 ± 0.68
100%	7.17	8.89	7.66	7.91 ± 0.89

Table 5. Inhibition zones of jackfruit leaf extract against Pseudomonas aeruginosa.

Treatment	Replication 1 (mm)	Replication 2 (mm)	Replication 3 (mm)	Mean ± SD (mm)
Positive control	5.663	1.109	1.687	2.820 ± 2.479
Negative control	0.000	0.000	0.000	0.000 ± 0.000
20%	1.109	2.654	2.662	2.142 ± 0.894
40%	2.662	4.698	5.623	4.328 ± 1.515
60%	2.623	7.824	3.681	4.709 ± 2.749
80%	6.779	3.008	6.333	5.373 ± 2.061
100%	9.167	8.892	5.663	7.907 ± 1.949

MIC and MBC evaluation

The liquid dilution assay was used to evaluate changes in optical density before and after incubation as an indicator of bacterial growth. The OD values showed variable responses among extract concentrations (Tables 6 and 7). In *Bacillus subtilis*, the ΔOD increased from 0.2820 ± 0.2490 at 20% to 0.7907 ± 0.1956 at 100%, indicating that the extract did not show a consistent concentration-dependent inhibitory pattern. In *Pseudomonas aeruginosa*, lower ΔOD values were observed at 20% and 40%, but higher concentrations showed increased OD changes and large standard deviations. Therefore, the MIC could not be conclusively determined from the present OD data. Clear MIC interpretation requires well-defined controls, consistent baseline correction, and confirmation that the selected lowest concentration truly prevents bacterial growth compared with the growth or solvent control.

Table 6. Optical density values in the MIC assay against Bacillus subtilis.

Concentration	Before incubation ± SD	After incubation ± SD	OD ± SD
100%	0.0500 ± 0.0100	0.8407 ± 0.1956	0.7907 ± 0.1956
80%	0.0500 ± 0.0100	0.5873 ± 0.2053	0.5373 ± 0.2053
60%	0.0500 ± 0.0100	0.5209 ± 0.2700	0.4709 ± 0.2700
40%	0.0500 ± 0.0100	0.4828 ± 0.1516	0.4328 ± 0.1516
20%	0.0500 ± 0.0100	0.3320 ± 0.2490	0.2820 ± 0.2490
Positive control	0.0500 ± 0.0100	0.2642 ± 0.0893	0.2142 ± 0.0893
Negative control	0 ± 0	0 ± 0	0 ± 0
Media control	0 ± 0	0 ± 0	0 ± 0
Extract control	0.0500 ± 0.0100	0.0500 ± 0.0100	0.0000 ± 0.0000

Table 7. Optical density values in the MIC assay against Pseudomonas aeruginosa.

Concentration	Before incubation ± SD	After incubation ± SD	OD ± SD
100%	0.0500 ± 0.0100	0.6979 ± 0.8272	0.6479 ± 0.8273
80%	0.0500 ± 0.0100	0.5663 ± 0.6941	0.5163 ± 0.6942
60%	0.0500 ± 0.0100	0.5623 ± 0.5565	0.5123 ± 0.5566
40%	0.0500 ± 0.0100	0.2674 ± 0.4231	0.2174 ± 0.4232
20%	0.0500 ± 0.0100	0.2662 ± 0.3153	0.2162 ± 0.3155
Positive control	0.0500 ± 0.0100	0.1109 ± 0.4635	0.0609 ± 0.4636
Negative control	0 ± 0	0 ± 0	0 ± 0
Media control	0 ± 0	0 ± 0	0 ± 0
Extract control	0.0500 ± 0.0100	0.0500 ± 0.0100	0.0000 ± 0.0000

The MBC assay provided a clearer bactericidal pattern compared with the MIC assay. At 100% extract concentration, no bacterial growth was observed in all three replications for both *B. subtilis* and *P. aeruginosa* (Tables 8 and 9). In contrast, bacterial growth was still observed at concentrations of 80%, 60%, 40%, and 20%. Therefore, the MBC of the jackfruit leaf extract was identified as 100% for both tested bacteria. This result indicates that the extract exhibited bactericidal activity only at the highest concentration tested. The absence of bacterial growth at 100% concentration suggests that the active compounds in jackfruit leaf extract were sufficient to kill bacterial cells rather than merely inhibit their growth. However, the presence of growth at lower concentrations indicates that the extract did not exert a bactericidal effect at 20%–80%. The bactericidal activity may be related to the presence of flavonoids, tannins, saponins, alkaloids, steroids, and triterpenoids, which may disrupt bacterial cell-wall integrity, increase membrane permeability, denature proteins, and interfere with bacterial metabolism.

Table 8. MBC assay of jackfruit leaf extract against Bacillus subtilis.

Extract concentration	Replication 1	Replication 2	Replication 3
100%	No growth	No growth	No growth
80%	Growth	Growth	Growth
60%	Growth	Growth	Growth
40%	Growth	Growth	Growth
20%	Growth	Growth	Growth

Table 9. MBC assay of jackfruit leaf extract against Pseudomonas aeruginosa.

Extract concentration	Replication 1	Replication 2	Replication 3
100%	No growth	No growth	No growth
80%	Growth	Growth	Growth
60%	Growth	Growth	Growth
40%	Growth	Growth	Growth
20%	Growth	Growth	Growth

Proposed antibacterial mechanism

The antibacterial activity of jackfruit leaf extract may be attributed to the combined effects of several secondary metabolites present in the extract. This explanation is supported by previous studies reporting that jackfruit leaves contain bioactive compounds with antibacterial

potential. Mawardika et al. (2023) reported that 70% ethanol extract of jackfruit leaves contained flavonoids, tannins, saponins, and triterpenoids and showed inhibitory activity against *Salmonella typhi*. Similarly, Sousa et al. (2021) reported that jackfruit leaf extracts exhibited bactericidal activity against foodborne pathogens such as *Escherichia coli* and *Salmonella enterica*.

Flavonoids and tannins may contribute to antibacterial activity by interacting with bacterial proteins, enzymes, and cell-wall components. Cushnie and Lamb (2005) explained that flavonoids may act through several mechanisms, including inhibition of nucleic acid synthesis, disruption of cytoplasmic membrane function, and inhibition of energy metabolism. Tannins have also been reported to inhibit bacterial growth through mechanisms such as cell-wall synthesis inhibition, membrane disruption, iron chelation, and suppression of bacterial virulence factors (Farha et al., 2020).

Saponins may increase membrane permeability and promote leakage of cellular components because of their surface-active properties, whereas terpenoids and steroids may interact with lipid components of bacterial membranes and destabilize membrane structure. These mechanisms may explain the bactericidal activity observed at the 100% extract concentration in this study. The inhibition of both *Bacillus subtilis* and *Pseudomonas aeruginosa* suggests that jackfruit leaf extract has antibacterial potential against both Gram-positive and Gram-negative bacteria. However, the outer membrane of Gram-negative bacteria, such as *P. aeruginosa*, may reduce the penetration of active compounds and contribute to differences in bacterial sensitivity.

CONCLUSIONS

The 70% ethanol extract of jackfruit leaves (*Artocarpus heterophyllus* Lam.) demonstrated antibacterial activity against *Bacillus subtilis* and *Pseudomonas aeruginosa*. The extract yield was 7.2%, and phytochemical screening confirmed the presence of alkaloids, flavonoids, tannins, saponins, steroids, and triterpenoids. FTIR analysis supported the presence of hydroxyl, aliphatic C–H, aromatic, carbonyl, and C–O functional groups, indicating the presence of bioactive secondary metabolites. The disc diffusion assay showed inhibition zones against both bacteria, while the MBC assay demonstrated that the 100% extract concentration produced no bacterial growth in all replications, indicating bactericidal activity at this concentration. However, the MIC could not be conclusively determined from the current OD data due to the absence of a consistent concentration-dependent inhibition pattern. Further studies are needed to verify the MIC value, isolate active compounds, standardize extract quality, evaluate safety, and assess formulation potential before product development.

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REFERENCES

Alouw, G., Fatimawali, F., & Lebang, J. S. (2022). Uji aktivitas antibakteri ekstrak etanol daun kersen (*Muntingia calabura* L.) terhadap bakteri *Staphylococcus aureus* dan *Pseudomonas aeruginosa* dengan metode difusi sumuran. *Jurnal Farmasi Medica/Pharmacy Medical Journal (PMJ)*, 5(1), 36–44.

Angelina, S. (2015). Pengaruh pemberian berbagai konsentrasi gula pasir terhadap produksi biomassa dan asam 5-aminolevulinat (ALA) oleh isolat bakteri BD-16. Universitas Negeri Jakarta.

Arifin, A. R., Pratama, R., & Hadi, S. (2025). Optimasi ekstraksi fenolik dan aktivitas antioksidan ekstrak etanol pada daun nangka. *Jurnal Fitofarmaka Indonesia*, 12(1), 45–54.

Aulia Debby Pelu, Hamka Sangkala, & Akbar Mahfudz Ismail. (2022). Uji aktivitas antibakteri ekstrak etanol daun nangka (*Artocarpus heterophyllus* Lam.) terhadap pertumbuhan bakteri *Staphylococcus aureus*. *Jurnal Sains dan Kesehatan*, 6(1), 46–54. https://doi.org/10.57214/jusika.v6i1.221

Balouiri, M., Sadiki, M., & Ibnsouda, S. K. (2016). Methods for in vitro evaluating antimicrobial activity: A review. *Journal of Pharmaceutical Analysis*, 6(2), 71–79.

Bedair, H., Hamed, M., & Mansour, F. (2024). New emerging materials with potential antibacterial activities. *Applied Microbiology and Biotechnology*, 108. https://doi.org/10.1007/s00253-024-13337-6

CLSI. (2020). *Performance standards for antimicrobial susceptibility testing*. Clinical and Laboratory Standards Institute.

Cushnie, T. P. T., & Lamb, A. J. (2005). Antimicrobial activity of flavonoids. *International Journal of Antimicrobial Agents,*

26(5), 343–356. https://doi.org/10.1016/j.ijantimicag.2005.09.002

Dwiyanti, I., & Suarya, P. (2023). Efektivitas ekstrak metanol daun nangka (*Artocarpus heterophyllus* Lam.) sebagai inhibitor alami terhadap korosi pada baja ST 37 dalam media korosif HCl. *Jurnal Kimia (Journal of Chemistry)*, 17(1), 192–203.

Farha, A. K., Yang, Q.-Q., Kim, G., Li, H.-B., Zhu, F., Liu, H.-Y., Gan, R.-Y., & Corke, H. (2020). Tannins as an alternative to antibiotics. *Food Bioscience, 38*, 100751. https://doi.org/10.1016/j.fbio.2020.100751

HK, S. H. (2016). Pemanfaatan larutan daun nangka (*Artocarpus heterophyllus*) dengan dosis berbeda terhadap infeksi bakteri pada larva ikan lele dumbo (*Clarias gariepinus*). Universitas Muhammadiyah Makassar.

Hochvaldová, L., Večeřová, R., Kolář, M., Prucek, R., Kvitek, L., Lapcik, L., & Panacek, A. (2022). Antibacterial nanomaterials: Upcoming hope to overcome antibiotic resistance crisis. *Nanotechnology Reviews*, 11, 1115–1142. https://doi.org/10.1515/ntrev-2022-0059

Katrin, D., Idiawati, N., & Sitorus, B. (2015). Uji aktivitas antibakteri dari ekstrak daun malek (*Litsea graciae* Vidal) terhadap bakteri *Staphylococcus aureus* dan *Escherichia coli*. *Jurnal Kimia Khatulistiwa*, 4(1), 7–12.

Kusumawati, E., Apriliana, A., & Yulia, R. (2017). Kemampuan antibakteri ekstrak etanol daun nangka (*Artocarpus heterophyllus* Lam.) terhadap *Escherichia coli*. *Jurnal Sains dan Kesehatan*, 1(7), 327–332.

Maulidah, N., Dewi, R. S., & Kurniawati, Y. (2022). Pengaruh ukuran partikel bahan baku terhadap rendemen dan kadar fenolik pada ekstraksi tumbuhan. *Jurnal Teknologi Pengolahan Hayati*, 8(3), 187–198.

Mawardika, H., Wahyuni, D., & Khasanah, S. M. (2023). Potensi antibakteri ekstrak daun nangka (*Artocarpus heterophyllus* L.) terhadap bakteri *Salmonella typhi*. *Jurnal Farmasi Indonesia*, 20(2), 195–204.

Nurhamidin, S. J., Wewengkang, D. S., & Suoth, E. J. (2022). Uji aktivitas ekstrak dan fraksi organisme laut spons *Aaptos aaptos* terhadap bakteri *Escherichia coli* dan *Staphylococcus aureus*. *Pharmacon*, 11(1), 1285–1291.

Ranasinghe, R. A. S. N., Maduwanthi, S. D. T., & Marapana, R. A. U. J. (2019). Nutritional and health benefits of jackfruit (*Artocarpus heterophyllus* Lam.): A review. *International Journal of Food Science*, 2019, 4327183. https://doi.org/10.1155/2019/4327183

Sousa, D. F. de, Campos Filho, P. C., & Conceição, A. O. da. (2021). Antibacterial activity of jackfruit leaves extracts and the interference on antimicrobial susceptibility of enteropathogen. *Food Science and Technology*. https://doi.org/10.1590/fst.49220

Suriani, & Muis, A. (2016). Prospek *Bacillus subtilis* sebagai agen pengendali hayati patogen tular tanah pada tanaman jagung. *Jurnal Penelitian dan Pengembangan Pertanian*, 35(1), 37–45. https://doi.org/10.21082/jp3.v35n1.2016.p37-45

Suyono, Y., & Farid, S. (2011). Identifikasi dan karakterisasi bakteri *Pseudomonas* pada tanah yang terindikasi terkontaminasi logam. *Jurnal Biopropal Industri*, 2(1), 8–13.

Tilarso, D. P., Muadifah, A., Handaru, W., Pratiwi, P. I., & Khusna, M. L. (2021). Aktivitas antibakteri kombinasi ekstrak daun sirih dan belimbing wuluh dengan metode hidroekstraksi. *Chempublish Journal*, 6(2), 63–74. https://doi.org/10.22437/chp.v6i2.21736

Toy, T. S., Lampus, B. S., & Hutagalung, B. S. (2015). Uji daya hambat ekstrak rumput laut *Gracilaria* sp. terhadap pertumbuhan bakteri *Staphylococcus aureus*. *e-GiGi*, 3(1).

Wahyuni, D., et al. (2024). [Please complete this reference based on the original source used for steroid/triterpenoid testing].

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