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Antibacterial Activity of Matoa (*Pometia pinnata*) Leaf Extracts and Fractions Against *Staphylococcus aureus* ATCC 6538P

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ABSTRACT

Matoa (*Pometia pinnata*) leaves contain alkaloids, flavonoids, tannins, saponins, phenols, steroids, and terpenoids that can act as antibacterial agents. This experimental study aimed to evaluate the antibacterial activity of 70% ethanolic extract and the water, n-hexane, and ethyl acetate fractions of matoa leaves against *Staphylococcus aureus* ATCC 6538P. Antibacterial testing was performed using the well diffusion method at concentrations of 10%, 20%, and 30% (w/v), prepared in 10% dimethyl sulfoxide (DMSO). Ciprofloxacin 1 µg/well and 10% DMSO were used as positive and negative controls, respectively. Inhibition zones were measured after 24 h incubation at 37 °C in triplicate. The 30% ethyl acetate fraction produced the largest inhibition zone (13.2 mm), followed by the 30% ethanolic extract (11.6 mm), whereas the n-hexane and water fractions yielded smaller zones (9.3 mm and 5.9 mm, respectively, at 30%). Ciprofloxacin produced a very strong inhibition zone of 33.0 mm, while 10% DMSO showed no inhibition. One-way ANOVA with Bonferroni post-hoc analysis indicated that increasing concentration significantly enhanced the inhibition zone for several test materials ($p < 0.05$). Overall, the 30% ethyl acetate fraction exhibited the strongest in-vitro antibacterial activity among all matoa leaf preparations tested, although its effect remained weaker than that of ciprofloxacin.

Keywords:
Staphylococcus aureus; *Pometia pinnata*; Matoa Leaves; Well diffusion; Antibacterial activity

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1. Introduction

The Indonesian population has long utilised various plant parts such as leaves, seeds, roots, tubers, and wood for maintaining health and treating disease. One of the plants used traditionally as herbal medicine is matoa (*Pometia pinnata*) [1]. Matoa has been reported to be useful in the management of wounds and fever [2]. The leaves contain alkaloids, flavonoids, saponins, phenols, steroids, terpenoids, and tannins, which have been associated with antibacterial activity [3].

Maceration with hydroalcoholic solvents is commonly used for extracting secondary metabolites from plant leaves because it is simple, does not require heating, and provides good extraction efficiency for polar and semi-polar compounds [5],[6].

Staphylococcus aureus ATCC 6538P is a Gram-positive bacterium frequently implicated in purulent infections and abscesses of hair follicles and sweat glands, as well as boils and infected burn wounds [2]. These infections may be transmitted via direct contact, particularly through contaminated hands, to skin lesions, post-surgical wounds, and other injured areas [4]. Given its clinical importance and widespread occurrence, *S. aureus* ATCC 6538P is commonly used as a reference strain in antibacterial studies.

Ethanolic extracts and fractions of matoa leaves have demonstrated antibacterial potential in previous research. Sidoretno (2022) reported that 96% ethanolic extract of matoa leaves at concentrations of 10%, 20%, and 30% produced inhibition zones of 11.06, 15.07, and 16.07 mm against *S. aureus* ATCC 6538P [4]. Azlin et al. (2023) found that ethyl acetate fractions at 10%, 20%, and 30% generated mean inhibition zones of 8.39, 10.64, and 12.00 mm, respectively [2]. Risna (2023) further showed that 70% ethanolic extract at concentrations of 1%, 1.5%, and 2% produced inhibition zones of 12.88, 13.15, and 11.36 mm using the disc diffusion method.

However, studies combining 70% ethanolic extract with water, n-hexane, and ethyl acetate fractions of matoa leaves and evaluating their antibacterial activity against *S. aureus* ATCC 6538P using the well diffusion method are still limited. The well diffusion method allows liquid preparations to be applied directly into agar wells and provides good sensitivity for screening crude extracts. Therefore, this study aimed to determine the inhibition zones produced by 70% ethanolic extract and different solvent fractions of matoa leaves at concentrations of 10%, 20%, and 30% against *S. aureus* ATCC 6538P using the well diffusion method.

2. Method

This experimental study used green matoa leaves collected from positions 1–4 from the leaf tip. Samples were obtained from North Tanoyan Village, Lolayan District, Bolaang Mongondow Regency, North Sulawesi. Antibacterial activity against *Staphylococcus aureus* ATCC 6538P was tested on the 70% ethanolic extract and water, n-hexane, and ethyl acetate fractions of matoa leaves at concentrations of 10%, 20%, and 30% using the well diffusion method.

Equipment and Materials

The equipment used included an autoclave, beakers, Petri dishes, cork borers, Erlenmeyer flasks, measuring cylinders, incubator, vernier caliper, watch glasses, spirit lamp, oven, water bath, micropipettes, test-tube racks, spreaders, test tubes, and analytical balance. The materials used were acetic acid, sulphuric acid, *S. aureus* ATCC 6538P, ciprofloxacin, dimethyl sulfoxide (DMSO), 70% ethanol, ethyl acetate, sterile cotton, Mueller-Hinton agar (MHA), nutrient agar (NA), 0.9% physiological NaCl, n-hexane, and matoa leaf simplicia.

Extraction

Dried matoa leaves were ground into powder and sieved through a 40-mesh sieve. A total of 500 g of simplicia powder was weighed and placed in a dark bottle, then 5 L of 70% ethanol was added as solvent at a ratio of 1:10 [7]. Maceration was carried out for 3 × 24 h with occasional stirring. The extract was separated by filtration and concentrated using a rotary evaporator at 70 °C until a thick extract was obtained [2].

Ethanol-Free Test

The viscous extract was mixed with 1 mL of acetic acid and 1 mL of sulphuric acid in a test tube and heated. The extract was considered free of ethanol if no ester odour was detected [6].

Fractionation

Ten grams of matoa leaf extract was dissolved in 100 mL of distilled water and placed in a separating funnel, then fractionated with 100 mL of n-hexane. After shaking and allowing the mixture to stand, two layers formed, with the n-hexane layer above and the aqueous layer below; the n-hexane layer was collected. Next, 100 mL of ethyl acetate was added to the remaining aqueous phase, shaken, and left to stand until two layers formed; the upper ethyl acetate layer was then collected. All fractions were concentrated using a water bath at 70 °C until a constant weight was achieved [8]. Each fraction yielded approximately 6 g (60%).

Preparation of Concentration Series

For preparation of the concentration series, each extract and fraction was weighed at 0.1 g (10%), 0.2 g (20%), and 0.3 g (30%), then dissolved in 1 mL of 10% DMSO. Thus, each 1 mL of solution contained 0.1, 0.2, or 0.3 g extract/fraction in 10% DMSO. For antibacterial testing, 5 µL of each solution was applied into 6-mm diameter wells.

Antibacterial Activity Test

Sterilisation of Equipment and Materials

The equipment and media used in this study were sterilised beforehand using an autoclave at 121°C for 30 minutes [9].

Preparation of Muller-Hinton Agar (MHA) Medium

A total of 9.9 g of MHA powder was weighed into a 500-mL Erlenmeyer flask, 260 mL of distilled water was added, and the mixture was stirred until homogeneous. The medium was sterilised in an autoclave at 121 °C for 30 min and then poured into Petri dishes [2] .

Preparation of Positive Control Solution and negative control

Ciprofloxacin 500-mg tablets were crushed, and 5 mg of powder was dissolved in 25 mL of 10% DMSO to obtain a stock solution of 0.2 µg/µL. For each well, 5 µL of this solution (equivalent to 1 µg ciprofloxacin per well) was used as the positive control [10]. The negative control consisted of 5 µL of 10% DMSO in each well.

Preparation of Bacterial Suspension

A single colony of *S. aureus* ATCC 6538P from NA was transferred into a test tube containing 5 mL of physiological saline. The resulting suspension was used as the inoculum within 15 min [11] .

Antibacterial Testing Using the Well Method

The bacterial suspension was standardised to a turbidity equivalent to 0.5 McFarland (1×10^8 CFU/mL). The suspension was spread evenly over the surface of MHA plates using a sterile spreader. Wells were made using a cork borer, and 5 µL of each test solution, positive control, and negative control was added into the respective wells. Plates were incubated at 37 °C for 18–24 h, and clear zones around the wells were measured as inhibition zones [11] .

Data analysis

The inhibition zone diameters (mm) were recorded as mean ± standard deviation (SD) from three replicates. Differences between concentrations for each test material

were analysed using one-way ANOVA followed by Bonferroni post-hoc test in SPSS, with a significance level of $p < 0.05$.

3. Results and Discussion

Extraction yield

This study aimed to determine the inhibition zone of ethanol extract of matoa leaves, water fraction, n-hexane fraction, and ethyl acetate fraction against *Staphylococcus aureus* ATCC 6538P bacteria at concentrations of 10%, 20%, and 30%. Matoa leaves were obtained from Tanoyan Utara Village, Lolayan District, Bolaang Mongondow Regency, North Sulawesi Province.

The method used in the extraction process was maceration because, in addition to being easy and simple, this method is commonly used for leaf samples because the active substances in the samples are not heat-resistant, which can damage the active substances [12]. Maceration was carried out using 70% ethanol solvent because it can better extract polar compounds [13]. The solvent ratio used is 1:10 to ensure the sample is fully submerged, and the more solvent used, the greater the amount of extract obtained [14]. The resulting extract is then concentrated to obtain a thick extract, and the yield is calculated. The Indonesian Ministry of Health (2017) states that a good yield is when the yield value is above 10% [15]. The yield obtained from the concentrated extract of matoa leaves was 10.5%, which can be considered good as it is above 10%. The yield results can be seen in **Table 1**.

Table 1. Yield results of ethanol extract from matoa leaves

Weight of crude drug (g)	Extract weight (g)	Yield (%)
500	52.764	10.5

The concentrated extract obtained was then fractionated using the liquid-liquid extraction method with a separating funnel. The purpose of fractionation is to separate compounds based on their polarity [2]. In this study, aquades, ethyl acetate and n-hexane solvents were used, where solvents with a higher density will be in the lower layer, while solvents with a lower density will be in the upper layer. The purpose of varying the solvents is so that the secondary metabolite compounds bound to the sample can be extracted in solvents based on their polarity. Polar compounds will enter polar solvents (distilled water), semi-polar compounds will enter the semi-polar solvent (ethyl acetate), and non-polar compounds will enter the non-polar solvent (n-hexane) [16].

Antibacterial activity

Ethanol extracts and water, ethyl acetate and n-hexane fractions were tested for antibacterial activity against *Staphylococcus aureus* ATCC 6538P bacteria. In this study, several concentrations were used, namely 10%, 20% and 30%. These concentrations were used to determine the concentration that could produce the greatest inhibitory effect against *Staphylococcus aureus* ATCC 6538P bacteria. DMSO (*dimethyl sulfoxide)* can dissolve thick extracts better than distilled water, therefore it was used to make a series of solutions with concentrations of [2]. The solutions were then used in the antibacterial activity test of the ethanolic extract of matoa leaves and the water, ethyl acetate, and n-hexane fractions against *Staphylococcus aureus* ATCC 6538P bacteria. The results of the antibacterial activity test are shown in **Table 2**.

Table 2. Inhibition zones of ethanolic extract and fractions of matoa leaves against *Staphylococcus aureus* ATCC 6538P

Test material	Concentration (% w/v)	Replicate I (mm)	Replicate II (mm)	Replicate III (mm)	Mean (mm)	SD
Ethanolic extract	10	8.3	8.9	9.6	8.9	1.3503
Ethanolic extract	20	9.9	10.3	10.9	10.3	1.3503
Ethanolic extract	30	11.2	11.7	12.1	11.6	1.3503
Water fraction	10	4.3	4.5	4.9	4.5	0.7095
Water fraction	20	5.1	5.7	5.5	5.4	0.7095
Water fraction	30	5.9	6.1	5.7	5.9	0.7095
n-hexane fraction	10	8.1	8.3	8.5	8.3	0.5508
n-hexane fraction	20	8.7	7.9	8.6	8.4	0.5508
n-hexane fraction	30	9.1	9.3	9.5	9.3	0.5508
Ethyl acetate fraction	10	10.9	10.8	10.7	10.8	1.2220
Ethyl acetate fraction	20	12.6	11.4	11.0	11.6	1.2220
Ethyl acetate fraction	30	13.9	13.2	12.7	13.2	1.2220
Positive control (ciprofloxacin)	–	33.0	33.0	33.0	33.0	0.0
Negative control (10% DMSO)	–	0.0	0.0	0.0	0.0	0.0

Note: Data are presented as inhibition zone diameters (mm), mean ± SD, from three replicates (n = 3) for each material and concentration. Positive control: ciprofloxacin; negative control: 10% DMSO

Based on the results in **Table 2**, a clear zone can be seen forming around the well and measured using a vernier caliper, where the thick extract of matoa leaves has antibacterial activity because it contains alkaloids, flavonoids, tannins and saponins that act as antibacterial agents [4], with an average inhibition zone of 8.9 mm (10% concentration), 10.3 mm (20%) and 11.6 mm (30%). The water fraction also has antibacterial activity because it contains flavonoid compounds [9] with an average inhibition zone diameter of 4.5 mm (10%), 5.4 mm (20% concentration) and 5.9 mm (30% concentration). The n-hexane fraction exhibits inhibition zone activity due to the presence of terpenoid compounds [17]. The inhibition zones produced by the n-hexane fraction are stronger than those of the aqueous fraction, with average inhibition zone diameters of 8.3 mm (10% concentration), 8.4 mm (20% concentration), and 9.3 mm (30% concentration). The ethyl acetate fraction contains flavonoid and alkaloid compounds that act as antibacterial agents with an average inhibition zone diameter of 10.8 mm (10% concentration), 11.6 mm (20% concentration) and 13.2 mm (30% concentration) [2]. For the positive control, the inhibition zone produced was 33 mm, which is classified as very strong, while the negative control did not produce an inhibition zone. Based on the results obtained, it can be said that the higher the concentration, the larger the inhibition zone produced.

Table 3. Bonferroni post-hoc comparisons of inhibition zones between concentrations for each test material

Test material	Concentration comparison (%, w/v)	Mean difference (mm)*	p-value	Significant (p < 0.05)
Ethanolic extract	10 vs 20	−1.43	0.053	No
Ethanolic extract	10 vs 30	−2.73	0.002	Yes
Ethanolic extract	20 vs 30	−1.30	0.078	No
Water fraction	10 vs 20	−0.87	0.025	Yes
Water fraction	10 vs 30	−1.33	0.003	Yes
Water fraction	20 vs 30	−0.47	0.248	No
n-hexane fraction	10 vs 20	−0.10	1.000	No
n-hexane fraction	10 vs 30	−1.00	0.019	Yes
n-hexane fraction	20 vs 30	−0.90	0.031	Yes
Ethyl acetate fraction	10 vs 20	−0.87	0.376	No
Ethyl acetate fraction	10 vs 30	−2.47	0.007	Yes
Ethyl acetate fraction	20 vs 30	−1.60	0.050	No

*Note: *Mean difference is calculated as (lower concentration − higher concentration); negative values indicate larger inhibition zones at the higher concentration.*

Post-hoc Bonferroni analysis (Table 3) showed that, for the ethanolic extract and ethyl acetate fraction, the 30% concentration produced significantly larger inhibition zones than the 10% concentration ($p = 0.002$ and $p = 0.007$, respectively). For the water fraction, both 20% and 30% were significantly higher than 10% ($p < 0.05$), whereas for the n-hexane fraction significant differences were observed between 10% vs 30% and 20% vs 30% ($p < 0.05$)

Table 4. Categories of antibacterial activity [19]

Inhibition zone diameter (mm)	Category
≥ 20	Very strong
10–20	Strong
5–10	Moderate
≤ 5	Weak

Previous studies have categorised antibacterial activity based on inhibition zone diameter as shown in **Table 4** [19]. Based on these criteria, the 10% ethanolic extract of matoa leaves falls into the *moderate* category, whereas the 20% and 30% extracts are classified as having *strong* antibacterial activity. This pattern is consistent with the ability of 70% ethanol to extract polar and semi-polar constituents such as flavonoids, tannins, saponins, alkaloids, and triterpenoids, which are widely associated with antibacterial effects [3],[17],[18]. The water fraction at 10% is categorised as *weak*, while at 20% and 30% it shifts to the *moderate* category. Water predominantly extracts highly polar compounds, including saponins, tannins, flavonoids, and alkaloids, so the antibacterial

effect is present but less pronounced than that of the ethanolic extract. In contrast, the n-hexane fraction at 10%, 20%, and 30% remains in the *moderate* category across all concentrations, which is in line with its ability to extract non-polar constituents such as steroids and other lipophilic compounds that can disrupt bacterial cell membranes [18]. The ethyl acetate fraction at 10%, 20%, and 30% is classified as *strong* because this solvent attracts semi-polar compounds such as flavonoids, alkaloids, tannins, saponins, and phenolics [2],[15]. The positive control, ciprofloxacin, falls into the *very strong* category, confirming the validity of the assay. Overall, these findings indicate that higher concentrations of extract or fraction generally produce larger inhibition zones, consistent with the increased availability of active antibacterial constituents. Moreover, evidence from other natural sources, including secondary metabolites isolated from *Penicillium* sp. with potent antimicrobial properties, supports the role of diverse natural metabolites as important antibacterial agents [21].

This antibacterial study nevertheless has several limitations. The well diffusion method measures activity primarily on the basis of the ability of compounds to diffuse through agar and therefore does not always reflect true intrinsic potency. Minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) values were not determined, so the minimum effective concentrations of each preparation remain unknown. Only three solvents were used for extraction and fractionation, and the number of bacterial strains tested was limited to a single reference strain, which restricts the generalisability of the results. Further studies involving MIC/MBC determination, additional solvent systems, and a broader panel of clinical isolates are required to confirm and extend these findings.

In addition, this study employed crude extracts and solvent fractions without detailed identification of individual active compounds. Unlike studies that have successfully isolated and characterised specific antimicrobial secondary metabolites using chromatographic and spectrometric techniques [21], the present work did not elucidate which constituents of the matoa leaf preparations are primarily responsible for the observed activity, nor did it assess possible cytotoxicity or safety profiles. Future research should therefore incorporate comprehensive phytochemical profiling, bioassay-guided fractionation, and toxicity testing to clarify the mechanism of action and to evaluate the feasibility of developing matoa leaf–derived compounds as safe and effective antibacterial agents.

The well diffusion method was chosen in this study because it offers a simple procedure and relatively high sensitivity for screening liquid extracts and fractions. The osmotic process facilitates diffusion of active compounds from the wells into the surrounding agar, making this method effective for detecting growth inhibition zones [11]. Mueller–Hinton agar (MHA) was used as the culture medium because it is a nutritionally rich, standardised medium that supports robust growth of many bacterial species and is widely recommended for antimicrobial susceptibility testing [2]. Ciprofloxacin was employed as the positive control because it is a broad-spectrum antibiotic that inhibits bacterial DNA synthesis, thereby effectively suppressing the growth of susceptible microorganisms and providing a benchmark for maximal inhibition [22]. Dimethyl sulfoxide (DMSO) was used as the negative control since it served as the solvent for the extracts and fractions and has been shown not to inhibit bacterial growth or exhibit bactericidal properties at the concentrations used in this study.

4. Conclusion

Ethanolic extract and solvent fractions of *Pometia pinnata* leaves inhibited the growth of *Staphylococcus aureus* ATCC 6538P in the well diffusion assay. At 30% (w/v), the ethyl acetate fraction and the ethanolic extract produced mean inhibition zones of 13.2 ± 1.22 mm and 11.6 ± 1.35 mm (n = 3), respectively, whereas the n-hexane and water fractions showed smaller zones of 9.3 ± 0.55 mm and 5.9 ± 0.71 mm, respectively. Ciprofloxacin (1 µg/well) generated an inhibition zone of 33.0 ± 0.0 mm, and 10% DMSO showed no inhibition. Inhibition generally increased with concentration, indicating a concentration-dependent effect within the tested range. Nevertheless, the antibacterial activity of the extract and fractions remained lower than that of ciprofloxacin, and further studies are required to characterise the active constituents, determine MIC and MBC values, and evaluate their potential clinical relevance.

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Conflicts of Interest:

The authors declare no conflict of interest regarding the publication of this paper.

References

[1] F. Marya, "Measurement of specific and non-specific parameters of ethanol extracts of matoa leaves (*Pometia pinnata* J.R. & G. Forst.)," *Jurnal Mandala Pharmacon Indonesia*, vol. 6, no. 1, pp. 1–12, 2020. [Online]. Available: https://doi.org/10.35311/jmpi.v6i01.39

[2] S. Z. Azlin, R, "Antibacterial activity test of ethyl acetate fraction of matoa leaves (*Pometia pinnata* J.R. & G. Forst.) against *Staphylococcus aureus*," *JFARM – Jurnal Farmasi*, vol. 1, no. 1, pp. 30–41, 2023. [Online]. Available: https://doi.org/10.58794/jfarm.v1i1.491

[3] R. Rahmawati, M. Tahir, and A. H. W. Amir, "Chemical compound content and pharmacological activity of matoa plants (*Pometia pinnata* J.R. Forster & J.G. Forster)," *As-Syifaa: Jurnal Ilmiah Farmasi*, vol. 13, no. 2, pp. 108–115, 2021. [Online]. Available: https://doi.org/10.56711/jifa.v13i2.778

[4] W. M. Sidoretno, "Potential of the ethanolic extract of matoa leaves (*Pometia pinnata* J.R. & G. Forst.) against *Staphylococcus aureus*," *JPK: Jurnal Proteksi Kesehatan*, vol. 10, no. 2, pp. 107–112, 2022. [Online]. Available: https://doi.org/10.36929/jpk.v10i2.402

[5] Y. A. Kusmartono and Bambang, "Optimization of solvent volume and maceration time for flavonoid extraction from star-fruit leaves (*Averrhoa bilimbi* L.)," *Jurnal Teknik Kimia*, vol. 10, pp. 58–64, 2016. [Online]. Available: https://media.neliti.com/media/publications/142082-ID-optimasi-volume-pelarut-dan-waktu-masera.pdf

[6] L. Pratiwi, E. Puspitasari, "Ethanol extract, ethyl acetate extract, ethyl acetate fraction, and *n*-hexane fraction of mangosteen," *JPSCR: Journal of Pharmaceutical Science and Clinical Research*, vol. 1, no. 2, p. 71, 2016. [Online]. Available: https://doi.org/10.20961/jpscr.v1i2.1936

[7] M. I. Fatah, D, "Effect of solvent concentration on the antioxidant activity of red

dragon fruit (*Hylocereus polyrhizus*) extract serum preparations," *JIFI: Jurnal Ilmiah Farmasi Imelda*, vol. 7, no. 2, pp. 61–70, 2024. [Online]. Available: https://doi.org/10.52943/jifarmasi.v7i2.1562

[8] M. Andira, A.,T. Siska,W, "Antibacterial activity test of extracts, water fractions, ethyl acetate fractions, and *n*-hexane fractions of Chinese leucaena leaves (*Leucaena leucocephala*) against *Staphylococcus aureus* ATCC 25923," Universitas Duta Bangsa, Indonesia, 2024. [Online]. Available: https://doi.org/10.61132/obat.v2i6.785

[9] W. H. P. Gerung, J. E. K. Runtuwene, and A. M. Lolo, "Antibacterial activity test of bottle-gourd leaf extract (*Averrhoa bilimbi* L.) against the growth of *Propionibacterium acnes*," *Pharmacon: Jurnal Ilmiah Farmasi – Universitas Sam Ratulangi*, vol. 10, no. 4, pp. 1087–1093, 2021. [Online]. Available: https://doi.org/10.35799/pha.10.2021.37403

[10] F. Andriyawan, "Activity testing of ethanol extract of *Melastoma malabathricum* L. leaves against *Escherichia coli in vitro*," *Jurnal Mahasiswa PSPD Fakultas Kedokteran Universitas Tanjungpura*, vol. 1, pp. 11–13, 2015. [Online]. Available: https://jurnal.untan.ac.id/index.php/jfk/article/view/11045

[11] L. S. Nurhayati, R. P. Rahayu, and R. A. Galih, "Comparison of antibacterial activity testing of yogurt starters using the well-diffusion and disc-diffusion methods," *Jurnal Teknologi Hasil Peternakan*, vol. 1, no. 2, p. 41, 2020. [Online]. Available: https://doi.org/10.24198/jthp.v1i2.27537

[12] E. Pujiastuti and D. Andreana, "Determination of total flavonoid content of peel ethyl-acetate extract of *Carica papaya* L.," *Menara: Journal of Health Science*, vol. 1, no. 2, pp. 58–71, 2022. [Online]. Available: http://jurnal.iakmikudus.org/index.php/mjhs

[13] N. H. Novian, "Analysis of ethanol extract of yellow squash fruit (*Cucurbita* sp.)," *Parapemikir: Jurnal Ilmiah Farmasi*, vol. 9, no. 1, pp. 54–59, 2020. [Online]. Available: http://ejournal.poltektegal.ac.id/index.php/parapemikir

[14] R. Y. Asworo and H. Widwiastuti, "Effect of simplicia-powder size and maceration time on the antioxidant activity of soursop peel extract," *Indonesian Journal of Pharmaceutical Education*, vol. 3, no. 2, pp. 256–263, 2023. [Online]. Available: https://doi.org/10.37311/ijpe.v3i2.19906

[15] R. Islamiyati, M. Alhabsyi, and D. Lolo, "Antioxidant activity test of ethyl-acetate extract of matoa leaves using DPPH free radicals," *Pharmacon: Jurnal Ilmiah Farmasi – Universitas Sam Ratulangi*, vol. 13, pp. 611–618, 2024. [Online]. Available: https://doi.org/10.35799/pha.13.2024.55951

[16] I. S. Fatmawati, Haeruddin, and W. O. Mulyana, "Antioxidant activity of ethyl acetate extract of starfruit leaves (Averrhoa bilimbi L.) using the DPPH method," *SAINS: Jurnal Kimia dan Pendidikan Kimia*, vol. 12, no. 1, pp. 41–49, 2023. [Online]. Available: https://doi.org/10.36709/sains.v12i1.31

[17] P. A. R. Utoro, D. P. Y. Sari, and N. A. Azzahra, "A brief literature review of the bioactivity of matoa leaf extract (*Pometia pinnata*) from Indonesia and its application in food products," *Journal of Tropical AgriFood*, vol. 4, no. 2, p. 67, 2022. [Online]. Available: https://doi.org/10.35941/jtaf.4.2.2022.9293.67-76

[18] N. M. G. R. Nomer, A. S. Duniaji, and K. A. Nocianitri, "Flavonoid and anthocyanin content of sappan-wood extract (*Caesalpinia sappan* L.) and its antibacterial activity against *Vibrio cholerae*," *Jurnal Ilmu dan Teknologi Pangan (ITEPA)*, vol. 8, no. 2, p. 216, 2019. [Online]. Available: https://doi.org/10.24843/itepa.2019.v08.i02.p12

[19] A. Saptowo, S. H. Rahmawati, and A. D. Hadi, "Antibacterial activity test of *Sekilang* bark extract (*Embelia borneensis* Scheff.) against *Propionibacterium acnes* and *Staphylococcus epidermidis*," *Al-Ulum: Jurnal Sains dan Teknologi*, vol. 7, no. 2, p. 93, 2022. [Online]. Available: https://doi.org/10.31602/ajst.v7i2.6331

[20] D. U. Aulia, R. Anwar, and R. Juansah, "Antibacterial activity of methanol extract and *n*-butanol fraction of *Euphorbia milii* leaves against *Staphylococcus aureus*," *Jurnal Biologi Tropis*, vol. 23, no. 1, pp. 315–323, 2023. [Online]. Available: https://doi.org/10.29303/jbt.v23i1.6154

[21] Y. Nurulita, Yuharmen, A. Fitri, I. E. Sari, D. N. Sary, and T. T. Nugroho, "Identification of secondary metabolites secreted by local peat-soil fungus Penicillium sp. LBKURCC34 from Riau as antimicrobials," *Chimica et Natura Acta*, vol. 10, no. 3, 2022. [Online]. Available: https://doi.org/10.24198/cna.v10.n3.45994

[22] B. Lombogia, S. L. Kandou, and E. B. Kaunang, "Inhibitory effect of snake-plant leaf extract (*Sansevieria trifasciata* folium) on the growth of *Escherichia coli* and *Streptococcus* spp.," *Jurnal e-Biomedik*, vol. 4, no. 1, 2016. [Online]. Available: https://doi.org/10.35790/ebm.4.1.2016.12230