

ANTIOXIDANT AND ANTIBACTERIAL ACTIVITIES OF ACTIVE COMPOUND FROM *Murraya paniculata* (L.) LEAVES

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ABSTRACT

The present study aimed to isolate and characterize bioactive compounds from *Murraya paniculata* leaves and to evaluate their antioxidant, antibacterial, and cytotoxic activities. The extraction was performed by maceration, followed by phytochemical screening and characterisation by FT-IR and GC-MS analyses. Phytochemical results confirmed the presence of alkaloids, flavonoids, tannins, and saponins, with flavonoids as the dominant compounds. FT-IR analysis indicated the presence of O-H, N-H, C=O, and C-O functional groups, while GC-MS analysis revealed a complex mixture with a dominant compound at a retention time of 13.542 min. The extract exhibited strong antioxidant activity with an IC₅₀ value of 12.64 ppm. Antibacterial activity against *Staphylococcus aureus* showed inhibition zones greater than 20 mm, indicating strong activity. Cytotoxicity was evaluated using the Brine Shrimp Lethality Test (BSLT), yielding an LC₅₀ of 369.83 ppm, indicating moderate toxicity. These results demonstrate that *Murraya paniculata* (L.) leaves are a promising source of natural bioactive compounds with potential pharmaceutical applications.

Keywords: *Murraya Paniculata* (L.), Phytochemical Screening, Antioxidant Activity, Antibacterial Activity, Cytotoxicity Assay.

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INTRODUCTION

Kemuning leaves (*Murraya paniculata* L.), classified as part of the Rutaceae family, are commonly found in the tropical and sub-tropical regions, especially in Indonesia. It is a widely used ornamental plant, but this is also reported to produce several bioactive secondary metabolites with therapeutic potential.¹ Furthermore, previous research has demonstrated that *Murraya paniculata* (L.) leaves possess bioactive compounds such as flavonoids, alkaloids, and phenolics, which are associated with antioxidant, antibacterial, and cytotoxic characteristics.² One class of bioactive compounds in *Murraya paniculata* (L.) that is currently receiving special attention is flavonoids. backbone consisting of two aromatic rings (A and B) connected by a heterocyclic ring (C). Therefore, the presence of hydroxyl groups (-OH), particularly on ring B, as well as the conjugated double-bond system, is the primary determinant of flavonoid antioxidant activity. More specifically, these groups will donate a hydrogen atom or an electron to neutralise free radicals. At the same time, the conjugated system helps stabilise the radicals by distributing electrons throughout the molecular structure.³ The leaf extract of *Murraya paniculata* (L.) has been reported to possess significant antioxidant and antibacterial activity against *Staphylococcus aureus*, as presented by Neta and Tim.⁴ Subsequently, the cytotoxic effects of *Murraya paniculata* (L.) leaf extract have also been explored by Qi and colleagues.⁵ Although the antioxidant, antibacterial, and cytotoxic activities of *Murraya paniculata* (L.) leaves have been widely reported, studies focusing on Fourier transform infrared spectroscopy (FT-IR) and gas chromatography-mass spectrometry (GC-MS) analysis of *Murraya paniculata* (L.) leaf extracts from Indonesia remain relatively rare. Therefore, FT-IR and GC-MS analyses are required to identify the active compounds of *Murraya paniculata* (L.), especially those grown in Indonesia. This study aims to isolate and characterize bioactive compounds from *Murraya paniculata* L leaves through phytochemical screening and characterisation analysis, as well as to evaluate their antioxidant activity using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) method, their antibacterial activity against *Staphylococcus aureus*, and their toxicity to larvae.

EXPERIMENTAL

Material

Kemuning leaves (*Murraya paniculata* (L.) were collected from Bandar Sono Village, North Sumatra, Indonesia. The chemicals used in this study included methanol (analytical grade) as an extraction solvent, DPPH (2,2-diphenyl-1-picrylhydrazyl) for antioxidant assay, and ascorbic acid as a standard. Muller Hinton Agar (MHA) was used as the culture medium for antibacterial testing against *Staphylococcus aureus*. Other reagents used for phytochemical screening included Dragendorff's reagent (for alkaloids), ferric chloride (for phenolics), magnesium powder and hydrochloric acid (for flavonoids), and Liebermann–Burchard reagent (for steroids and terpenoids). All chemicals used were of analytical grade and purchased from Sigma-Aldrich (Germany), without further purification.

Extraction and Purification

The dried *Murraya paniculata* (L.) leaves were powdered and macerated with an appropriate solvent. The resulting extract was filtered to retrieve the bioactive components. Following this, phytochemical screening was implemented as an initial analysis before further analysis.^{6,7}

Identification of Pure Compound

The functional groups of the obtained extract were analysed using phytochemical screening and FT-IR spectroscopy with a Shimadzu IRTracer-100 spectrometer. The FT-IR analysis was performed in the range of 4000–400 cm⁻¹ using the KBr pellet method. Furthermore, gas chromatography–mass spectrometry (GC–MS) analysis was performed to identify bioactive compounds from *Murraya paniculata* (L.) leaves using a Shimadzu GC-MS-QP2010 Plus equipped with an Rxi-5MS column. The identified compounds were then compared with the NIST library.⁶

Antioxidant Activity

To evaluate the antioxidant activity of the obtained extract, the DPPH (2,2-diphenyl-1-picrylhydrazyl) assay was used. Briefly, 5 mL of the extract at 5 µg/mL and 5 mL of the ascorbic acid standard (1 µg/mL) were each mixed with 5 mL of DPPH solution prepared in methanol. Subsequently, the mixtures were kept at 37°C for 30 minutes. Following that, the absorbance was measured at 515 nm using a UV–Vis spectrophotometer (Shimadzu UV-1800). The same procedure was repeated for extract concentrations of 10, 20, 50, and 100 µg/mL, as well as for ascorbic acid concentrations of 2, 5, 10, and 20 µg/mL. The percentage of radical scavenging activity was calculated using the following equation.^{6,8}

$$\% \text{ Inhibition} = \frac{(\text{Absorbance of control solution} - \text{Absorbance of sample})}{\text{Absorbance of control}} \times 100\%$$

Antibacterial Activity

Antibacterial activity was determined using the agar diffusion method. Muller Hinton Agar (MHA) was aseptically poured into Petri dishes and allowed to solidify. A bacterial suspension of *Staphylococcus aureus* was spread evenly on the agar surface using a sterile cotton swab. The extract was applied onto sterile discs (1.5 cm diameter) placed on the inoculated agar. The plates were incubated for 24 h, and the diameter of the inhibition zone was measured.^{6,9}

Cytotoxicity Assay

Cytotoxicity was analysed using the Brine Shrimp Lethality Test (BSLT) with *Artemia salina* larvae. The eggs were hatched in artificial seawater under continuous illumination for 24–48 h. Ten active nauplii were transferred into vials containing varying concentrations of the extract. After 24 h incubation at room temperature, the number of dead larvae was recorded. Mortality was calculated, and the LC₅₀ value was determined by probit analysis.^{10,11}

RESULTS AND DISCUSSION

Phytochemical Screening Analysis

The phytochemical screening results, as indicated in (Table-1), showed that the maceration method successfully extracted *Murraya paniculata* leaves, yielding bioactive secondary metabolites, as evidenced by the presence of alkaloids, flavonoids, tannins, and saponins, confirming the extraction process's

efficiency in isolating polar phytochemicals. At the same time, steroids and terpenoids were not detected. Furthermore, (Table-1) shows that the flavonoid compound was the most dominant compound detected in the extract. Therefore, the obtained extract of *Murraya paniculata* leaves has potential applications as an antioxidant, antibacterial, and cytotoxic agent, since flavonoids are well known for exhibiting these biological activities.¹²

Table-1: Phytochemical Screening of the Leaves of *Murraya paniculata* (L.)

Phytochemical Test	Change	Result
Alkaloids	A brick red precipitate is formed with Dragendroff's reagent	+
	A yellowish-white precipitate is formed with Maeyer's reagent	+
Flavonoids	Black colloid is formed.	++
Tanins	A dark blue or greenish-black solution forms.	+
Saponins	Foam is formed, which lasts $\pm$ 10 minutes.	+
Steroids, Terpenoids	A brownish or violet ring is formed.	-

FT-IR Analysis

The FT-IR spectrum represented in (Fig.-1) indicates that the typical bands commonly found in *Murraya paniculata* (L.) leaf extract were the absorption peak at 3265.1 cm^{-1} , corresponding to O–H and N–H stretching. The peak at 1640 cm^{-1} , which indicated the presence of the C=O bond. Subsequently, the band at 1416 cm^{-1} , which is attributed to O-H bending, and the presence of 1013 cm^{-1} is associated with the C-O band. In addition, the peak at 2124.6 cm^{-1} is attributed to C $\equiv$ N stretching, while the peak at 980 cm^{-1} is attributed to C–H bending. Collectively, these functional groups confirm the presence of phenolic and nitrogen-containing compounds, thereby supporting the phytochemical results and indicating flavonoids as the primary bioactive constituents of the extract.¹³

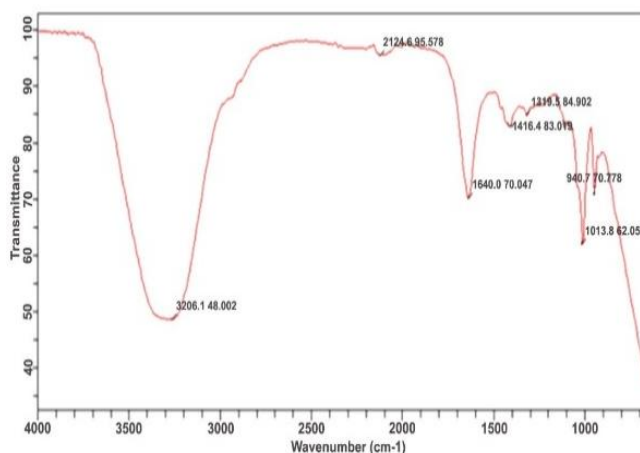
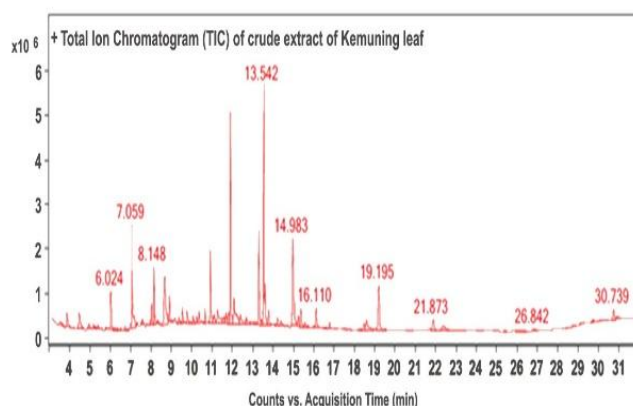


Fig.-1: FT-IR Spectrum of *Murraya paniculata* (L.) Leaf Extract

GC-MS Analysis

The Total Ion Chromatogram (TIC) shows that the crude extract of *Murraya paniculata* contains a complex mixture of compounds with several peaks at various retention times. The main peak at 13.542 minutes corresponds to the dominant component, presumed to be a major bioactive compound, such as a flavonoid. Other peaks in the range of 6.024–30.739 minutes indicate the presence of compounds with different properties, with the early peaks (6–9 minutes) typically representing low-molecular-weight volatile compounds such as simple alkaloids or phenolic derivatives. In contrast, peaks at higher retention times are associated with more complex compounds such as flavonoids, tannins, and saponins. Overall, this GC–MS profile aligns with the phytochemical screening results and suggests that the presence of these bioactive compounds may contribute to the extract's antioxidant, antibacterial, and cytotoxic activities.^{4,14}

Fig.-2: *Murraya paniculata* (L.) Leaf Extract Chromatogram

Antioxidant Activity

Murraya paniculata (L.) extract exhibited significant concentration-dependent antioxidant activity, as demonstrated by the DPPH assay, as shown in (Table-2), and the inhibition percentage is demonstrated in (Fig.-3). Based on (Fig.-3), the percentage inhibition increased from 6.82% at 1 ppm to 77.14% at 20 ppm, indicating a greater free radical scavenging capacity as the extract concentration increased. These results indicate that flavonoid compounds effectively stabilise DPPH free radicals through the donation of hydrogen atoms.¹⁵ Next, an IC_{50} value of 12.64 ppm indicates very strong antioxidant activity. These results are consistent with previous studies reporting that the antioxidant characteristics of a bioactive compound are considered highly effective if its IC_{50} value is lower than 50 ppm.¹⁶ These findings are likely associated with the presence of flavonoid compounds in *Murraya paniculata* (L.). Furthermore, the results highlight the potential of *Murraya paniculata* extract as a valuable natural antioxidant source, with promising applications in pharmaceutical, nutraceutical, and functional food industries.

Table-2: DPPH Antioxidant Test Results

Blank	Concentration (ppm)	Absorbance (A)	% Inhibition (%)	IC_{50}
0.9471	1	0.8825	6.820821455	12.64447164
0.9471	2	0.8380	11.51937493	
0.9471	5	0.7415	21.70837293	
0.9471	10	0.5675	40.08024496	
0.9471	20	0.2165	77.14074543	

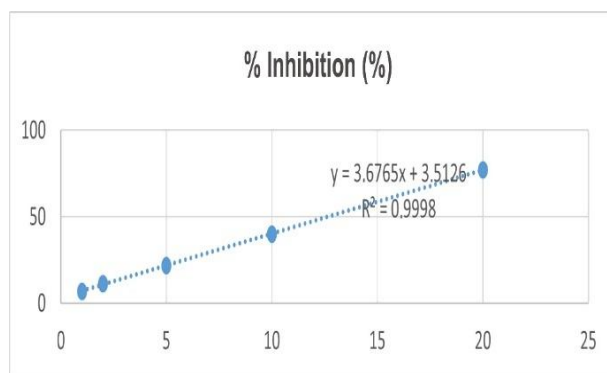


Fig.-3: % Inhibition Graph

Antibacterial Activity

The antibacterial activity of *Murraya paniculata* (L.) extract against *Staphylococcus aureus* is presented in (Table-3). The extract exhibited clear inhibitory effects against the tested bacterium, as evidenced by the formation of inhibition zones at all tested concentrations. The diameter of the inhibition zone increased from 19.5 mm at 0 g to 25.9 mm at 25 g, indicating a concentration-dependent antibacterial effect. These findings agree with previous studies showing that plant extracts with larger inhibition zones

exhibit stronger antibacterial activity.¹⁷ The observed inhibitory effect against *Staphylococcus aureus* indicates that the extract demonstrates considerable antibacterial potential.

Table-3: Antibacterial Activity of the Obtained Extract of *Murraya paniculata* (L.)

Bacteria	Extract (g)	Clear zone diameter (mm)	Antimicrobial Zone Index
<i>Staphylococcus aureus</i>	0	19.5	1.75
	5	22.7	2.33
	10	23.5	3.07
	15	24.6	3.93
	20	24.9	3.98
	25	25.9	4.09
	Control +	31.5	4.26

Cytotoxicity Assay

The cytotoxic activity of *Murraya paniculata* (L.) leaf extract was evaluated using the Brine Shrimp Lethality Test (BSLT). No larval mortality was observed at 10 and 100 ppm, whereas complete mortality (100%) occurred at 1000 ppm, indicating a clear concentration-dependent toxic effect. The LC₅₀ value of 369.83 ppm indicates that the extract is moderately toxic.¹⁸ This activity is likely associated with the presence of flavonoid compounds, identified as the dominant constituents by FT-IR and GC-MS analyses.¹⁹ These findings suggest that *Murraya paniculata* (L.) leaves possess promising bioactivity and warrant further pharmacological investigation. The cytotoxic potential of *Murraya paniculata* (L.) leaf extract was evaluated using the BSLT. As shown in (Table-4), no mortality was observed at concentrations of 10 and 100 ppm, whereas complete mortality (100%) occurred at 1000 ppm. This concentration-dependent increase in larval mortality indicates that the extract possesses measurable cytotoxic activity.⁵

Table-4: Toxicity Test Extract Leaves of *Murraya paniculata* (L.)

C (ppm)	Number of test animals	Number of dead larvae per replication			Death rate	% Death	Average Probit Value
		1	2	3			
10	10	0	0	0	0	0 %	0
100	10	0	0	0	0	0 %	0
1000	10	10	10	10	10	100 %	8.09

CONCLUSION

Bioactive compounds from *Murraya paniculata* leaves were successfully extracted using the maceration method and characterised by phytochemical screening, FT-IR, and GC-MS analyses. The outcomes exhibited that flavonoids are the dominant compounds in the extract. The extract exhibited strong antioxidant activity (IC₅₀ = 12.64 ppm), significant antibacterial activity against *Staphylococcus aureus* (inhibition zone > 20 mm), and moderate cytotoxicity (LC₅₀ = 369.83 ppm). These findings highlight the potential of *Murraya paniculata* leaves as a natural source of antioxidant and antibacterial agents for further pharmaceutical development.

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CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

AUTHOR CONTRIBUTIONS

The present work is related to SDG-3: *Good Health and Well-being*. All the authors contributed significantly to this manuscript, participated in reviewing/editing and approved the final draft for publication. The research profile of the authors can be verified from their ORCID ids, given below:

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REFERENCES

1. T. Yulistyarini and J.T. Hadiah, *IOP Conference Series: Earth Environmental Science*, **724**, (2021), <https://doi.org/10.1088/1755-1315/724/1/012082>
2. S. Gea, Hariyati, Andriyani and K.M. Pasaribu, *International Journal of Ecophysiology*, **5**, 18(2023), <https://doi.org/10.32734/ijoe.v5i2.13459>
3. S. Chen, X. Wang, Y. Cheng, H. Gao, and X. Chen, *Molecules*, **28**, 1(2023), <https://doi.org/10.3390/molecules28134982>
4. M.C.S. Neta, C. Vittorazzi, A.C. Guimarães, J.D.L. Martins, M. Fronza, D.C. Endringer and R. Scherer, *Pharmaceutical Biology*, **55**, 190(2017), <https://doi.org/10.1080/13880209.2016.1254251>
5. Y. Qi, L. Wang, N. Wang, S. Wang, X. Zhu, T. Zhao and Q. Jiang, *Frontiers in Pharmacology*, **15**, (2024), <https://doi.org/10.3389/fphar.2024.1337161>
6. R. Hardiyanti, L. Marpaung, I.K. Adnyana and P. Simanjuntak, *Rasayan Journal of Chemistry*, **12**(3), 1822(2019), <https://doi.org/10.31788/RJC.2019.1235353>
7. W.H. Irham and R. Hardiyanti, *Rasayan Journal of Chemistry*, **14**(4), 2386(2021), <https://doi.org/10.31788/RJC.2021.1446345>
8. R. Hardiyanti, L. Marpaung, I.K. Adnyana and P. Simanjuntak, *Rasayan Journal of Chemistry*, **12**, 1569(2019), <https://doi.org/10.31788/RJC.2019.1235272>
9. S. Saragih, R. Hardiyanti and I.P. Mahendra, *Rasayan Journal of Chemistry*, **14**(1), 578(2021), <https://doi.org/10.31788/RJC.2021.1415883>
10. A. Armi, L.H. Pinem, Y. Setiawan, Y.D. Ayu, S. Hartati, L.O.M. Anwar and A. Rachmita, *Media Penelitian dan Pengembangan Kesehatan*, **35**, 1339(2025), <https://doi.org/10.34011/jmp2k.v35i4.2816>
11. M. Damayanti, A. Indarjo and S. Sedjati, *Journal of Marine Biotechnology and Immunology*, **3**, 49(2025), <https://doi.org/10.61741/jmbi.2025.v3.p49-54>
12. Q. Ma, L. Zhang, A. Nie, Y. Shi, Z. Liu, R. Zhang and Z. Sun, *Frontiers in Plant Science*, **16**, 1(2025), <https://doi.org/10.3389/fpls.2025.1717793>
13. J. An, Z. Zhang, A. Jin, M. Tan, S. Jiang and Y. Li, *Food Science and Nutrition*, **13**, (2025), <https://doi.org/10.1002/fsn3.70191>
14. T.S.B. Monir, S. Afroz, I. Jahan and T.Hossain, *Journal of Applied Life Science International*, **23**, 1(2020), <https://doi.org/10.9734/jalsi/2020/v23i430153>
15. N. Mohammadi, A.D.S. Lima, L. Azevedo and D. Granato, *Current Research in Food Science*, **8**, 100714(2024), <https://doi.org/10.1016/j.crfs.2024.100714>
16. J.G. Yang, B.G. Liu, G.Z. Liang and Z.X. Ning, *Molecules*, **14**, 46(2009), <https://doi.org/10.3390/molecules14010046>
17. M. Bubonja-Šonje, S. Knezević and M. Abram, *Arhiv za Higijenu Rada i Toksikologiju*, **71**, 300(2020), <https://doi.org/10.2478/aiht-2020-71-3396>
18. S. Fattahi, E. Zabihi, Z. Abedian, R. Pourbagher, A.M. Ardekani, A. Mostafazadeh and H. Akhavan-Niaki *International Journal of molecular and Cellular Medicine*, **3**, 102(2014).
19. S. Lenny, T. Barus, L. Marpaung and M.P. Nasution, *Malaysian Journal of Analytical Sciences*, **17**(2), 255(2013).

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