Trop J Nat Prod Res, January 2025; 9(1): 411 - 414 **ISSN 2616-0684 (Print)**
ISSN 2616-0692 (Electronic)

Tropical Journal of Natural Product Research

Available online at https://www.tjnpr.org

Original Research Article

Synergistic Effect of *Peperomia pellucida L. Kunth Aerial Part and* Amoxicillin against Methicillin-Resistant *Staphylococcus aureus* (MRSA)

Husnil Ania[1], Sartini Sartini[2], Herlina Rante[2]

[1]*Faculty of Pharmacy, Hasanuddin University, Makassar, South Sulawesi, 90245, Indonesia.*
[2]*Laboratory of Microbiology, Department of Pharmaceutical Science and Technology, Faculty of Pharmacy Hasanuddin University, 90245, Indonesia.*

ARTICLE INFO

Article history:
Received 13 January 2024
Revised 27 May 2024
Accepted 14 November 2024
Published online 01 February 2025

ABSTRACT

The combination of natural substances with antibiotics is a novel approach to combating antibiotic resistance. Several studies have shown that plant bioactive compounds can boost antibiotic action against pathogenic bacteria that are resistant to antibiotics. The aim of this study was to determine the synergistic effect of the aerial part of *Peperomia pellucida* (*P. pellucida*) with amoxicillin against Methicillin-Resistant *Staphylococcus aureus* (MRSA). *P. pellucida* aerial part extract was obtained by maceration in 70% ethanol followed by partitioning with n-hexane to give n-hexane soluble and n-hexane insoluble fractions. The phytochemical constituents of the extract and fractions were determined by thin layer chromatography (TLC) and spray reagents. The antibacterial activity of the extract and fractions as well as its synergistic effect with amoxicillin was assessed using the broth microdilution assay. The extract and fractions at 10 mg/mL demonstrated significant antibacterial activity against MRSA. The bacterial growth inhibition zone diameters were 9.33 ± 1.25 mm, 10.16 ± 0.28 mm, and 9.00 ± 1.00 mm for the ethanol extract, n-hexane soluble fraction, and n-hexane insoluble fraction of *P. pellucida*, respectively. The hexane soluble fraction of *P. pellucida* showed synergistic effect with amoxicillin against MRSA, resulting in a Fractional Inhibitory Concentration Index (FICI) of 0.25. The results obtained from this study suggest that *P. pellucida* n-hexane fraction could enhance the antibacterial effect of amoxicillin against methicillin-resistant *Staphylococcus aureus*.

Keywords: Synergy, *Peperomia pellucida*, Amoxicillin, Synergism, Methicillin-resistant *Staphylococcus aureus.*

Introduction

Antimicrobial resistance (AMR) has been a major healthcare challenge worldwide, and antimicrobial-resistant bacteria cause significant morbidity and mortality.[1] AMR has emerged rapidly on a global scale in recent years, spreading faster than previously thought.[2] According to the World Health Organization (WHO), antibiotic resistance was one of the world's top ten health issues in 2019.[3] Antimicrobial drugs play an important role in the prevention and treatment of infectious diseases.[4]

Methicillin-resistant *Staphylococcus aureus* (MRSA) is a group of gram-positive bacteria that is genetically distinct from other strains of *Staphylococcus aureus*. MRSA is responsible for several difficult-to-treat infections in humans. MRSA mostly affects the skin and delicate tissues, and it is responsible for about 15 to 60 percent mortality from infectious illnesses.[5] MRSA may infect both children and adults of all ages. According to various American polls, it was found that up to 30 percent of youngsters have MRSA on their skin or nose, and a majority of them were unaware that they habour MRSA. Many important opportunistic infections have resulted from the presence of MRSA on the skin, nose and other tissues.[6]

*Corresponding author. E mail: husnilaniawahid@gmail.com
Tel: 082151286262

Citation: Husnil A, Sartini S., Herlina R. Synergistic Effect of Peperomia pellucida L. Kunth Aerial Part and Amoxicillin against Methicillin-Resistant Staphylococcus aureus (MRSA). Trop J Nat Prod Res. 2025; 9(1): 411 – 414 https://doi.org/10.26538/tjnpr/v9i1.52

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria.

MRSA bacteria are resistant to practically all β-lactam antibiotics, and this resistance can range from mild to severe. Amoxicillin is a β-lactam antibiotic that is still commonly used due to its high gastrointestinal absorption; however there has been resistance to this antibiotic in many people. The antibiotic amoxicillin has a resistance rate of 67.6%.[5] *Staphylococcus aureus* isolated from humans and animals in Indonesia has amoxicillin resistance of 95.52%.[6,7]

The rise in amoxicillin-resistant bacteria is the leading cause of treatment failure and an increase in mortality rate. Therefore, it is critical to produce antibacterial agents that do not only reduce drug resistance but also improve treatment outcomes.[8]

A new medicine or therapeutic strategy against MRSA is desperately needed.[9,10] Many studies have been undertaken to examine natural compounds as novel alternatives against MRSA and to produce effective antibiotics for tackling the challenges associated with MRSA.[11-13]

Antibacterial activities of *Peperomia pellucida* have been demonstrated in numerous studies.[14-16] Many bioactive substances such as proteins, tannins, glycosides, steroids, flavonoids, alkaloids, phenols, terpenes, essential oils, and saponins have been found in the plant through phytochemical studies.[17,18] The present study therefore seek to investigate the concentration of *Peperomia pellucida* extract that can potentially enhance the antibacterial activity of amoxicillin against methicillin-resistant Staphylococcus aureus (MRSA).

Materials and Methods

Bacterial strain

Clinical isolates (obtained from pus specimen from the wound of the patient) of methicillin resistant *Staphylococcus aureus* were collected from Microbiology Laboratory of Hasanuddin University Hospital, Makassar, Indonesia.

Drugs and Chemicals
Amoxicillin (Supelco), Hexane (Merck), Ethyl acetate (Merck), sulfuric acid (Merck), DMSO (Merck), TSA (Triptic Soy Agar) (Oxoid®), TSB (Triptic Soy Broth) (Oxoid®), Triphenyltetrazolium chloride (Merck KGaA , 64271 Darmstatd®). Dragendorff, Lieberman Burchard, Iron(III) chloride ($FeCl_3$), and Aluminium chloride ($AlCl_3$).

Plant collection and identification
Peperomia pellucida herbaceous plant (aerial part) was obtained from a commercial market in Makassar, South Sulawesi Indonesia. The plant material was identified in the Department of Biology, Faculty of Mathematics and Natural Science, Universitas Hasanuddin makassar, Sounth Sulawesi, Indonesia.
Preparation of extract and fractions
Peperomia pellucida herbal powder was macerated with 70% ethanol at sample to solvent ration of 1:5, at room temperature for 3 days. The ethanol extract was subjected to liquid-liquid partitioning with n-hexane. Briefly, the extract was weighed and dissolved in ethanol, then the extract solution was poured into a separating funnel, and n-hexane was added, homogenized, and allowed to stand for a few minutes until separation occurred. The n-hexane-soluble and n-hexane-insoluble fractions were collected into separate containers and then evaporated to dryness.
Phytochemical screening
The phytochemical constituents of the extract and fractions were determined by thin layer chromatography (TLC). TLC was carried out by spotting the stock solution of the extract and fractions on a silica gel 60 F254 plate, then eluting with hexane: ethyl acetate (7:3). The phytochemicals on the TLC chromatogram were identified by colour changes after spraying with several spray reagents, namely; Dragendorff's, Lieberman Burchard, Iron (III) chloride ($FeCl_3$), and aluminium chloride ($AlCl_3$) reagents.
Determination of Antibacterial activity
The antibacterial activity of *P. pellucida* herb extract and fractions was determined using the disk diffusion technique. The extract and fractions (50 mg each) was dissolved in 5 mL of 10% DMSO. Then 20 µL of each solution was pipetted and dropped onto a blank paper disk and allowed to dry. After drying, the disk was placed on the surface of Triptic Soy Agar (TSA) medium containing suspension of MRSA at 10^6 CFU/mL, incubated at 37°C for 24 h. The diameter of the inhibition zone formed was measured.[14] The extract/fraction with the highest inhibition zone diameter was used in the synergistic test with amoxicillin. Erythromycin was used as the positive control while DMSO (10%) was used as the negative control.
Determination of Minimum Inhibitory Concentration (MIC) of P. pellucida hexane-soluble fraction
The MIC of *P. pellucida* hexane-soluble fraction was evaluated using the microdilution technique in Tryptic Soy Broth (TSB) media. The concentrations of the fraction ranged from 20 to 0.625 mg/mL. Each 96 well microplate contained extract stock, TSB media and MRSA bacterial culture (equivalent to 0.5 Mc Farland 1.5×10^6 CFU/mL). The plate was incubate at 37°C for 24 h. Few drops of 2,3,5-Triphenyltetrazolium chloride reagent was added to each well, and the microplate was further incubated for another 30 min. Visual observations were made. The MIC of the extract was determined as the lowest concentration that does not exhibit bacterial growth or a red colour change.[19]

Determination of Minimum Inhibitory Concentration (MIC) of Amoxicillin
The MIC of amoxicillin was also assessed using the microdilution technique in Tryptic Soy Broth (TSB) media. Concentrations of amoxicillin ranging from 0.016 to 0.0005 mg/mL were used. Each well of the 96 well microplate contained extract stock, amoxicillin, a combination of extract and amoxicillin, TSB media and MRSA bacterial culture (equivalent to 0.5 Mc Farland 1.5×10^6 CFU/mL). The plate was incubate at 37°C for 24 h. Few drops of 2,3,5-Triphenyltetrazolium chloride reagent was added to each well, and the microplate was further incubated for another 30 min. Visual observations were made.

Test for Synergistic Activity
The test for synergistic activity between *P. pellucida* hexane-soluble fraction and amoxicillin against MRSA was determined using the checkerboard assay method.[11] The concentration of the hexane-soluble fraction ranged from 20 to 0.625 mg/mL while that of amoxicillin ranged from 0.016 to 0.00025 mg/mL. Each 96 well microplate contained stock extract, amoxicillin, TSB medium and MRSA bacterial culture (equivalent to 0.5 Mc Farland 1.5×10^6 CFU/mL). the plate was incubated at 37°C for 24 h. The MIC is the lowest concentration that does not exhibit bacterial growth or a red color change. Following MIC, a synergistic effect test was conducted utilizing the Fractional Inhibitory Concentration Index (FICI). The FICI is calculated using the formula below.[19]

$$\text{FIC} = FIC_A + FIC_B = \frac{\text{MIC of plant extract in combination}}{\text{MIC plant extract only}} + \frac{\text{MIC of plant extract in combination}}{\text{MIC plant extract only}}$$

Where; FICI ≤ 0.5 is stated to have a synergistic effect; FICI > 0.5 to1 is denoted additive; FICI>1 to 4 is denoted indifferent; FICI > 4 is indicated as having an antagonistic effect.
Data analysis
The inhibition zone diameter (IZD) was reported as mean ± standard deviation (SD). The Fractional Inhibitory Concentration Index (FICI) was calculated from the results obtained from the MIC.

Results and Discussion

Percentage yields of P. pellucida extracts and fractions
Extraction of the *P. pellucida* herb was carried out using the maceration method with 70% ethanol as the solvent (1:5). The percentage yield of the extract was 9.90% (Table 1). However, a higher percentage yield of 18.28% was reported in a previous study by Adomi and Idudun (2023).[20] The variation in the percentage yield obtained in the different studies might have been influenced by differences in the amount of powdered plant materials used, the solvent and extraction method. The percentage yields obtained for the hexane-soluble and hexane insoluble fractions were 57.22% and 27.69%, respectively (Table 2).

Table 1: Percentage Yield of *P. pellucida* ethanol extract

Consistency	Weight of powdered sample (g)	Extract weight (g)	% Yield
Thick	220.12	21.81	9.90

Table 2: Percentage yield from partition of *P. pellucida* herb extract

Fraction	Extract weight (g)	% Yield
Hexane Soluble	12.49	57.22
Hexane Insoluble	6.04	27.69

Phytochemical constituents of P. pellucida extract and fractions
The result of the phytochemical screening of *P. pellucida* extract and fractions is presented in Table 3. The result shows that *P. pellucida* contains flavonoids, tannins, steroids and terpenoids. Phytochemical analysis of the initial extract (70% ethanol) and hexane insoluble fraction showed the presence of tannins, steroids and terpenoids. Previous phytochemical screening tests have shown the presence of a variety of phytoconstituents in *P. pellucida*. Some of the constituents previously found in the plant include glycosides, flavonoids, tannins

and steroids/triterpenoids.[21] The work of Ling *et al.* (2021)[22] found phenolic compounds, terpenoids, saponins, proteins and glycosides in *P. pellucida* extract. The slight variation in the results obtained from the different studies may be ascribed to several factors, such as differences in solvents used for extraction, extraction methods, screening reagents used, differences in geographic locations, and environmental conditions.

Table 3: Phytochemical constituents of *Peperomia pellucida* extract and fractions

Compound	Reagent used	Colour of Stain	Extract/Fraction		
			Ethanol Extract	**Hexane Soluble Fraction**	**Hexane Insoluble Fraction**
Alkaloids	Dragendorff	Orange	-	-	-
Flavonoids	$AlCl_3$	Yellow	-	+	-
Phenolic		Blue black	-	-	-
Tannin	$FeCl_3$	Greenish	+	-	+
Phenolic		brown		-	-
		Bluish Black	-		
Steroids and terpenes	Liberman Burchard	Blue and green in interphase	+	+	+

(+) means presence of the chemical; (-) means absence of the chemical

Antibacterial activity of P. pellucida herb extract

The results of the antibacterial activity test are shown in Table 4 and Figure 1. The results showed that the 70% ethanol extract, hexane soluble fraction and hexane insoluble fraction of *P. pellucida* exhibited antibacterial activity against MRSA. The diameter of the inhibition zone of the 70% ethanol extract was 9.33 ± 1.25 mm, while the hexane soluble and hexane insoluble fractions gave inhibition zone diameter of 10.16 ± 0.28 mm and 9.00 ± 1.00, respectively. Previous study by Ibrahim and Yahya (2020)[14] found that the diameter of the inhibition zone of *P. pellucida* ethanol extract at the highest concentration against *S. aureus* was 10.67 ± 0.58 mm. However, in this study, the extract of *P. pellucida* was used as a modulator to enhance the antibacterial effect of amoxicillin an antibiotic to which several bacteria have shown resistance to.

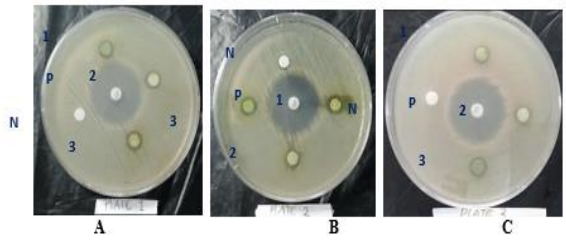

Figure 1: Antibacterial activity of *P. pellucida* extract and fractions against MRSA. **N:** Negative control (10% DMSO), **P:** Positive control (Erythromycin), **1:** 70% ethanol extract, **2:** Hexane soluble fraction, **3:** Hexane insoluble fraction. **(A):** Replicate 1, **(B):** Replicate 2, and **(C):** Replicate 3

Previous researches have shown that flavonoids, tannins, and terpenoids have antibacterial activity against MRSA.[23-25] In addition, the astringent property of tannins contribute to their antibacterial activity, they shrink cell wall, damage bacterial cell membranes and disrupt cell permeability, resulting in bacterial cell death. Other compounds like flavonoids exhibit their antibacterial activity by disrupting the structure of bacterial cell wall by interacting with extracellular protein and inhibiting bacterial mobility.[5] The mechanism of action of terpenoids as antibacterials is by lysis of bacterial cell walls. Terpenoids generate strong polymer connections with porins (transmembrane proteins) on the bacterial cell wall, and destroys the porins. Damage to porins limit the entry and exit of essential nutrients, resulting in bacterial cell wall destruction and eventual cell death.[25]

Minimum Inhibitory Concentration (MIC) and Synergistic effect of P. pellucida and amoxicillin

The test for synergism between *P. pellucida* (n-hexane soluble fraction) and amoxicillin was carried out by measuring the MIC of *P. pellucida*, amoxicillin, and a combination of both. Table 5 shows the MICs of the individual samples and their combination. The MIC of *P. pellucida* n-hexane soluble fraction was 10 mg/mL, while that of amoxicillin was 0.016 mg/mL. The MIC of the combination of *P. pellucida* n-hexane soluble fraction and amoxicillin was 1.25 mg/mL. This indicates that *P. pellucida* herb extract could decrease the MIC of amoxicillin, which might improve the antibacterial impact against MRSA. According to the Clinical Laboratory Standards Institute (CLSI), bacterial resistance to penicillin class of antibiotics is confirmed if the minimum inhibitory concentration (MIC) against *Staphylococcus aureus* is ≥ 0.00025 mg/mL. So it can be concluded that the test bacteria (MRSA) used in this study is truly resistant.

Also shown in Table 5 is the fractional inhibitory concentration index (FICI). A FICI value of 0.25 was found when *P. pellucida* n-hexane soluble fraction was combined with amoxicillin, revealing a synergistic effect. This study showed that the herb extract combined with the antibiotic amoxicillin inhibited the growth of the bacteria tested at lower concentrations than when the single drugs were tested separately. The combined effect of active chemicals from extracts and antibiotics resulted in synergistic interactions. Antimicrobial substances might limit microbial growth in processes distinct from those generally known in antimicrobial medicines, and they may have significant clinical benefit in the treatment of microbial resistance

Table 4: Antibacterial activity of *P. pellucida* extract and fractions

Extract	Inhibition zone diameter (mm)
Ethanol 70%	9.33 ± 1.25

Insoluble hexane	9.00 ± 1.00
Soluble hexane	10.16 ± 0.28

Table 5: Minimum inhibitory concentration (MIC) of n-hexane fraction of *P. pellucida* in combination with amoxicillin and FICI values

Sample	MIC (mg/mL)	FICI
P. pellucida n-hexane fraction	10	
Amoxicillin	0.016	
MIC of *P. pellucida* n-hexane fraction in the combination of *P. pellucida* and Amoxicillin	1.25	0.25
MIC Amoxicillin in the combination of *P. pellucida* and Amoxicillin	0.002	

Conclusion

The combination of *P. pellucida* n-hexane fraction with the antibiotic amoxicillin greatly reduced the MIC of the individual samples against MRSA. The Fractional Inhibitory Concentration Index (FICI) was found to be 0.25, indicating a synergistic effect between *P. pellucida* n-hexane fraction and amoxicillin. These findings therefore suggest that *P. pellucida* n-hexane fraction may enhance the antibacterial action of amoxicillin against methicillin-resistant *Staphylococcus aureus* (MRSA) in clinical setting.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

References

1. Micoli F, Bagnoli F, Rappuoli R, Serruto D. The role of vaccines in combatting antimicrobial resistance. Nat Rev Microbiol. 2021; 19(5):287-302.
2. Christaki E, Marcou M, Tofarides A. Antimicrobial Resistance in Bacteria: Mechanisms, Evolution, and Persistence. J Mol Evol. 2020; 88(1):26-40.
3. White A and Hughes JM. Critical Importance of a One Health Approach to Antimicrobial Resistance. Ecohealth. 2019; 16(3):404-409.
4. Abushaheen MA, Muzaheed, Fatani AJ, Alosaimi M, Mansy W, George M, Acharya S, Rathod S, Divakar DD, Jhugroo C, Vellappally S, Khan AA, Shaik J, Jhugroo P. Antimicrobial resistance, mechanisms and its clinical significance. Dis Mon. 2020; 66(6):100971.
5. Panchal VV, Griffiths C, Mosaei H, Bilyk B, Sutton JAF, Carnell OT, Hornby DP, Green J, Hobbs JK, Kelley WL, Zenkin N, Foster SJ. Evolving MRSA: High-level β-lactam resistance in *Staphylococcus aureus* is associated with RNA Polymerase alterations and fine tuning of gene expression. PLoS Pathog. 2020;16(7):e1008672.
6. Nandhini P, Kumar P, Mickymaray S, Alothaim AS, Somasundaram J, Rajan M. Recent Developments in Methicillin-Resistant Staphylococcus aureus (MRSA) Treatment: A Review. Antibiotics. 2022; 11(5):1-21.
7. Fitranda M, Salasia SIO, Sianipar O, Dewananda DA, Arjana AZ, Aziz F, Wasissa M, Lestari FB, Santosa CM. Methicillin-resistant Staphylococcus aureus isolates derived from humans and animals in Yogyakarta, Indonesia. Vet World. 2023; 16(1):239-245.
8. Saquib SA, AlQahtani NA, Ahmad I, Arora S, Asif SM, Javali MA, Nisar N. Synergistic antibacterial activity of herbal extracts with antibiotics on bacteria responsible for periodontitis. J Infect Dev Ctries. 2021; 15(11):1685-1693.
9. Zhang W, Margarita GE, Wu D, Yuan W, Yan S, Qi S, Xue X, Wang K, Wu L. Antibacterial Activity of Chinese Red Propolis against *Staphylococcus aureus* and MRSA. Molecules. 2022;27(5):1-17.
10. Buzgaia N, Awin T, Elabbar F, Abdusalam K, Lee SY, Rukayadi Y, Abas F, Shaari K. Antibacterial activity of arbutus pavarii pamp against methicillin-resistant staphylococcus aureus (MRSA) and uhplc-ms/ms profile of the bioactive fraction. Plants. 2020; 9(11):1-16.
11. Sartini S, Natsir Djide M, Amir MN, Permana AD. Phenolic-rich green tea extract increases the antibacterial activity of amoxicillin against Staphylococcus aureus by in vitro and ex vivo studies. J Pharm Pharmacogn Res. 2020; 8(6):491-500.
12. Kwiatkowski P, Łopusiewicz Ł, Pruss A, Kostek M, Sienkiewicz M, Bonikowski R, Koszko IW, Dołęgowska B. Antibacterial activity of selected essential oil compounds alone and in combination with β-lactam antibiotics against MRSA strains. Int J Mol Sci. 2020; 21(19):1-15.
13. Bao M, Zhang L, Liu B, Li L, Zhang Y, Zhao H, Ji X, Chen Q, Hu M, Bai J, Pang G, Yi J, Tan Y, Lu C. Synergistic effects of anti-MRSA herbal extracts combined with antibiotics. Future Microbiol. 2020; 15(13):1265-1276.
14. Ibrahim MA and Yahaya NAM. Phytochemical Screening and Antibacterial Activity Determination of *Peperomia pellucida* Extract. Trop Agrobiodivers. 2020; 1(1):04-06.
15. Tablang JO, Ron PCC, Jacob JKS. Phytochemical Screening and Antibacterial Properties of Silverbush (*Peperomia pellucida*) against Selected Cultured Bacteria. Glob J Med Plant Res. 2020;8(1):1-6.
16. Teoh L, Gnanasegaran N, Adnan AFM, Taha RM. The comparative antimicrobial and anticancer of chemical extract from in vitro and in vivo peperomia pellucida plantlet. J Appl Biol Biotechnol. 2021; 9(2):115-123.
17. Babu YTR, Satapathy P, Krishna CM, Ramalakshmana J, Padal SB. GC-MS analysis and screening of antimicrobial potentialities of the medicinal herb *Peperomia pellucida* (L .) Kunth. 2023;17(1001):1376-1389.
18. Farid N, Meinar Dwi Rantisari Thayeb A, Saleh A. Formulation and Test of Antibacterial Activity Gel Mask Peel Off Stem and Suruhan Leaves (Peperomia Pellucida L.) Extract Against Propionibacterium Acne under a Creative Commons Attribution-NonCommercial 4.0 International License (CC BY-NC 4.0). J Eduhealth. 2022; 13(02):2022.
19. Abdulabbas SS, Ibrahim ZK, Muhammed HA, AL-Khazali NAY, Ahmed ST, Ai- Tuma AF, Mahdi FMS. Determination of Fractional Inhibitory Concentration (FIC) Index as A Measure of Synergy of Antibiotics in *E. coli* O157:H7. Egypt J Hosp Med. 2022; 89(2):7571-7575.
20. Adomi PO and Idundun R. Evaluation of Antimicrobial Activities of Medicinal Plants Used against Microbial Infections in Pregnant Women in Delta State, Nigeria. 2023; 23(9):9-17.
21. Silahooy C. Characterization of Alluvial Soil for Growth of Chinese Betel (*Peperomia pellucida* L.). Int J Multidiscip Appl Bus Educ Res. 2023; 4(5):1476-1482.
22. Vol RDP. Water Extract of Leaves of *Peperomia pellucida* by Cold. 2021; 7(4):64-68.
23. Shamsudin NF, Ahmed QU, Mahmood S, Ali Shah SA, Khatib A, Mukhtar S, Alsharif MA, Parveen H, Zakaria ZA. Antibacterial Effects of Flavonoids and Their Structure-Activity Relationship Study: A Comparative Interpretation. Molecules. 2022; 27(4):1149.
24. Sivasamugham LA, Nimalan V, Subramaniam G. Antibacterial effects of Musa sp. ethanolic leaf extracts against methicillin-resistant and susceptible *Staphylococcus aureus*. South African J Chem Eng. 2021; 35(2020):107-110.
25. Apridamayanti P, Sari R, Rachmaningtyas A, Aranthi V. Antioxidant, antibacterial activity and FICI (Fractional Inhibitory Concentration Index) of ethanolic extract of *Melastoma malabathricum* leaves with amoxicillin against pathogenic bacteria. Nusant Biosci. 2021; 13(2):140-147.

ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)