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Research Article

Two Sesquiterpenes from *n*-Hexane Fraction of *Curcuma soloensis* Rhizomes and Their Antimicrobial Activities

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Abstract

Curcuma soloensis is one of the medicinal plants that has the potential to be a source of bioactive compounds. The antimicrobial study of the bioactive compounds from *C. soloensis* was still limited. This study aimed to isolate the bioactive compounds from *C. soloensis* rhizomes and to evaluate their potential as antimicrobial agents. *C. soloensis* rhizome extraction was done using the maceration method with acetone and then fractionated with *n*-hexane: methanol (1:1). The compounds were separated and purified using vacuum liquid chromatography and radial chromatography. The structure of the isolated compounds was determined using the nuclear magnetic resonance (^{1}H and ^{13}C NMR) spectroscopy analysis and comparison with literature data. Antimicrobial activity assays of the extract, *n*-hexane fraction, and isolated compounds were carried out by microdilution methods against *Escherichia coli*, *Staphylococcus aureus*, *Candida albicans*, and *Malaszezia furfur*. Two bioactive compounds from the *n*-hexane extract of *C. soloensis* rhizome have been isolated: ar-curcumene and *ar*-turmerone. Antimicrobial test results on acetone extract, *n*-hexane fraction, and isolated compounds showed that *ar*-turmerone had the highest activity against *S. aureus* with a MIC value of 15.6 µg / mL. In comparison, *ar*-curcumene showed the same activity against all test microbes with a MIC value of 62.5 µg / mL. This study showed that secondary metabolite compounds of *C. soloensis* rhizomes have the potential to be developed as antimicrobial agents.

Keywords: Antimicrobial; *ar*-curcumene; *ar*-turmerone; *C. soloensis*

1. INTRODUCTION

Antibiotics, chemical compounds of microorganisms' metabolic products, are antimicrobial drugs that have been seen as a 'blessing' for mankind since more than half a century ago. But lately, the ability of these antibiotic compounds has begun to decline because the targeted microorganisms have developed immunity or resistance gradually [1]. On the other hand, the human need for antibiotics is not only focused on treating diseases but also on their prevention, particularly concerning their transmission through food. This fact motivates intense research efforts to obtain novel antibiotics. Antimicrobial compounds derived from natural products are now again being used. Natural antimicrobial agents are distinguished by their harmless nature, safety, and diversity of structures [2].

Indonesia as a region with a diversity of plant species allows the discovery of antimicrobial active compounds from Indonesian medicinal plants, one of which is from the genus *Curcuma* (family Zingiberaceae). The Curcuma genus is an important medicinal plant in Indonesia because more than 2,031 traditional medicine formulas (6.34% of the formulas identified) used species of Curcuma rhizomes [3]. The study of Curcuma's constituent has revealed a variety of biological properties, including antibacterial, anti-inflammatory, hepatoprotective, anti-cholesterol, antioxidant, and anticancer properties. [4-5]. Slightly more

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than 20 of the 110 species in the genus Curcuma have been investigated phytochemically; *C. longa*, *C. xanthorrhiza*, and *C. zedoaria* were the most extensively researched. [4].

One species of Curcuma that has not been studied scientifically but has potential as a medicinal plant *is Curcuma soloensis*. This plant thrives in Indonesia with the local name 'temu tis' or 'temu glenyeh'. *C. soloensis* rhizome is usually used with other herbs for the treatment of skin infections [6]. The rhizomes *of C. soloensis* are also used by people for dermatological disorders, especially wounds and burns [7].

Some studies report that the *n*-hexane extract of *C. soloensis rhizomes* has biological activity as a gastroprotective [7], and anticancer [8,9]. Acetone extract of *C. soloensis* rhizomes also has antifungal activity against *Aspergillus fumigatus, Candida albicans, Epidermophyton sp, Penicillum sp,* and *Trichophyton rubrum* [10]. The essential oil of *C. soloensis* rhizomes was also reported to have an antiproliferative effect against human carcinoma cell lines [6] and inhibit the growth of *Staphylococcus aureus, S. epidermis,* and *S. haemolyticus* [11]. The ethanol extract of *C. soloensis* showed antimicrobial activity against *Streptococcus mutans* and *C. albicans*[12]. Some compounds successfully isolated from the rhizomes of *C. soloensis* are bisacurone, curcumin [13], and *ar*-turmerone [14]. These compounds have been isolated from the *C. domestica* rhizomes and showed antimicrobial activity against some bacteria[15]. Based on some previous research, it appears that there was a potential for bioactive compounds of *C. soloensis* rhizomes as antimicrobials, but has not received more specific studies, related to bioactive compounds that act as antimicrobials.

In this research, the antimicrobial activity of bioactive compounds isolated from rhizomes of *C. soloensis* has been studied. Antimicrobial testing was carried out on *Escherichia coli*, *S. aureus*, *C. albicans*, and *Malassezzia furfur* using the microdilution method. Identification of isolated compounds was carried out using NMR (*Nuclear Resonance Magnetic*) spectroscopy, so we have the information on bioactive compounds that have the potential as antimicrobial.

2. RESEARCH METHODS

Materials and Instruments

Plant material (*C. soloensis* rhizomes) was collected from Bandung of West Java, Indonesia, and identified at the Environmental Laboratory of the Faculty of Biology, Universitas Jenderal Soedirman. The tested microbes (*E. coli, S. aureus, C. albicans, M. furfur*), were collected from the Microbiology Laboratory Faculty of Medicine, Universitas Jenderal Soedirman.

The instruments used in this research were a microplate spectrophotometer for antimicrobial analysis (Automatic Microplate Laser: All Sheng, AMR-100), ^{1}H NMR and ^{13}C NMR spectrometer for the identification of isolated compounds (Agilent DD2: 500 (^{1}H) MHz and 125 (^{13}C) MHz, solvent $CDCl_3$).

Procedures

Isolation of compounds

The rhizome of *C. soloensis* (8 kg) was washed, cut into thin and small pieces then dried by drying or oven at low temperature (30-40 ºC), and then mashed until it became powder. The powder (1.1 kg) was macerated with acetone for 3x24 hours. Every 24 hours the extract was taken by filtering, and the residue from the filtering was macerated again until three repetitions. The acetone extract was concentrated using a rotary evaporator. The concentrated acetone extract (107 g) was partitioned with *n*-hexane: methanol (1:1), and the fraction of n-hexane was accommodated and then concentrated with a rotary evaporator. The *n*-hexane fraction (18 g) was fractionated by vacuum liquid chromatography (VLC) using silica gel, and eluent of *n*-hexane, a mixture of n-hexane and chloroform (8:2, 6:4, 4:6, 2:8), and chloroform. The fractions that showed the same spot profile in the TLC that were observed with UV lamps were then combined and obtained four subfractions namely Fr.1(8.9 g), Fr.2 (2.25 g), Fr.3 (0.7 g) and Fr.4 (0.9 g). Two main fractions (Fr.1 and Fr.2) were further separated through radial chromatography until the pure compound was obtained. A compound was said to be pure when it showed a single spot on the TLC. The fraction of Fr.1 was purified by radial chromatography using n-hexane eluents, and a pure compound (1) in the form of colorless oil was obtained (56 mg). While the Fr.2 fraction was purified using an eluent of n-hexane: chloroform (9:1), a pure compound (2) in the form of yellowish clear oil was obtained (207 mg). Both isolates were then analyzed for their chemical structure by NMR spectroscopy and tested for antimicrobial activity.

Determination of the structure of the compound

The structure of the isolated compound was determined by ^{1}H NMR, ^{13}C NMR, and HSQC (Heteronuclear Single Quantum Coherence) spectrometers. The isolated compound has been known, so the data obtained was compared with literature data.

Antimicrobial assays by microdilution method [16]

Antimicrobial activity assays were carried out by liquid dilution method. The concentration of the sample stock solution was 1000 µg /mL. The concentration of test samples used was 500, 250, 125, 62.5, 31.25, 15.62, and 7.81 µg/mL. The sample was dissolved in 10% DMSO.

Microbes were cultured for 24 hours for bacteria and 48 hours for fungi at 27 ºC under aerobic conditions in an agar medium (Mueller Hinton Agar (MHA) for bacteria and Potato Dextrose Agar (PDA) for fungi). The

microbes were then suspended in 0.9% NaCl solution (w/v) and the density was equalized to 0.5 Mc Farland.

A total of 200 µL of liquid media (Mueller Hinton Broth (MHB) for bacteria and Potato Dextrose Broth (PDB) for fungi was filled into each microplate well (96 wells). The sample's stock solution (1000 µg/mL) was introduced in 200 µL to the first well. To conduct the solution concentration series, 200 µL of solution was transferred from the first well to the second, and then 200 µL of solution was withdrawn from the second well and put into the third, and so on up to the eighth well. The amount of solution in each well was 200 µL. Further, into each well was inserted 10 µL of microbial suspension. The microplate was then incubated at 37 ºC for 24 hours. Microbial growth was determined using a microplate spectrometer at a wavelength of 600 nm. The minimum inhibitory concentration (MIC) is the lowest concentration that can inhibit microbial growth. This experiment was carried out twice. As positive controls, amoxicillin was utilized for bacteria, and miconazole was used for fungus.

3. RESULTS AND DISCUSSION

Isolated Compounds

The results of physical properties observations and spectroscopic measurements (^{1}H and ^{13}C NMR) of isolated compounds obtained two known sesquiterpene compounds, namely *ar*-curcumene (1) and *ar*-turmerone (2), in **Figure 1.**

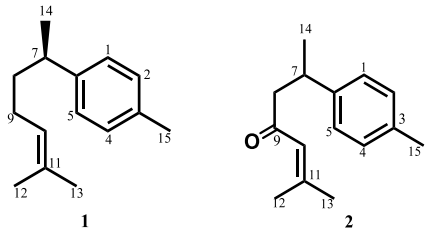

Figure 1. *ar*-Curcumene (**1**) and *ar*-turmerone (**2**)

ar-Curcumene

ar-Curcumene (**1**) is a sesquiterpene compound with a bisabolene-type skeleton (**Figure 1**). This compound is in the form of colorless oil.

The analysis of ^{1}H NMR spectral data of compound **1**, showed the presence of four methyl signals (δH 1.22 (d, *J* = 6.9 Hz); 1.53 (s*); 1.68* (s) and 2.33 (*s*) *ppm*), two proton signals of para-substituted methylbenzene (*δH* 7.06 (d, *J* = 8.0 Hz) and 7.10 *(*d, *J* = 8.0 Hz), two methylene signals (δ_H 1.56-1.62 (m) and 1.88-1.99 *(m*) ppm), one methine signal (δ_H 2.66 (m*)* ppm) and one vinilic proton signal *(*δ_H *5.11 (*t, *J = 6.9* Hz) ppm). The spectral data of ^{13}C NMR compound **1** showed the presence of 13 signals that representing 15 carbons, consisting of seven C-sp3, namely four methyl carbons (δ_C 17.9; 21.2; 22.7 and 25.9 ppm), two methylene carbons (δ_C 26.4 and 38.5 ppm) and one methine carbon (δ_C 39.2 ppm), and eight C-sp2 atoms from six aromatic carbons (δ_C 127.3 (2C); 129.0 (2C); 135.5 and 144.6 ppm) and two aliphatic carbons (δ_C 124.6 and 131.5 ppm).

Table 1. Spectral data of ^{1}H, ^{13}C-NMR and HSQC *ar*-curcumene

C atomic position	δH (*much*. *J* in Hz) ppm		δC in ppm	
	1	**1***	**1**	**1***
1	7.06 (1H, d, 8.0)	7.03 (1H, *d*, 7.8)	127.3	126.9
2	7.10 (1H, d, 8.0)	7.08 (1H, *d*, 7.8)	129.0	129.0
3	-	-	135.5	135.1
4	7.10 (1H, *d*, 8.0)	7.08 (1H, *d*, 7.8)	129.0	128.9
5	7.06 (1H, *d*, 8.0)	7.03 (1H, *d*, 7.8)	127.3	126.9
6	-	-	144.6	144.6
7	2.66 (1H, *m*)	2.60 (1H, *m*)	39.2	39.0
8	1.56-1.62 (2H, *m*)	1.60-1.67(2H, *m*)	26.4	26.2
9	1.88-1.99 (2H, *m*)	1.80-1.95(2H, *m*)	38.5	38.5
10	5.11 (1H, *t*, 6,9)	5.10 (1H, *t*, 6,9)	124.6	124.6
11	-		131.5	131.3
12	1.69 (3H, *s*)	1.68 (3H, *s*)	25.9	25.7
13	1.53 (3H, *s*)	1.53 (3H, *s*)	17.8	17.6
14	1.22 (3H, *d*, 6.9)	1.21 (3H, *d*, 7.0)	22.6	22.4
15	2.32 (3H, *s*)	2.32 (3H, *s*)	21.1	20.9

1: isolated compound, in $CDCl_3$ 500 MHz (^{1}H) and 125 MHz (^{13}C);
1* : [17]

These signals indicated that the compound is a sesquiterpene compound with a bisabolane skeleton. Based on these spectroscopic data, compound **1** is *ar*-curcumene and has the molecular formula $C_{15}H_{22}$. Comparison of ^{1}H and ^{13}C NMR data of compound **1** (**Table 1**) shows similar to previously reported *ar*-curcumene data [17].

ar-Turmerone

ar-Turmerone (**2**) is also a sesquiterpene with a bisabolene skeleton. This compound is a yellowish oil with a chemical structure as shown in **Figure 1**.

^{1}H NMR spectral data of compound **2** showed the presence of four methyl proton signals at C-12 (δH 2.12 ppm, s); C-13 (δ_H 1.88 ppm, s); C-14 (δ_H 1.31 ppm, d, *J* = 7.2 Hz); and C-15 (δ_H 2.33 ppm, s), four proton signals from the benzene ring substituted at positions 3 and 6 (C-1 and C-5, δ_H 7.07 ppm, d, *J* = 1.3 Hz); C-2 and C-4, δ_H 7.10 ppm (d, *J* = 7.8 Hz); one methylene signal (C-8, δH 2.62-2.87 ppm, m); one methine signal (C-7, δ_H 3.26, ppm, *sext*, *J* = 7.2 Hz); as well as one vinylic proton signal (C-10, δ_H 5.81 ppm, s). Based on 13C NMR spectral data of compound **2** showed the presence of fifteen signals representing six C-sp3 atoms, namely at C-7, C-8, C-12, C-13, C-14, and C-15, and nine C-sp2 atoms including one C-carbonyl (δc 199.7 ppm).

The signals from the NMR spectrum indicate that compound **2** has the same skeleton as compound **1**, namely bisabolene. The difference between compounds **1** and **2** is that there are substituents in the aliphatic skeleton, compound **2** has a carbonyl group (C=O) at C-9, which is characterized by the absence of a proton signal at C-9 and the appearance of a carbon signal at δc of 19.7 ppm. Based on the NMR data, it was suggested that compound **2** was *ar*-turmerone ($C_{15}H_{22}O$). Conformance may be seen when ^{1}H NMR, ^{13}C NMR, and HSQC data of compound 2 (**Table 2**) are compared to *ar*-turmerone data that have been published[14]

The bisabolene skeleton is a monocyclic sesquiterpene with a six-ring, which is formed from the cyclization of trans-farnesyl cations to form bisabolyl cations[18]. The bisabolyl cation is a precursor forming sesquiterpene compounds with a bisabolene skeleton. *ar*-curcumene compound formed from the dehydrogenation reaction of bisabolene, then *ar*-curcumene undergoes oxidative reactions to form *ar*-turmerone[17] .

Antimicrobial Activity

The antimicrobial activity of extracts, fractions, and compounds isolated from *C. soloensis rhizomes* showed diverse activity. The data showed that acetone extract, *n*-hexane fraction, and isolated compounds have the potential to be developed as antimicrobial agents. The minimum inhibitory concentration (MIC) of a test sample indicates its capacity to inhibit bacteria; the lower the MIC value, the higher the antimicrobial activity. Until recently, it has not been known with certainty what the standard value for the level of activity of antimicrobial compounds. According to some studies, a compound has a very high potential for usage as an antimicrobial if its MIC value is between 0.02-10.0 μg/mL [19], On the other hand, a compound can be considered antimicrobial if its MIC value is between 100-1000 μg/mL [20].

Table 2. Spectral data ^{1}H, ^{13}C-NMR and HSQC of *ar*-turmerone

C atomic position	δH (*much*. *J* Hz) ppm		δC ppm	
	2	**2***	**2**	**2***
1	7.07 (1H, *d*, 1.3)	6.98 (1H, *s*)	124.1	126.6
2	7.13 (1H, *d*, 7.8)	6.98 (1H, s)	126.2	129.1
3	-	-	136.2	135.5
4	7.13 (1H, *d*, 7.8)	6.98 (1H, *s*)	126.2	129.1
5	7.07 (1H, *dd*, 1.3; 7.8)	6.98 (1H, *s*)	124.1	126.6
6	-	-	143.7	143.7
7	3.26 (1H, *sext*, 7.2)	3.20 (1H, *m*)	35.4	35.3
8	2.62-2.87 (2H, *m*)	2.52 (2H, *m*)	52.1	52.7
9	-	-	199.7	199.8
10	5.81 (1H, s)	5.88 (1H, s)	126.1	124.1
11	-	-	155.7	155.1
12	2.12 (3H, *s*)	2.06 (3H, *s*)	21.4	20.6
13	1.88 (3H, *s*)	1.80 (3H, *s*)	27.4	27.6
14	1.31 (3H, *D*, 7.2)	1.18 (3H, *s*)	22.0	22.0
15	2.33 (3H, *s*)	2.25 (3H, *s*)	21.1	20.9

2: isolated compound, in $CDCl_3$ 500 MHz (^{1}H) and 125 MHz (^{13}C);
2*: [14]

Table 3. Antimicrobial activities of extract, fraction, and isolated compounds

Sample	MIC (µg/mL)			
	E. coli	***S. aureus***	***C. albicans***	***M. furfur***
Acetone extract	125	125	125	125
n-Hexane fraction	250	250	62.5	62.5
ar-Curcumene	62.5	62.5	62.5	62.5
ar-Turmerone	62.5	15.6	31.2	31.2
Chloramphenicol	3.9	1.9	-	-
Miconazole	-	-	6.25	6.25

In general, the antimicrobial activity of isolated compounds (*ar*-curcumene and *ar*-turmerone) has higher activity than acetone extracts and *n*-hexane fractions. The highest activity was shown by *ar*-turmerone against *S. aureus* with a MIC value of 15.6 µg/mL, while *ar*-curcumene showed the same activity in all four test microbes with a MIC value of 62.5 µg/mL. Previous research has shown that *ar*-turmerone was inactive against *S. aureus*[21]. This is likely due to differences in the assay methods used, the environment where bacteria grow, and the type of bacterial culture used. In addition, *ar*-Curcumene isolated from the fresh aerial parts of *Senecio selloi* was found to be effective against some microbial such as *S. aureus*, *Bacillus subtilis*, *E. coli*, *Pseudomonas aeruginosa, C. albicans, C. glabrata,* and *Saccharomyces cerevisiae* [22].

The varying antimicrobial properties for each compound in a particular microbe are not only determined by the type and characteristic of the microbe but also by the physical and chemical properties as well as the structure and functional groups of the compound [23]. The study of the structure and activity of isolated compounds in this study was stated qualitatively and focused on the activity level of isolated compounds for the same microbes.

ar-Curcumene and *ar*-turmerone compounds are sesquiterpene compounds with a monocyclic bisabolene skeleton that has a benzene ring. The difference between the two compounds is that in *ar*-turmerone there is a ketone group at the acyclic chain (C-9). The difference in the antifungal activity of the two compounds above against the four test microbes might be caused by the presence of ketone groups in C-9 of *ar*-turmerone. The antimicrobial activity *of ar*-turmerone against all four test microbes showed higher MIC values (MIC 15.6 - 62.5 µg/mL) than *ar*-curcumene (MIC 62.5 µg/mL). This suggests that the presence of carbonyl groups in *ar*-turmerone was thought to increase antimicrobial activity.

The mechanism of action of terpenoid compounds in inhibiting the growth or killing of microbes has not been widely reported. Some studies reported that terpenoid compounds work by disrupting the function of cell membranes. The lipophilicity of terpenoid compounds can enhance the permeability and fluidity of cell membranes, obstruct ion transport and respiration, and prevent membrane proteins from binding [24].

4. CONCLUSIONS

The isolation of bioactive compounds from the *n*-hexane fraction of *C. soloensis* rhizomes obtained two pure sesquiterpene compounds. Based on ^{1}H NMR, ^{13}C NMR, and HSQC spectroscopy analysis, the isolated compounds were identified as sesquiterpene compounds with bisabolene molecular skeletons, namely *ar*-curcumene ($C_{15}H_{22}$) and *ar*-turmerone ($C_{15}H_{22}O$).

The antimicrobial activity of acetone extract, *n*-hexane fraction, and isolated compounds of *ar*-curcumene and *ar*-turmerone showed diverse activity. The data showed that *ar*-turmerone had the highest activity against *S. aureus* with a MIC value of 15.6 µg/mL, while *ar*-curcumene showed the same activity against all test microbes with a MIC value of 62.5 µg/mL.

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REFERENCES

1. Keita K, Darkoh C, Okafor F. Secondary plant metabolites as potent drug candidates against antimicrobial-resistant pathogens. *SN Appl Sci*. 2022;4(8). doi:10.1007/s42452-022-05084-y
2. Elmaidomy AH, Shady NH, Abdeljawad KM, et al. Antimicrobial potentials of natural products against multidrug resistance pathogens: a comprehensive review. *RSC Adv*. 2022;12(45):29078-29102. doi:10.1039/d2ra04884a

3. Subositi D, Wahyono S. Study of the genus Curcuma in Indonesia used as traditional herbal medicines. *Biodiversitas*. 2019;20(5):1356-1361. doi:10.13057/biodiv/d200527
4. Dosoky NS, Setzer WN. Chemical composition and biological activities of essential oils of Curcuma species. *Nutrients*. 2018;10(9):10-17. doi:10.3390/nu10091196
5. Rahaman MM, Rakib A, Mitra S, et al. The genus Curcuma and inflammation: Overview of the pharmacological perspectives. *Plants*. 2021;10(1):1-19. doi:10.3390/plants10010063
6. Hong SL, Lee GS, Syed Abdul Rahman SN, et al. Essential oil content of the rhizome of *Curcuma purpurascens* Bl. (Temu Tis) and its antiproliferative effect on selected human carcinoma cell lines. *Sci World J*. 2014;2014. doi:10.1155/2014/397430
7. Rouhollahi E, Zorofchian Moghadamtousi S, Hamdi OAA, et al. Evaluation of acute toxicity and gastroprotective activity of *Curcuma purpurascens* BI. rhizome against ethanol-induced gastric mucosal injury in rats. *BMC Complement Altern Med*. 2014;14(1):1-10. doi:10.1186/1472-6882-14-378
8. Rouhollahi E, Moghadamtousi SZ, Al-Henhena N, et al. The chemopreventive potential of *Curcuma purpurascens* rhizome in reducing azoxymethane-induced aberrant crypt foci in rats. *Drug Des Devel Ther*. 2015;9:3911-3922. doi:10.2147/DDDT.S84560
9. Rouhollahi E, Zorofchian Moghadamtousi S, Paydar M, et al. Inhibitory effect of *Curcuma purpurascens* BI. rhizome on HT-29 colon cancer cells through mitochondrial-dependent apoptosis pathway. *BMC Complement Altern Med*. 2015;15(1):1-12. doi:10.1186/s12906-015-0534-6
10. Diastuti H, Asnani A, Chasani M. Antifungal activity of *Curcuma xanthorrhiza* and *Curcuma soloensis* extracts and fractions. *IOP Conf Ser Mater Sci Eng*. 2019;509(1):9-14. doi:10.1088/1757-899X/509/1/012047
11. Murningsih T, Rezeki S, Chairul H, Priyono S. The chemical composition and anti-bacteria activity analysis of essential oil of "Temu glenyeh" (*Curcuma soloensis* Val. *War AKAB Indones*. 2000;12:37-45.
12. Okta OP, Laili RTN. Antimicrobial activity of temu blenyeh (*Curcuma purpurascens* Blume) ethanol extract against on *Streptococcus mutants* and *Candida albicans*. *Ad-Dawaa' J Pharm Sci*. 2023;6(1):93-101. doi:10.24252/djps.v6i1.37703
13. Vitasari RA, Wibowo FR, Marliyana SD, Wartono MW. Isolation and identification of curcumin and bisacurone from rhizome extract of temu glenyeh (*Curcuma soloensis*. Val). *IOP Conf Ser Mater Sci Eng*. 2016;107(1). doi:10.1088/1757-899X/107/1/012063
14. Marliyana SD, Wartono MW, Wibowo FR, Munasah G. Isolasi dan Identifikasi Senyawa Seskuiterpen dari *Curcuma soloensis* Val. (Temu Glenyeh). *J Kim Val*. 2018;4(2):137-142. doi:10.15408/jkv.v4i2.7443
15. Ragasa CY, Rideout JA, Avenue T, Sciences B. Sesquiterpenoids and diarylheptanoids from *Curcuma domestica*. 2005;18:21-24.
16. Diastuti H, Syah YM, Juliawaty LD, Singgih M. Antibacterial *Curcuma xanthorrhiza* extract and fractions. *J Math Fundam Sci*. 2014;46(3):224-234. doi:10.5614/j.math.fund.sci.2014.46.3.2
17. Du ZT, Zheng S, Chen G, Lv D. A short synthesis of bisabolane sesquiterpenes. *Molecules*. 2011;16(9):8053-8061. doi:10.3390/molecules16098053
18. Bergman ME, Davis B, Phillips MA. Occurrence, and mechanism of action. *Molecules*. Published online 2019:3961.
19. Saleem M, Nazir M, Ali MS, et al. Antimicrobial natural products: An update on future antibiotic drug candidates. *Nat Prod Rep*. 2010;27(2):238-254. doi:10.1039/b916096e
20. Abreu AC, McBain AJ, Simões M. Plants as sources of new antimicrobials and resistance-modifying agents. *Nat Prod Rep*. 2012;29(9):1007-1021. doi:10.1039/c2np20035j
21. Marliyana SD, Wibowo FR, Wartono MW, Munasah G. Evaluation of antibacterial activity of sesquiterpene ar-turmerone from *Curcuma soloensis* Val. rhizomes. *IOP Conf Ser Mater Sci Eng*. 2019;578(1):0-3. doi:10.1088/1757-899X/578/1/012060
22. Narjara Santos da Silva G, Pozzatti P, Rigatti F, et al. Antimicrobial evaluation of sesquiterpene ar-curcumene and its synergism with imipenem. *J Microbiol Biotechnol food Sci*. 2015;4(5):434-436. doi:10.15414/jmbfs.2015.4.5.434-436
23. Lobiuc A, Pavăl NE, Mangalagiu II, et al. Future Antimicrobials: Natural and functionalized phenolics. *Molecules*. 2023;28(3). doi:10.3390/molecules28031114
24. Huang W, Wang Y, Tian W, et al. Biosynthesis investigations of terpenoid, alkaloid, and flavonoid antimicrobial agents derived from medicinal plants. *Antibiotics*. 2022;11(10). doi:10.3390/antibiotics11101380