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Chemical Profiling and Antibacterial Activity of Javanese Turmeric (*Curcuma xanthorriza*) Essential Oil on Selected Wound Pathogen

Suryati[1,2*], Dachriyanus[2], Irwandi Jaswir[1], Faridah Yusof[3]

[1] *International Institute of Halal Research and Training, IIUM Malaysia*
[2] *Faculty of Pharmacy, Andalas University, Padang, West Sumatera, Indonesia*
[3]*Departement of Biotechnology Engineering, Faculty of Engineering, IIUM Malaysia*
[*]*Corresponding author. Email:suryatifudin@gmail.com*

ABSTRACT
Essential oil is a volatile liquid with a pungent taste and aroma. The essential oil has been used since many years ago for many purpose including as antimicrobial agent. Javanese turmeric reported containing essential oil. Therefore, the purpose of this study was to identify chemical profiling of essential oil of Javanese turmeric (*Curcuma xanthorriza*) rhizome which collected from Padang, West Sumatera as well as to evaluate antibacterial activity toward selected wound infected bacteria. The hydrodistillation technique was used for extracting the essential oil and the chemical profiling was identified by using gas chromatography-mass spectrometry (GC-MS). The percentage of essential oil obtained was 0.557 %. The result revealed the essential oil of rhizome contained α-Curcumene, β-Curcumene, Di-epi-.α.-cedrene-(I), (-)-Xanthorrhizol, and S-(-)-Camphor as major constituents. Antibacterial activity was performed by broth microdilution technique The essential oil was active against test bacteria with the Minimum Inhibition Concentration (MIC) value was 5 mg/ml, 6.25 mg/ml and 10 mg/ml for *Staphylococcus epidermidis*, *Staphylococcus aureus* and *Enterococcus faecalis* respectively.

Keywords: *Zingiberaceae, essential oils, Curcuma xanthorriza, GC-MS*

1. INTRODUCTION

There was reported more than a million people suffered from burn injuries annually [1]. This injury may be infected by microorganism if the wound was not getting well treatment and may developed to the chronic wound. The essential oil promising to be an alternative source for inhibiting this microbial infection. Essential oil has gained a lot of people attentions for treatment of disease due to it is affordable and safer than synthetic medication [2].

Essential oils are secondary metabolites which composed of 20 – 60 components which are terpenes as major constituents and non-terpene constituents. It is volatile and has pungent odor. Essential oils can be extracted from any parts of plant such as flowers, roots, bark, leaves, seeds, peel, fruits, wood, and whole plant. The essential oils have been traditionally used for many purposes including in cosmetic and aromatherapy, as food preservative and flavoring agents as well as for prevention and treatment of diseases. Moreover, essential oil possesses many interesting pharmacological effect such as antiseptic, antibacterial, antiviral, antioxidant, anti-parasitic, antifungal, and insecticidal activities [2,3].

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Curcuma xanthorriza which commonly named as Javanese turmeric and locally called as Temu Lawak belongs to Zingiberaceae family. It was originated from Indonesia – Malaysia and nowadays extensively cultivated in South East Asia [4] According to Ministry of Agriculture, Indonesia, Javanese turmeric is one of 66 of Indonesian medicinal herb commodities and has been used as one of ingredients in Indonesian traditional drink which called as "Jamu"[5]. It contained about 4-6% of essential oil. It was traditionally used to overcome lack of appetite, constipation, hemorrhoid, acne, diarrhea, and seizure medication, to destroy gallstones, to treat kidney and liver diseases, rheumatic pain, rheumatism, and arthritis, and to treat thrush and vaginal discharge. Many pharmacological activities reported that it acted as antimicrobial, anti-inflammatory, antitumor, antioxidant, hepatoprotective and etc [4].

Although many articles reported on Javanese turmeric composition and antibacterial activity, but there was no article reported about this plant which grown in the West Sumatera. Many factors would affect the chemical composition of essential oil which were method of rhizome preparation, geographical condition and freshness of rhizome. Thus this study was aimed to identify the chemical composition of the essential oil of *Curcuma xanthorriza* rhizome collected from Padang, West Sumatera.as well as to evaluate antimicrobial activity towards selected wound pathogen.

2. METHODS

2.1 Chemical and Test Microorganism

Analytical grade chemicals and solvents were used throughout this study. Selected wound infected bacteria were chosen for the test microorganisms. They were *Staphylococcus epidermidis*, *Enterococcus faecalis* and *Staphylococcus aureus* which obtained from Indonesian Food and Drug Authority. As stock cultures, all test bacteria were kept at 37°C on nutrient agar slants.

2.2 Plant Collection

Fresh Javanese turmeric rhizome was obtained from a local market in Padang, West Sumatera, Indonesia. This plant was identified by a botanist, Dr. Norainas. The specimen was then kept in the ANDA herbarium at Andalas University's Faculty of Mathematics and Natural Sciences in Padang, Indonesia.

2.3 Extraction of Essential Oil

Unpeeled rhizomes were sorted and washed under running tapped water prior to distillation. It was then cut into pieces. 1 kg of fresh rhizome was transferred into a Clevenger distillation apparatus then further proceeded for hydrodistillation for a few hours. The oil was collected in a dark bottle and stored at 40 degrees Celsius until further use.

2.4 Analysis of Essential Oil

Agilent Technology 7890A / 5975C inert MSD with Triple-Axis Detector was used to analyze the essential oil. For this analysis, a capillary column HP-5MS 5 percent Phenyl Methyl Silox (30 m x 250 m x 0.25 m) was used. The oven temperature was set to 80°C for 1 minute before gradually increasing to 110°C at a rate of 2°C/min, 140°C at a rate of 3°C/min, 170°C at a rate of 4°C/min, and finally 180°C at a rate of 5°C/min.

Analysis of Essential oil was performed by GC/MS Agilent Technology 7890A / 5975C inert MSD with Triple-Axis Detector as seen in **Table 1.**

Table 1. The GC-MS condition

Parameter	Condition
GC Condition **Inlet** Heater	: 250°C
Pressure	: 11.7 psi
Split ratio	: 200 : 1
Split flow	: 240 mL/min
Columns Flow rate	: 1.2 mL/min
Pressure	: 11.7 psi
Oven	Initial temperature : 80^0C
	Initial time : 1 minute
Temperature program	80-110°C at a rate 2°C/min, 110-140°C at a rate 3°C/min, 140-170°C at a rate 4°C/min, 170-200°C at a rate 5°C/min
MS Condition	MS Source : 230°C MS Quad : 150°C Tune type : EI

2.5 Antibacterial Activity

Antibacterial activity was performed in 96-well plate. First, the bacteria culture was suspended in sterile saline then the turbidity was compared to the standard 0.5 McFarland. Then the bacterial suspension was diluted in MHB medium in a ratio of 1:150. Fifty microliters of MHB media was transferred into all well except well in the row A followed by 50 µl test sample into row A and B then followed by two-fold dilution by transferring 50 µl of well B into C. The step was repeated until row G and then 50 µl of well G discarded. So that, the sample concentrations were obtained as follows, 10%, 5%, 2.5%, 1.25%, 0.63% and 0.31%. Finally, 50 µl of bacterial suspension was added in each row A to H except for grow and sterility control and 50 µl of ciprofloxacin solution as positive control was transferred into row G. Subsequently the plate was incubated for 18-24 hours at 37 ° C. The minimum of inhibition concentration (MIC) value was determined by observing the color of solution. The purple color indicated the viable bacteria, otherwise colorless solution indicated no viable bacteria [6]

3. RESULT AND DISCUSSION

3.1 Chemical Composition of Essential Oil

Fresh rhizome of *Curcuma xanthorriza* yielded 0.557 % of essential oil. The essential oil of Javavense turmeric rhizome was composed of monoterpenes and sesquiterpenes with percentage of 63.41% and 36.59% respectively as described in **Table 2**. Sesquiterpenes hydrocarbon was found as major component with the total percentage of 46.34%. Moreover, oxygenated monoterpenes identified in the percentage of 21.95%. Chemical analysis revealed 6 major compounds which counted 64.54% of total identified compound as described in **Table 3**. α-curcumene was identified as major compound with the percentage of 22.007% followed by β-curcumene 13.271%. The chemical profiling of all component identified in the essential oil as seen in the **Figure 1.**

Table 2. Main classes and subclasses of *Curcuma xanthorriza*

Class, Subclass group of compound		Percentage (%)
Monoterpenoids		
	Monoterpene hydrocarbons	12.20
	Oxygenated monoterpenes	21.95
Sesquiterpenoids		
	Sesquiterpene hydrocarbons	46.34
	Oxygenated Sesquiterpenes	17.07
Others		2.44
Total identified		100

Table 3: Chemical Composition of the essential oil of fresh rhizome Curcuma xanthorriza from West Sumatera

Compound	Molecular Weight	% Composition
α-curcumene	202.172	22.007
β-curcumene	204.188	13.271
Di-epi-α-cedrene-(I)	204.188	8.913
(-)-Xanthorrhizol	218.167	8.445
S-(-)-Camphor	152.12	6.807
trans-α-Bergamotene	204.188	5.098

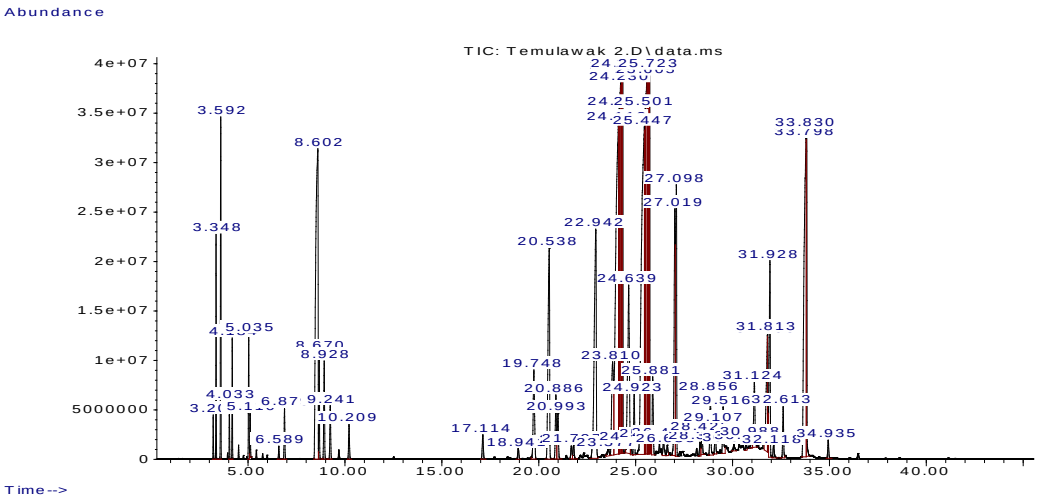

Figure 1. Chromatogram of essential oil of Javanese turmeric rhizome

This finding showed there was the different major component identified in our rhizomes compared to the another studies. *C. xanthorriza* was collected from Chiang Mai, Thailand revealed monoterpene (88.53%) was predominantly in the rhizome of *Curcuma xanthorriza*.. α-terpinolene (24.86%), p-cymen-7-ol (12.17%) was as major constituents [7]. Otherwise Javanese turmeric collected from Nongkai province, Thailand revealed 1,8 cineol (37.58%) and curzerenone (13.70%) as major component [8]. Another study conducted on Javanese turmeric dry rhizomes collected from Pasingangan, Banyumas, Central Java showed cineole, camphor, α-curcumin, androsta, dan α-chamigren as the major component [9].

3.2 Antibacterial Activity

Antibacterial activity of the essential oil was performed by broth microdilution technique toward selected wound pathogen. The result was as seen in **Table 4**. The essential oil inhibited all test microorganisms. The lowest MIC values was showed on *Staphylococcus epidermidis* with value of 5 mg/ml. However, the MIC value for *Enterococcus faecalis* and *S. aureus* were 10 mg/ml and 6.25 mg/ml respectively.

Table 4. Antibacterial activity of the essential oil of Javanese turmeric

Sample Concentration (mg/ml)	Colour observation		
	Staphylococcus epidermidis	*Enterococcus faecalis*	*Staphylococcus aureus*
10	Colorless*	Colorless	Colorless
5	colorless	purple	Purple
2.5	purple	purple	Purple
1.25	purple	purple	Purple
0.63	purple	purple	Purple
0.31	purple	purple	Purple
MIC value	5 mg/ml	10 mg/ml	6.25g/ml

*colorless indicated no bacteria growth

purple color indicated bacteria growth

The activities of essential oils were affected by the composition, functional groups present in active components, and synergistic interactions [10]. The Javanese turmeric essential oil contained phenolic compound particularly xanthorrhizol which may responsible for its antibacterial activity. In addition, the presence of oxygenated compound S-(-)-Camphor may increase its antibacterial activity [11]. All test microorganisms belong to Gram positive bacteria. The Gram positive bacteria surrounded by thick peptidoglycan and the presence of the lipophilic ends of lipoteichoic acid present in cell membrane would facilitate the infiltration of hydrophobic component in essential oil. Therefore, these essential oil acted through disruption of cell walls or membranes of microbes by altering its cell permeability, resulting in the forfeit of essential molecules such as ATP, RNA, protein, and DNA

4. CONCLUSION

The main chemical component found in the essential oil of Javanese turmeric rhizome was α-curcumene. Javanese turmeric essential oil inhibited selected wound pathogen. The geographical condition would affect the chemical composition and the bioactivities of the essential oil.

AUTHORS' CONTRIBUTIONS

Suryati performed sample collection, extraction, data analysis and wrote the manuscript. Dachriyanus, Irwandi Jaswir and Faridah Yusof gave financial support and supervised the project. All authors have checked and agreed the manuscript to publish.

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REFERENCES

[1]. Selvakumar PM, Rajkumar S RJ, MSA MN. Phytochemicals as a potential source for anti-microbial, anti-oxidant and wound healing - a review. MOJ Bioorganic Org Chem. 2018;2(2):61–70. https://doi.org/10.15406/mojboc.2018.02.0058

[2]. A. Shaaban H. Essential Oils as Antimicrobial Agents: Efficacy, Stability, and Safety Issues for Food Application. In: IntechIvanova, Nadezhda Gugleva, Viliana. 2016.

[3]. Hyldgaard M, Mygind T, Meyer RL. Essential oils in food preservation: Mode of action, synergies, and interactions with food matrix components. Front Microbiol. 2012;3(JAN):1–24. https://doi.org/10.3389/fmicb.2012.00012

[4]. Rahmat E, Lee J, Kang Y. Javanese Turmeric (*Curcuma xanthorrhiza* Roxb):Phytochemistry , Biotechnology , and Pharmacological Activities. Hindawi Evidence-Based Complement Altern Med. 2021;15. Available from: https://doi.org/10.1155/2021/9960813

[5]. Salim Z, Munadi E. Info Komoditi Tanaman Obat. Badan Pengkajian dan Pengembangan Perdangan Kementrian Perdagangan Republik Indonesia. 2017. https://doi.org/10.7748/ldp.5.4.28.s16

[6]. Balouiri M, Sadiki M, Ibnsouda SK. Methods for in vitro evaluating antimicrobial activity: A review. J Pharm Anal. 2016;6(2):71–9. https://doi.org/10.1016/j.jpha.2015.11.005

[7]. Akarchariya N, Sirilun S, Julsrigival J, Chansakaowa S. Chemical profiling and antimicrobial activity of essential oil from *Curcuma aeruginosa* Roxb., *Curcuma glans* K. Larsen & J. Mood and *Curcuma cf. xanthorrhiza* Roxb. collected in Thailand. Vol. 7, Asian Pacific Journal of Tropical Biomedicine. 2017. p. 881–5. https://doi.org/10.1016/j.apjtb.2017.09.009

[8]. Jarikasem S, Thubthimthed S, Chawananoraseth K, Suntorntanasat T, Brophy JJ. Essential oils from three Curcuma species collected in Thailand. Acta Hortic. 2005;675:37–40. https://doi.org/10.17660/ActaHortic.2005.675.4

[9]. Meilaningrum DN, Tjiptasurasa, Rahayu WS. Minyak Atsiri, Perbandingan Kadarnya Pada Rimpang Temulawak (*Curcuma xanthorrhiz*a Roxb.) yang Dikeringkan dengan Metode Sinar Matahari dan Oven beserta Profil Kromatografi Gas Spektrometri Massa (KGSM). Pharmacy. 2009;6(3):115–25.

[10]. Chouhan S, Sharma K, Guleria S. Antimicrobial Activity of Some Essential Oils—Present Status and Future Perspectives. Medicines. 2017;4(3):58. https://doi.org/10.3390/medicines4030058

[11].Baharudin MKA, Hamid SA, Susanti D. Chemical composition and antibacterial activity of essential oils from three aromatic plants of the zingiberaceae family in Malaysia. J Phys Sci. 2015;26(1):71–81.