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Antifungal activity of curcuma xanthorrhiza and curcuma soloensis extracts and fractions

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Abstract. In this research, the antifungal activity of acetone extracts, and fractions of *n*-hexane, chloroform and ethylacetate of *C. xanthorrhiza* and *C. soloensis* rhizomes have been conducted. The antifungal activity was carried out by using agar dilution method and evaluated against *Aspergillus fumigatus*, *Candida albicans*, *Epidermophyton sp*, *Penicillium sp* and *Trichophyton rubrum*. The result showed that acetone extract and chloroform fraction of *C. xanthorrhiza* exhibited significant activities against *A. fumigatus*, *Epidermophyton sp*, *Penicillium sp* and *T. rubrum* with MIC 12.5-25.0 µg/mL. The *n*-hexane fraction of *C. xanthorrhiza* showed significant activity on *Epidermophyton sp* with MIC 12.5 µg/mL. Meanwhile, the extract and fraction of *C. soloensis* showed moderate and weak activities against all tested fungal with MIC 50-200 µg/mL.

Keywords: *C. xanthorrhiza*, *C. soloensis*, extract and fraction, antifungal.

1. Introduction

Fungus is one of the microbes that cause infection, especially in tropical countries. Tropical climate with high air humidity as in Indonesia was strongly supports the growth of fungus. The proliferation of fungus infections is also supported by low public awareness of environmental hygiene, sanitation, and healthy lifestyles. One attempt to suppress the spread of fungal infections is through the use of antibiotics or synthetic food preservatives. But within a certain time, the ability of antibiotics is gradually decreased, because the targeted microbes were developing its immunity. Development of microbial resistance has stimulated researchers to find new antibiotics either by synthesis or from natural compounds, of particularly from plants [1].

Curcuma is an important medicinal plant in Indonesia, because more than 50 recipes of herbs circulating in Indonesia using *Curcuma* rhizome. These herbs are used to treat various diseases, including gastrointestinal and liver disorders, kidney inflammation, gall stones, hemorrhoids, rheumatism, high cholesterol, menstruation, lack of breast milk and appetite [2, 3]. In addition, *Curcuma* rhizome is also widely used as a spice on a variety of cuisine, giving the yellow color on food, to keep the body fresh, and for cosmetic raw materials [3].

Previous study showed that *C. xanthorrhiza* rhizome extracts can lower cholesterol levels in patient with high cholesterol. *C. xanthorrhiza* is also known as hepatoprotector, the regular consumption of boiling of three slices of rhizome *C. xanthorrhiza* and one piece of papaya leaf can decrease serum glutamic pyruvic transaminase (SGPT) and SGOT (serum glutamic oxaloacetic transaminase) of hepatitis patients to normal, for a week [4]. Extract of *C. xanthorrhiza* rhizome has been scientifically proven to have hypothermic effects [5], analgesic and antidiuretic activity [6, 7], immunostimulant [8],



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anticarcinogenic [9] and antibacterial [10] Kertia *et al.* [11] reported that *C. xanthorrhiza* is commonly used to prevent arthritis (osteoarthritis) due to its anti-inflammatory effect [12]. The results of the research on four species of *Curcuma* namely *C. longa*, *C. caesia*, *C. amada*, and *C. aromatica* also known that water extract and alcohol extract *C. longa* and *C. aromatica* showed antimicrobial activity after tested against *Staphylococcus aureus*, *Bacillus subtilis*, *C. albicans* and *Aspergillus flavus* [13]. The essential oils of *C. soloensis* was reported to inhibit the growth of *Staphylococcus aureus*, *S. epidermis*, and *Streptococcus haemolyticus* [14]. The extracts of *n*-hexane, methylene chloride and ethylacetate of *C. soloensis* rhizome were also showed the antifungal activity against *C. albicans* [15].

The phytochemical studies of the *Curcuma* rhizomes indicate that they contain two mayor types of secondary metabolites, namely diarilheptanoid (curcuminoid) and terpenoid mainly sesquiterpenes [16]. Curcumin is the most widely studied diarylheptanoid compound, including hepatoprotector, antioxidant, antitumor, anticancer, anti-inflammatory, anti-HIV and antimicrobial [12, 16]. Santorizol as one of the main terpenoid compounds in the *Curcuma* rhizome is known to have high activity against some pathogenic bacteria [17-19] some *Candida* [20], *Malassezia* [21] and filamentous fungi [22]. Germacrone and furanodienon was known to also have antibacterial activity [23, 24].

Some of the results of this study showed the potential of *Curcuma* rhizome as antimicrobial, but has not been studied comprehensively, especially its activity as an antifungal. In this study will be conducted the antifungal properties of extract and fraction of *C. xanthorrhiza* and *C. soloensis* rhizomes against *A. fumigatus*, *C. albicans*, *Epidermophyton sp*, *Penicillium sp.*, and *T. rubrum*.

2. Material and Methods

2.1. Materials

We used rhizome of *C. xanthorrhiza* and *C. soloensis* (collected from Solo, Indonesia), redestilled solvents of *n*-hexane, ethylacetate and methanol, chloroform (Merck), demineralized water, fungal strains: *A. fumigatus*, *C. albicans*, *Epidermophyton sp.*, *Penicillium sp.*, and *Trichophyton rubrum*, Sabaroud Dextrose Agar (Oxoid), Ketoconazole (Merck) and dimethyl sulfoxide (Merck).

2.2. Extraction and fractionation of *C. xanthorrhiza* and *C. soloensis* rhizomes

The fresh rhizome of *C. xanthorrhiza* (10 kg) and *C. soloensis* (10 kg) were washed with water to remove the impurities, then cut to small pieces and air dried for 5 days. The dry rhizomes were ground into powder. A dry powder of *C. Xanthorrhiza* (1.2 kg) and *C. soloensis* rhizomes (1.0 kg) were extracted with acetone (three times) for three days, at room temperature. The each of acetone extract was filtered and concentrated using a rotary evaporator. Furthermore, the acetone extract was partitioned into *n*-hexane: methanol (1:1). Then *n*-hexane soluble extract (*n*-hexane fraction) was concentrated with a rotary evaporator. In other hand, the methanol soluble extract was partitioned into chloroform: water (1:1). The chloroform soluble extract (chloroform fraction) was concentrated with a rotary evaporator, then the water soluble was extracted into ethylacetate to give ethylacetate fraction.

2.3. Antifungal activity assays [25]

In vitro antifungal activity assays was carried out with agar dilution methods against five fungal i.e. *A. fumigatus*, *C. albicans*, *Epidermophyton sp*, *Penicillium sp*, and *Trichophyton rubrum*. The concentration of sample was 400, 200, 100, 50, 25, 12.5, 6.25 µg / mL. The sample was dissolved in 10% DMSO in distilled water.

Selected fungal were cultured for 48 hours at 27 °C under aerobic conditions on agar media (Sabaroud Dextrose Agar). Afterwards, the fungal were suspended in a 0.9% NaCl solution (w/v). The concentration of fungal suspension was adjusted to 10⁷ fungal cells /mL.

A total of 1.0 ml of each test solution was put in a test tube, then each 3.0 mL of agar (SDA) was added which was still liquid. Place each tube in a tilted position and allow it to stand until the sample-media solution solidifies. The fungal suspension (10 µL) was then inoculated to the agar medium surface which containing the sample solution, then they were incubated for 48 hours at 27°C. Furthermore, the fungal growth was observed. The lowest concentration of solution where no fungal growth was stated

as the minimum inhibitory concentration (MIC). The same method was carried out to the negative control (without extract or fraction) and standard antibiotics ketoconazole (positive control). The assay was repeated three times.

3. Result and Discussions

Extraction of the dried powder of *C. xanthorrhiza* (1.2 kg) and *C. soloensis* (1.0 kg) rhizomes with acetone both yielded the brownish yellow paste 87.7 g and 58 g respectively. The liquid-liquid fractionation of acetone extract of *C. xanthorrhiza* with *n*-hexane, chloroform and ethyl acetate respectively, was yielded *n*-hexane fraction 55.6 g, chloroform fraction 10.5 g and ethyl acetate fraction 3.7 g, while the liquid-liquid fractionation of acetone extract of *C. soloensis* into *n*-hexane, chloroform and ethyl acetate was obtained *n*-hexane fraction 38.2 g, chloroform fraction 6.5 g and ethyl acetate fraction 2.1 g. The weight of *n*-hexane fractions of *C. xanthorrhiza* and *C. soloensis* were most than chloroform and ethyl acetate fractions, these showed that practically most of the mass acetone extract soluble in *n*-hexane fractions. The previous study reported that *n*-hexane fraction of *Curcuma* contain the essential oils with the major constituents was sesquiterpene, while chloroform fractions was contain curcuminoids as its main component [19].

The antifungal activities of the acetone extract, *n*-hexane, chloroform and ethylacetate fractions of *C. xanthorrhiza* and *C. soloensis* was presented in Table 1 as minimum inhibitory concentration (MIC) values.

Table 1. MIC values of *C. xanthorrhiza* and *C. soloensis* extracts and fractions

Fungal	MIC ($\mu\text{g/mL}$)								
	Kzl	Cx-A	Cx-H	Cx-C	Cx-E	Cs-A	Cs-H	Cs-C	Cs-E
<i>Fumigatus</i>	6.25	50	25	25	100	100	50	50	200
<i>Albicans</i>	12.5	50	50	25	50	100	50	50	100
<i>Epidermophyton sp</i>	6.25	12.5	12.5	12.5	50	200	50	50	50
<i>Penicillium sp</i>	6.25	25	100	12.5	100	200	200	100	200
<i>T. rubrum</i>	6.25	12.5	50	25	100	100	50	50	100

Kzl=ketoconazole; Cx= *C. xanthorrhiza*; Cs=*C. soloensis*; A=acetone extract; H=*n*-hexane fraction; C= chloroform fraction; E= ethylacetate fraction.

As shown in Table 1, the extract and fractions of *C. xanthorrhiza* and *C. soloensis* were potential as antifungal agent due to both have the MIC values $<1000 \mu\text{g/mL}$ [25]. The highest antifungal activity (MIC $12.5 \mu\text{g/mL}$) was showed by acetone extract of *C. xanthorrhiza* against *Epidermophyton sp* and *T. rubrum*, *n*-hexane fraction of *C. xanthorrhiza* against *Epidermophyton sp*, and chloroform fraction of *C. xanthorrhiza* against *Epidermophyton sp* and *Penicillium sp*. While the ethylacetate fraction of *C. xanthorrhiza* showed moderate and weak activities with MIC values $50-100 \mu\text{g/mL}$. The extract and fraction of *C. soloensis* also showed moderate and weak antifungal activity with MIC values $50-200 \mu\text{g/mL}$. The acetone extract of *C. soloensis* showed weak activity ($\geq 100 \mu\text{g/mL}$). against all the tested fungal with MIC $100-200 \mu\text{g/mL}$, the *n*-hexane and chloroform fractions of *C. soloensis* showed moderate activities ($< 100 \mu\text{g/mL}$) against *A. fumigatus*, *C. albican*, *Epidermophyton sp*, and *T. rubrum* with MIC values $50 \mu\text{g/mL}$.

The difference in antifungal activity level caused by each extract and fractions have different component. The *n*-hexane fraction of *C. xanthorrhiza* and *C. soloensis* were containing essential oils (type sesquiterpenoids and monoterpenes) as the main component, while the chloroform fraction was containing curcuminoids as major component [19]. Previous research reported that the essential oils and curcuminoids of *Curcuma* have biological activities, one of them as antimicrobial activity [6, 27].

The antifungal mechanism of terpenoids and curcuminoid has been reported. The study have shown that the site action of cyclic hydrocarbon, including terpenoids and curcuminoids was at cell membrane. Terpenoids was interfere permeability of cell membranes, which had a consequence a permeability increase and loss of cellular constituents. These causes inhibition of enzyme, which are crucial to the

energy system in a cell [28]. Meanwhile, curcuminoids was disturbs the membrane potential and disrupts membrane integrity. The previous study assumed that curcumin forms electrostatic and/or hydrophobic interaction with fungal cell membrane and cell wall causing membrane disruption [29].

4. Conclusions

The acetone extract and chloroform fraction of *C. xanthorrhiza* exhibited significant activities against *Epidermophyton sp.*, *Penicillium sp.* and *Trichophyton rubrum* with MIC (minimum inhibitory concentration) 12.5-25.0 µg/mL. The *n*-hexane fraction of *C. xanthorrhiza* showed significant activity on *Epidermophyton sp.* with MIC 12.5 µg/mL. Meanwhile, the extracts and fractions of *C. soloensis* showed moderate and weak activities against all tested fungal with MIC 50-200 µg/mL.

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References

- [1] Saleem M, Nazir M, Ali M S, Hussain H, Lee Y S, Riaz N and Jabbar A 2010 Antimicrobial natural products: an update on future antibiotic drug candidates *Nat. Prod. Rep.* **27** 2 238-54
- [2] Achmad S A, Hakim E, Makmur L, Syah Y, Juliawaty L and Mujahidin D 2009 *Ilmu Kimia dan Kegunaan Tumbuh-Tumbuhan Obat Indonesia* (Bandung: Institut Teknologi Bandung)
- [3] Tilaar M, Wih W and Ranti A 2010 The Green Science of Jamu: Pendekatan Pragmatik untuk Kecantikan dan Kesehatan. In: *Dian Rakyat*, (Jakarta)
- [4] Trubus R 2009 *Herbal Indonesia Berkhasiat: Bukti Ilmiah dan Cara Racik PT Trubus Swadaya, Depok*
- [5] Yamazaki M, Maebayashi Y, Iwase N and Kaneko Y 1988 Studies on Pharmacologically Active Principles from Indonesian Crude Drugs. II.: Hypothermic Principle from *Curcuma xanthorrhiza* ROXB *Chem. Pharm. Bull.* **36** 6 2075-8
- [6] Mahmood M H, Bachar S C, Islam M S and Ali M S 2004 Analgesic and diuretic activity of *Curcuma xanthorrhiza* *Dhaka University J. Pharm. Sci.* **3** 1
- [7] Devaraj S, Esfahani A S, Ismail S, Ramanathan S and Yam M F 2010 Evaluation of the antinociceptive activity and acute oral toxicity of standardized ethanolic extract of the rhizome of *Curcuma xanthorrhiza* Roxb *Molecules* **15** 4 2925-34
- [8] Kim A-J, Kim Y-O, Shim J-S and Hwang J-K 2007 Immunostimulating activity of crude polysaccharide extract isolated from *Curcuma xanthorrhiza* Roxb *Biosci. Biotechnol. Biochem.* **71** 6 1428-38
- [9] Park J H, Park K K, Kim M J, Hwang J K, Park S K and Chung W Y 2008 Cancer chemoprotective effects of *Curcuma xanthorrhiza* *Phytother. Res. Int. J. Devoted to Pharmacol. Toxicol. Eval. Nat. Prod. Deriv.* **22** 5 695-8
- [10] Mary H P, Susheela G K, Jayasree S, Nizzy A, Rajagopal B and Jeeva S 2012 Phytochemical characterization and antimicrobial activity of *Curcuma xanthorrhiza* Roxb *Asian Pac. J. Trop. Biomed.* **2** 2 S637-S40
- [11] Kertia N, Sudarsono, A. D I A D, Mufrod, Catur E, Rahardjo P and Asdie A H 2005 Pengaruh pemberian kombinasi minyak atsiri temulawak dan ekstrak kunyit dibandingkan dengan piroksikam terhadap angka leukosit cairan sendi penderita dengan osteoarthritis lutut *Majalah Farmasi Indonesia* **16** 3
- [12] Ozaki Y 1990 Antiinflammatory effect of *Curcuma xanthorrhiza* ROXB. and its active principles *Chem. Pharm. Bull.* **38** 4 1045-8
- [13] Harit J, Barapatre A, Prajapati M, Aadil K R and Senapati S 2013 Antimicrobial activity of rhizome of selected *Curcuma* variety *Int. J. Life Sci. Biotech. Pharma. Res.* **2** 3 183-9
- [14] Murningsih T, Rezeki S, H-Priyono S and Taufiq A 2000 The chemical composition and anti bacteria activity analysis of essential oil of " Temu glenyeh" (*Curcuma soloensis* Val.) *Warta AKAB (Indonesia)* **12** 37-45

- [15] Harliana D 2006 *Aktivitas Antijamur Ekstrak Rimpang Temu Glenyeh* Jurusan Kimia Universitas Sebelas Maret Surakarta
- [16] Ravindran P, Babu K N and Sivaraman K 2007 *Turmeric. The Genus Curcuma*: CRC Press)
- [17] Hwang J, Shim J and Pyun Y 2000 Antibacterial activity of xanthorrhizol from *Curcuma xanthorrhiza* against oral pathogens *Fitoterapia* **71** 3 321-3
- [18] Lee L Y, Shim J-S, Rukayadi Y and Hwang J-K 2008 Antibacterial activity of xanthorrhizol isolated from *Curcuma xanthorrhiza* Roxb. against foodborne pathogens *J. Food Prot.* **71** 9 1926-30
- [19] Diastuti H, Syah Y M, Juliawaty L D and Singgih M 2014 Antibacterial *Curcuma xanthorrhiza* extract and fractions *J. Math. Fundam. Sci.* **46** 3 224-34
- [20] Rukayadi Y, Yong D and Hwang J-K 2006 In vitro anticandidal activity of xanthorrhizol isolated from *Curcuma xanthorrhiza* Roxb *J. Antimicrob. Chemother.* **57** 6 1231-4
- [21] Rukayadi Y and Hwang J K 2007 In vitro anti-Malassezia activity of xanthorrhizol isolated from *Curcuma xanthorrhiza* Roxb *Lett. Appl. Microbiol.* **44** 2 126-30
- [22] Rukayadi Y and Hwang J K 2007 In vitro antimycotic activity of xanthorrhizol isolated from *Curcuma xanthorrhiza* Roxb. against opportunistic filamentous fungi *Phytotherapy Research: An International Journal Devoted to Pharmacological and Toxicological Evaluation of Natural Product Derivatives* **21** 5 434-8
- [23] Diastuti H, Syah Y M, Juliawaty L D and Singgih M 2013 Sesquiterpen Furanodienon dari Rimpang *Curcuma Xanthorrhiza* dan Aktivitas Antibakterinya *Molekul* **8** 2 101-10
- [24] Therese K, Bagyalakshmi R, Madhavan H and Deepa P 2006 In-vitro susceptibility testing by agar dilution method to determine the minimum inhibitory concentrations of amphotericin B, fluconazole and ketoconazole against ocular fungal isolates *Indian J. Med. Microbiol.* **24** 4 273
- [25] Abreu A C, McBain A J and Simoes M 2012 Plants as sources of new antimicrobials and resistance-modifying agents *Nat. Prod. Rep.* **29** 9 1007-21
- [26] Dosoky N and Setzer W 2018 Chemical Composition and Biological Activities of Essential Oils of *Curcuma* Species *Nutrients* **10** 9 1196
- [27] Zorofchian Moghadamtousi S, Abdul Kadir H, Hassandarvish P, Tajik H, Abubakar S and Zandi K 2014 A review on antibacterial, antiviral, and antifungal activity of curcumin *Biomed. Res. Int.* **2014**
- [28] Policegoudra R, Rehna K, Rao L J and Aradhya S 2010 Antimicrobial, antioxidant, cytotoxicity and platelet aggregation inhibitory activity of a novel molecule isolated and characterized from mango ginger (*Curcuma amada* Roxb.) rhizome *J. Biosci.* **35** 2 231-40
- [29] Kumar A, Dhamgaye S, Maurya I K, Singh A, Sharma M and Prasad R 2014 Curcumin targets cell wall integrity via calcineurin-mediated signaling in *Candida albicans* *Antimicrob. Agents Chemother.* **58** 1 167-75

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