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Antioxidant Activity of *Syzygium samarangense* L. and Their Endophytic Fungi

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ABSTRACT. Leaves of jambu air (*Syzygium samarangense* L.) has been used by local residents as medicine for various diseases caused by free radical agents in human's body. This study aims to find a new source of antioxidants from medicinal plants and their endophytic fungi *Syzygium samarangense* leaves was fractionated by maceration method using gradient solvent i.e. hexane, ethyl acetate, and methanol. The antioxidant activity of the leaf fractions was determined using 1,1-diphenyl-2-picryl hydrazyl (DPPH) method. The chemical compound was isolated from acetone fraction by chromatographic techniques and their chemical structure was identified using spectroscopy techniques. The endophytic fungi was isolated from *S. samarangense* leaves and continued to cultivation in potato-dextrose broth (PDB) medium for four weeks. The medium was extracted using ethyl acetate and determined its antioxidant activity. Fungi's isolate with highest activity was analyzed to identify its molecular. Ethyl acetate fraction of *S. samarangense* leaves showed highest antioxidant activity. Spectroscopy analysis result concluded the isolated compound is 5,7-dihydroxy-6,8-dimethyl flavanone. Four endophytic fungi had been isolated from *S. samarangense* leaves. Antioxidant activity test showed that ethyl acetate extract of endophytic fungi BJA-1 has the highest value. Molecular identification of BJA-1 shows high homology with *Lasiodiplodia venezuelensis* strain CBS 129753.

Keywords: *Syzygium samarangense*, endophytic fungi, antioxidant activity, *Lasiodiplodia venezuelensis*

INTRODUCTION

Free radical is a highly reactive species that can interfere with cell integrity, react with the cell's structural component such as biological macromolecule for membrane building, and react with a functional components such as enzymes and DNA. In human's body, free radical triggers various degenerative disease including cancer, atherosclerosis, hypertension, rheumatoid, arthritis, diabetes, decrease of immunodeficiency system, and accelerates the aging process. Protection of the human body against free radical generally required a particular substance, namely antioxidant (Mary, George, Mathew, & Varghese, 2017; Yadav, Yadav, & Yadav, 2014).

Syzygium samarangense L. known as jambu air, is one of Myrtaceae's family (Mary et al., 2017). Most of part of this plant used for medicinal purpose. Its root, leaves, flowers, and fruits have been used traditionally to cure bronchitis, diabetes, cracked tongue, itchy, asthma, reduce swelling, infection of pathogenic bacteria, digest dysfunction, and kidney disease. Study confirms that extract of these plants shows antioxidant activity, antihyperglycemic cytotoxic, antibacterial, spasmolytic, and immunomodulatory. The *S. samarangense* leaves

contain three C-methylated chalcone, flavonoid, gallic acid, triterpenoids, and volatile terpenoids (Edema & Alaga, 2012; Khandaker, Alebidi, Hossain, Mat, & Boyce, 2015; Khandaker & Boyce, 2016; Insanu, Rmadhania, Halim, Hartati, & Wirasutisna, 2018).

Endophytic is microorganism living inside plant tissue but its existence did not interfere the plant's life. The living organisms can be fungi or bacteria and has been associated since early of plant development (Kaur, Kaur, & Kaur, 2018; Golinska et al., 2015). Either bacteria or fungi live in colony inside plant tissue. Almost all of plants type provides this kind of organism. Every plants has unique living organism that plays significant role as biocontrol agent in plant's development (Santoyo, Hagelsieb, Mosqueda, & Glick, 2016).

Klimova, Rodríguez-Peña, & Sánchez (2017) identified 46 kinds of the endophytic organism inside plant tissue. These organisms consist of 10 bacteria and 36 fungi. Among 36 kinds of fungi, 31 of them come from *Ascomycota* phylum. All of those organisms are capable of producing secondary metabolites with specific antioxidant activity. Endophytic fungi are a good bioactive source due to its ability in producing bioactive in short period and does not require large growing area.

This research is aimed to study the antioxidant activity of secondary metabolites from host plant dan from its endophytes. Results of this research can be used as scientific reference to find new antioxidant agent sources.

EXPERIMENTAL SECTION

Materials and Instrumentation

Fresh *S. samarangense* leaves were taken from Indralaya district, Ogan Ilir regency, South Sumatera. The identification was held at Biology Department, Sriwijaya University. Material for isolation: column chromatography using silica gel Merck 60G (70-230 mesh), TLC Kiesel gel 60 GF₂₅₄, 0,25 mm, 20 x 20 cm using Merck (Art.5554), a series of organics solvent with p.a. and technical grade (distilled before use), distilled water. Reagent for antioxidant activity from Sigma-Aldrich pure chemical industries United Kingdom: DMSO, DPPH, and standard ascorbic acid. Medium for isolation of endophytic fungi using Oxoid: PDA and PDB (200 g potato, 20 g dextrose, in 1 L of H₂O).

Instrumentation in the research were spectrophotometer ultraviolet Beck DU-7500, Perkin Elmer-FTIR spectrometer, ¹H-NMR and ¹³C-NMR spectra on Agilent DD2 (¹H-500 MHz; ¹³C-125 MHz in CDCl₃).

Procedure

Extraction and antioxidant activity test of *S. samarangense* leaves

1.5 kilograms of fresh *S. samarangense* leaves were cleaned and cut to pieces. The sample was dried in room temperature until its mass remain constant. The dried sample was pulverized and macerated using gradient solvent i.e. *n*-hexane, ethyl acetate and methanol. All macerate product was condensed using rotary evaporator to obtain concentrate extract of *n*-hexane, ethyl acetate and methanol fraction. Each fraction was tested against antioxidant activity using DPPH methods. The most active extract was isolated and then identification was carried out to obtain pure compounds.

Antioxidant test using DPPH method

Fraction obtained from previous procedure was dissolved to made 1000, 500, 250, 125, 62.5, 31.25, 15.625 µg/mL concentration. 0.2 mL of each concentration was added by 3.8 mL DPPH 0.5 mM. Mixture was homogenized and left in the dark tube for 30 minutes. The absorption was measured using UV-Vis spectrophotometer at λ_{max} 517 nm. In this test, ascorbic acid was used as a standard positive control and methanol as a negative control. Antioxidant activity can represent by the detention value of DPPH absorption, which was calculated through absorption inhibiting percentage of DPPH and IC₅₀ value (Selvi, Joseph, & Jayaprakasha, 2003; Elfita, Muharni, Munawar, & Rizki, 2012).

$$\%Inhibition = \frac{Control\ Abs. - sample\ Abs.}{Control\ Abs.} \times 100$$

Isolation and identification of pure compound from an active extract

The active extract was analyzed using thin layer chromatography method using solvent of various eluen. The extract (20 g) then was separated to vacuum column chromatography (20 cm x 8 cm) with silica gel (230-400 mesh) as a stationary phase. The pre-absorption prepared sample was injected into the column and was eluted using gradient-polarity solvents, i.e., *n*-hexane-ethyl acetate (10:0→0:10) and ethyl acetate-methanol (10:0→0:10). The eluate was collected in bottles and classified according to column fraction, which was determined by thin layer chromatography results. TLC analysis for each eluate result in 4 column fraction (F1= 7.4 g; F2= 3.2 g; F3= 2.8 g; F4= 5.6 g). Each column fraction was concentrated using rotary evaporator. F3 fraction was used one to process in open column chromatography using *n*-hexane-ethyl acetate-methanol as solvent and the result is 6 column fraction labelled as F3.1 to F3.6 was obtained. Column fraction F3.4 was re-separated in column chromatography and then subject to re-crystallization resulting 23 mg pure compound as a yellow-pale powder which labelled as compound 1. The pure compound was identified using UV, IR, ¹H-NMR, ¹³C-NMR, HSQC, and HMBC spectroscopy and the result was compared with reference data.

Sterilization of *S. samarangense* leaves and isolation of endophytic fungi

Leaves sample were cleaned using flowing water for ± 5 minutes and its surface was sterilized using ethanol 70% for ± 3 minutes followed by aquadest for ± 1 minute and rubbed with HgCl₂ (mercury II chloride) 5% (m/v). After 1 minute, sample was rinsed using sterilized aquadest and cut sideways into several pieces. The sterilized pieces were placed on petri dish contain PDA supplemented with penicillin G (100 units mL) and incubated at 37 °C for 2 x 24 hours. Sample was being observed every day for 3 – 7 days until fungi was grow. Growth fungi colony in PDA medium which is showing different morphology such as shape, color and size was separated. The separation process was carried out by moving single type of colony to new PDA medium in a streak plate and incubated at 37 °C for 2 x 24 hours. The pure fungi colony then prepared for working culture and stock culture by growing it in skewed PDA medium (Muharni, Fitrya, Ruliza, Susanti, & Elfita, 2014; Barik, Tayung, Jagadev, & Dutta, 2010; Khan et al., 2010).

Selection of antioxidant activity from endophytic fungi extracts

All strain of endophytic fungi was cultivated. Fungi inoculum (±10⁶ spores/mL) was inoculated 5% (v/v) into 300 mL PDB medium then placed into 1-liter bottles and incubate at room temperature for four weeks. The medium contains secondary metabolites compound was partitioned into ethyl acetate and evaporated (Elfita, Munawar, Muharni, Pratiwi, & Rahmadania, 2016). Finally, the concentrated medium proceeded to an antioxidant activity screening process to obtained active extract.

Identification of fungi

The fungi was identified in the Biological research Center-LIPI Cibinong. The fungi isolate was identified molecularly based on partial genetic analysis on *Internal Transcribed Spacer* (ITS) *ribosomal DNA locus* of fungi. Pure fungi cultures obtained (BJA-1) was cultivated in PDA medium for three days followed by growing on PDB medium for three days. Mycelium biomass of fungi was harvested, and its DNA was extracted using reagent nucleon PHYTOpure (Amersham LIFE SCIENCE). PCR amplification was conducted on ITS using primary ITS 4: 5'–TCC TCC GCT TAT TGA TAT GC–3' and primary ITS 5: 5'–GGA AGT AAA AGT CGT AAC AAG G–3' (White, Bruns, Lee, & Taylor, 1990). Purification of PCR product was carried out by using *PEG Precipitation methods* (Hiraishi, Kamagata, & Nakamura, 1995), followed by sequencing cycle. The results of sequencing were purified in Ethanol Purification Methods. Base sequence reading was analyzed using *automated DNA Sequencer* (ABI PRISM 3130 Genetic Analyzer (Applied Biosystems)).

Raw data from sequence results was being *trimming* dan *assembling* using SeqTrace program. The sequence data (contig) was compared to determine the homology/ similarities molecularly with BLAST (<http://ncbi.nlm.nih.gov/BLAST/>). BLAST analysis is based on *database nucleotide collection (nt/nr)* which limited by *sequence to type material*. The BLAST analysis results in the form of an FASTA aligned sequences file with the extension .txt taken from the highest percentage of similarity values. BLAST results were processed using the Clustal W application to produce files with MSF and PHYLIP extensions. PHYLIP files were processed with the Seqboot application with 1000 replicates. The results of the Seqboot application were processed into the DNAdist application with 1000 replicates and then into the Neighbour application with 1000 replicates to produce an outtree. This outtree file was processed into the Consense application to produce the final file extension .phb. The phylogenetic tree was visualized by Treeview v1.4.3 program (Yohandini, Julinar, & Muharni, 2017).

RESULTS AND DISCUSSION

Dried *S. samarangense* leaves as much as 615 g was obtained 1.5 kg of fresh leaves. 500 g of powdered was extracted consecutively using *n*-hexane, ethyl acetate and methanol for 2 x 24 hours with three times repetitions. Concentrate of 31.7 g *n*-hexane fraction, 28.2 g ethyl acetate fraction and 25.6 g methanol fraction were obtained after vacuum evaporation. All three fraction were tested for antioxidant activity, and results are shown in following Table 1. Ethyl acetate showing the highest antioxidant activity.

Table 1. IC₅₀ values of *S. samarangense* leaves of all fractions, and ascorbic acid on antioxidant activity test using DPPH method.

Test sample	Fraction	IC ₅₀ (µg/mL)
<i>S. samarangense</i> 's leaves	<i>n</i> -Hexane	94.01
	Ethyl acetate	74.37
	Methanol	929.22
Ascorbic Acid	Ascorbic Acid	22.23

The chemical compound from ethyl acetate fraction of *S. samarangense* leaves

Compound 1. A yellow-pale powder. UV spectra (MeOH) absorption at λ_{max}: 295 and 341 nm. UV spectra (MeOH + NaOH) absorption at λ_{max}: 339 and 341 nm. IR peak at 3271.8 (OH), 2925.1 (CH₂phatic), 1633.2 (C=O), 1607.7-1468.8 (C₆H₅). NMR (¹H-500 MHz; ¹³C-75 MHz; HSQC and HMBC in CDCl₃) see Figure 1 and Table 2.

The ¹H-NMR spectrum of compound 1 shows nine signals with 16 total protons. There were five aromatic protons appears at δ_H 7.30 - 7.50 ppm. Furthermore, two methyl signals were detected at δ_H 2.08 (3H; s) and 2.07 (3H; s). Moreover, there are three protons at chemical shift around 3-6 ppm and two hydroxyl protons.

According to phytochemical test, compound 1 is a flavonoid. The typical characteristic of flavonoid indicates by proton signal at low field δ_H 12.27 (1H; s) which marks the existence of –OH chelat at C-5 and chelated carbonyl carbon at C-4. Multiplicity pattern of these five aromatic protons implies it is an aromatic ring labeled as ring B. Ring A, therefore has no aromatic proton. The appearance of three proton signal at 3-6 ppm indicates ring C from compound 1 has no double bond neither has substituted hydroxyl groups. This suggests that the flavonoid obtained is a flavanone group compound.

¹³C-NMR spectra show 17 signals in which 15 of them belong to flavanone structure and 2 of them are methyl groups bound to ring A. The compound 1 is identify as flavanone can be confirmed from the appearance of carbonyl carbon signals at δ_C 196.6 ppm. The spectrum also shows that the compound has three oxyaryl carbon atoms (=C-O) that are at δ_C 159.5; 157.8 and 161.0 ppm. It indicates the existence of two hydroxyl groups at flavonoid structure, which bound to ring A. Identification of carbon and proton signals furthermore were set according to NMR 2D spectrum (Table 2). HMQC spectrum was used to identify carbon signals which bound to H and showing H – C correlation having distance by two or three bonds.

According to ¹H-NMR, ¹³C-NMR, HMQC, and HMBC spectrum obtained compared to the reference, it can be concluded that compound 1 is 5,7-dihydroxy 6,8-dimethyl flavanone (Figure 2). The molecular formula is C₁₇H₁₆O₄ with DBE = 10.

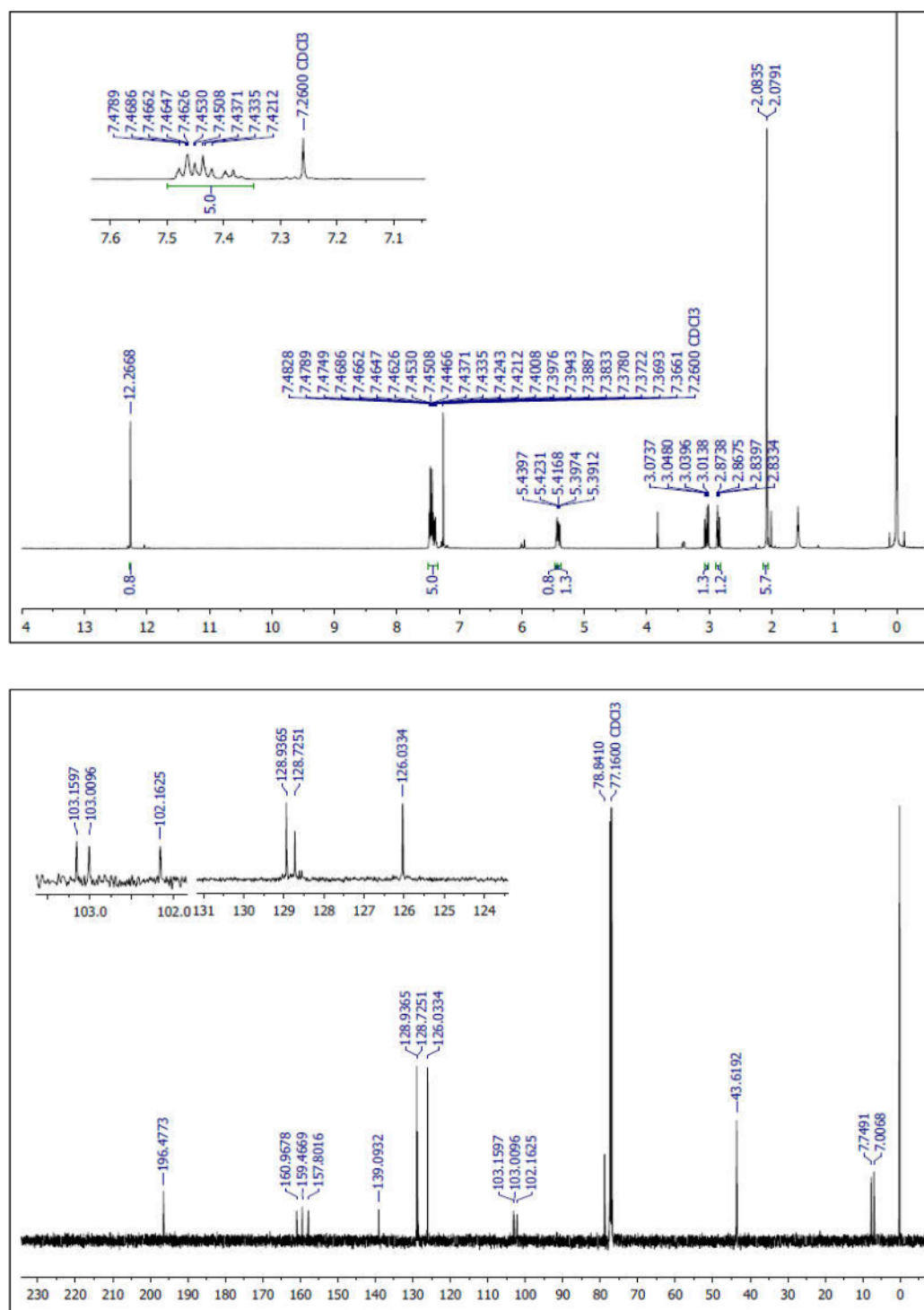


Figure 1. 500 MHz ¹H-NMR and 125 MHz ¹³C-NMR spectra of compound 1 in CDCl₃.

Table 2. NMR data of Compound 1 recorded at ¹H-500 MHz; ¹³C-125 MHz in CDCl₃ and 1* (5,7-dihydroxy-6,8-dimethyl flavanone: recorded at ¹H-500 MHz; ¹³C-125 MHz in CD₃OD) (Memon et al., 2015)

Position of C	δ _c ppm 1	δ _c ppm 1*	δ _H ppm (ΣH, multiplicity, J (Hz)) 1	HMBC 1
2	78.8	78.6	5.42 (24; dd; J=3.15; 3.15 Hz)	
3	43.6	42.82	3.04 (24; dd; J=3.15; 12.85 Hz) 2.84 (1H; dd; J=12.85; 3.15 Hz)	78.8; 196.5
4	196.5	196.6		
5	159.5	163.0		
6	102.2	102.8		
7	161.0	162.8		
8	103.0	101.9		
9	157.8	158.9		
10	103.2	103.5		
1'	139.1	139.4		
2'	128.9	128.3	7.42 (1H; m)	128.9; 139.1
3'	126.0	125.8	7.46 (39; m)	126.0; 128.7
4'	128.7	128.0	7.40 (1H; m)	126.0
5'	126.0	125.8	7.46 (1H; m)	126.0; 128.7
6'	128.9	128.3	7.42 (1H; m)	128.9; 139.1
6-CH ₃	7.7	6.75	2.08 (3H; s)	161.0; 159.5; 102.2
8-CH ₃	7.0	6.0	2.07 (3H; s)	103.0; 157.8
5-OH			12.27 (1H; s)	103.2; 159.5
7-OH			5.44 (1H; s)	

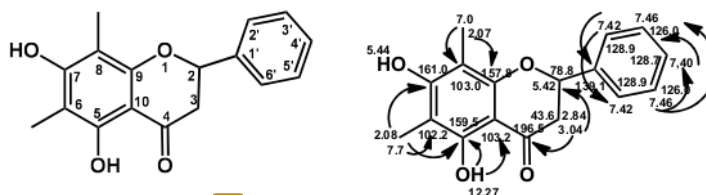


Figure 2. Structure of 5,7-dihydroxy 6,8-dimethyl flavanone and HMBC correlation

Result of antioxidant test shows that compound 1 is inactive with IC₅₀ value > 100 µg/mL. During DPPH measurement, reaction occurs between radical species of DPPH and tested compound. The absorption spectrum of residual radical species of DPPH after react with substrat was observed. Molecular structure of a compound affects its antioxidant activity. Compounds from flavonoid groups has abundant hydroxyl group which plays significant roles to antioxidant activity. Antioxidant activity increases if the compound has a unit known as cathecol or 1,2-dihydroxyfenil. Compound with cathecol unit or 1,2-dihydroxyfenil is more easily experience proton abstraction by free radical to form more stable radical. This stability established because of free radical forms intramolecular hydrogen bond with hydroxyl and diketones groups by its side. Like wise, compound with hydroxyl groups at para position against carbonyl group has proton easily abstracted by free radical to form more stabil radicals. This stability occurrence is caused by distribution and delocalization of electrons. The double bond and carbonyl groups in basic structure with heterocyclic rings will enhance its antioxidant activity. The radical stabilization was created through conjugation

and electron delocalization (Procházková, Boušová, & Wilhelmová, 2011; Brunetti et al., 2013; Sawai & Sakata, 1998; Heim, Tagliaferro, & Bobilya, 2002). However, compound 1 from flavonoid groups has least hydroxyl group in addition to no double bond on its heterocyclic ring. Moreover, the existence of chelated hydroxyl group along with carbonyl makes proton abstraction by free radical to form more stable radical is more difficult.

Antioxidant activity of endophytic fungi

Four endophytic fungi were isolated from *S. samarangense* leaves and were labeled as BJA1 – BJA4. The fungi was stored in petri dish contains medium as working culture and stock culture for colony shapes observation. Each fungus isolate cultivated in 3 x 300 mL liquid PDB medium for four weeks. Partition extraction of cultivation medium conducted with ethyl acetate were able to yields 1.47; 1.62; 1.75; and 1.34 concentrate extracts respectively. Four fungi extracts were tested for antioxidant activity. The IC₅₀ of all *S. samarangense* leaves fractions, and ethyl acetate extract of endophytic fungi fraction using DPPH methods is display in Table 3.

Table 3. IC₅₀ values of ethyl acetate extracts of endophytic fungi isolated from *S. samarangense* leaves and ascorbic acid on antioxidant activity test using DPPH method.

Test sample	Fraction	IC ₅₀ (µg/mL)
Endophytic fungi	Ethyl acetate BJA-1	49.96
	Ethyl acetate BJA-2	852.86
	Ethyl acetate BJA-3	84.86
	Ethyl acetate BJA-4	616.08
Ascorbic Acid	Ascorbic Acid	22.23

Endophytic fungi cultivation medium was effectively extracted by using ethyl acetate to isolate a secondary metabolites compound from endophytic fungi. Ethyl acetate solvent is selectively able to extract phenolic compound with low molecular mass and polyphenol with high molecular mass (Yadav et al., 2014; Killa et al., 2015). IC₅₀ value at Table 1 confirms that ethyl acetate extract of endophytic fungi, BJA-1 is more active than leaves extract and second metabolite of its host plant.

Medicinal plants usually contain endophytic fungi which produce identic antioxidant activity its secondary metabolites. *Syzygium samarangense* is one of the medicinal plants widely used in Indonesia, and some other countries contain various phytochemical compound such as saponins, alkaloids, flavonoids, tannins, phenolic, glycosides and terpenoid which contribute to its properties (Edema & Alaga, 2012; Madhavi Ram, 2015).

Identification of endophytic fungi

The result of identification according to phylogenetic tree built in 1000 bootstrap replicates shows strain BJA-1 has highest homology with *Lasiodiplodia venezuelensis* strain CBS 129753 (Figure 3). Sequence of base area ITS rDNA: Contig BJA-1:

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TAGGTGAACCTGCGGAAGGATCATTACCGAGTTC
TCGAGCTCCGGCTCGACTCTCCACCCTTTGTGA
ATGTACCTCTGTTGCTTTGGCGGCCCGGGCCGCC
AGAGGACCTTCAAACCTCCAGTCAGTGAACGCAG
ACGTCTGAAAACAAGTCAATAAACTAAAACCTTTC
AACACCGATCTCTTGGTTCTGGCATCGATGAAG
AACGCAGCGAAATGCGATAAGTAATGTGAATTG
CAGAATTCAGTGAATCATCGAATCTTTGAACGCA
CATTGCGCCCTTGGTATTCCGGGGGGCATGCCT
GTTTCGAGCGTCATTACAACCCTCAAGCTCTGCTT
GGCATTGGGCTCCGTCCTCACTGCGGACGCGCCT
CAAAGACCTCGCGGGTGGCTGTTCCGCCCTCAAG
CGTAGTAGAATACACCTCGCTTTGGAGCGGTCCGG
CGTCGCCCCGCCGACGAACTTCTGAACTTTTCT
CAAGGTTGACCTCGGATCAGGTAGGATACCCG
CTGAACTTA
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Molecular identification and analysis with phylogenetic tree of BJA-1 shows high homology with *Lasiodiplodia venezuelensis* strain CBS 129753. This proves that endophytic fungi is capable to produce secondary metabolites with higher biological activity than its host. The *Lasiodiplodia venezuelensis* strain is recommended for further investigations.

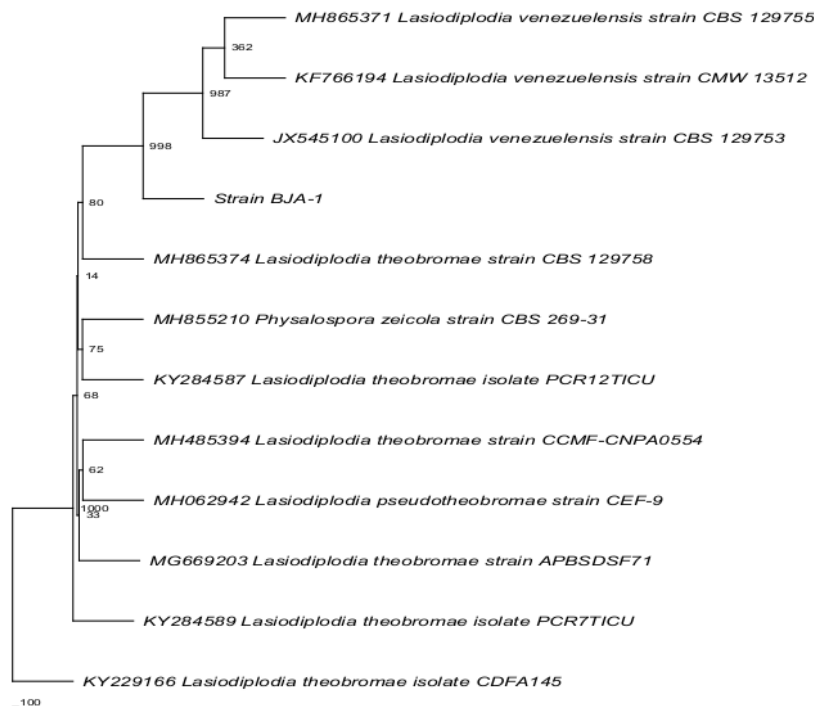


Figure 3. Phylogenetic tree of strain BJA-1 was visualized by Treeview program with 1000 bootstrap replicates

Bioactive compound exploration from endophytic fungi has the advantage of short lifespan and compound availability to produce in large scale through the fermentation process. Isolation of bioactive compound from host plants has many obstacles because the scarcity of host plant and its relatively long life cycle. Hence the endophytic fungi have good prospect as a source to discover new antioxidant compound.

CONCLUSIONS

In this study, a pure compound from the active fraction of antioxidant has been found, but the compound is not antioxidant active. Further studies are needed to elute the secondary metabolites of antioxidants found in the ethyl acetate fraction of the leaves of *S. samarangense*. This study also revealed the presence of secondary metabolites that have antioxidant activity produced by *Lasiodiplodia venezuelensis* endophytic fungus strain CBS 129753 from *S. samarangense* leaves. Further studies are needed to isolate and purify responsible bioactive compounds as antioxidants in ethyl acetate extract of endophytic fungus. These findings indicate that endophytic fungi from antioxidant plants could be potential for a source of antioxidant compounds.

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REFERENCES

- Barik, B.P., Tayung, K., Jagadev, P.N., & Dutta, S.K. (2010). Phylogenetic placement of an endophytic fungus *Fusarium oxysporum* isolated from *Acorus calamus* rhizomes with antimicrobial activity. *European Journal of Biological Sciences*, 2, 8-16.
- Brunetti, C., Martina, D., Ferdinando, M.D., Fini, A., Pollastri, S., & Tattini, M. (2013). Flavonoids as antioxidants and developmental regulators: relative significance in plants and humans. *International Journal of Molecular Sciences*, 14, 3540-3555.
- Edema, M.O., & Alaga, T.O. (2012). Comparative evaluation of bioactive compounds in *Hibiscus sabdariffa* and *Syzygium samarangense* juice extracts. *African Crop Science Journal*, 20 (3), 179-187.
- Elfita, E., Muharni, M., Munawar, M., & Rizki, R. (2012). Isolation of antioxidant compound from endophytic fungi *Acremonium* sp from the twigs of Kandis Gajah (*Garcinia griffithii* T, Anders). *Makara Journal of Science Series*, 16 (1), 46-50.
- Elfita, E., Munawar, M., Muharni, M., Pratiwi, G., & Rahmadania, R. (2016). A benzoyl compound isolated from endophytic fungi of kandis gajah (*Garcinia griffithii*) and asam kandis (*Garcinia cowa*). *Makara Journal of Science*, 20 (4), 18-23.
- Golinska, P., Wypij, M., Agarkar, G., Rathod, D., Dahm, H., & Rai, M. (2015). Endophytic actinobacteria of medicinal plants: Diversity and bioactivity. *Antonie Van Leeuwenhoek International Journal of General*, 108 (2), 267-289.
- Heim, K.E., Tagliaferro, A.R., & Bobilya, D. J. (2002). Flavonoid antioxidant: chemistry, metabolism and structure-activity relationships. *Journal of Nutritional Biochemistry*, 13, 572-584.
- Hiraishi, A., Kamagata, Y., & Nakamura, N. (1995). Polymerase chain reaction amplification and restriction fragment length polymorphism analysis of 16S rRNA genes from methanogens. *Journal of Fermentation and Bioengineering*, 79, 523-529.
- Insanu, M., Rmadhania, Z.M., Halim, E.N., Hartati, R., & Wirasutisna, K.R. (2018). Isolation of 5,7-dihydroxy, 6,8-dimethyl flavanone from *Syzygium aqueum* with its antioxidant and xanthine oxidase inhibitor activities. *Pharmacognosy Research*, 10 (1), 60-63.
- Kaur, J., Kaur, R., & Kaur, A. (2018). Evaluation of antidiabetic and antioxidant potential of endophytic fungi isolated from medicinal plants. *International Journal of Green Pharmacy*, 12 (1), 6-12.
- Khan, R., Shahzad, S., Choudhary, M.I., Shakeel, A., Khan, S.A., & Ahmad, A. (2010). Communities of endophytic fungi in medicinal plant *Withania somnifera*. *Pakistan Journal of Botany*, 42 (2), 1281-1287.
- Khandaker, M.M., Alebidi, A.I., Hossain, A.S., Mat, N., & Boyce, A.N. (2015). Physiological and biochemical properties of three cultivars of wax apple (*Syzygium samarangense* [Blume] Merrill & L.M. Perry) fruits. *Journal of Sustainability Science and Management*, 10 (1), 66-75.
- Khandaker, M.M., & Boyce, A.N. (2016). Growth, distribution and physiochemical properties of wax apple (*Syzygium samarangense*): A Review. *Australian Journal of Crop Science*, 10 (12), 1640-1648.
- Khiralla, A., Mohamed, I., Thomas, J., Mignard, B., Spina, R., Yagi, S., & Mattar, L.D. (2015). A pilot study of antioxidant potential of endophytic fungi from some Sudanese medicinal plants. *Asian Pacific Journal of Tropical Medicine*, 8 (9), 701-704.
- Klimova, E.M., Rodríguez-Peña, K., & Sánchez, S. (2017). Endophytes as sources of antibiotics. *Biochemical Pharmacology*, 134, 1-17.
- Madhavi, M., & Ram, M.R. 2015. Phytochemical screening and evaluation of biological activity of root extracts of *Syzygium samarangense*. *International Journal of Research in Pharmacy and Chemistry*, 5 (4), 753-763.
- Mary, L.M.K., George, N.S.P., Mathew, N., & Varghese, S.E. 2017. Antioxidant activity of wax jambu. *European Journal of Pharmaceutical and Medical Research*, 4 (10), 227-234.
- Memon, A.H., Ismail, Z., Al-Suede, F.S.R., Aisha, A.F.A., Hamil, M.S.R., Saeed, M.A.A., Laghari, M., & Majid, A.M.S.A. (2015). Isolation, characterization, crystal structure elucidation of two flavanones and simultaneous RP-HPLC determination of five major compounds from *Syzygium campanulatum* Korth. *Molecules*, 20, 14212-14233.

- 1 Muharni, M., Fitrya, F., Ruliza, M.O., Susanti, D.A., & Elfita, E. (2014). Di-(2-ethylhexyl)phthalate and pyranone derivated from endophytic fungi *Penicillium* sp the leave of Kunyit Putih (*Curcuma zedoaria*). *Indonesian Journal of Chemistry*, 14 (3), 290 – 296.
- 16 Procházková, D., Boušová, I., & Wilhelmová, N. (2011). Antioxidant and prooxidant properties of flavonoids. *Fitoterapia*, 82, 513–523.
- Santoyo, G., Hagelsieb, M.G., Mosqueda, D.C.O.M., & Glick, B.R. (2016). Plant growth-promoting bacterial endophytes. *Microbiological Research*, 183, 92–99.
- 9 Sawai, Y., & Sakata, K. (1998). NMR Analytical Approach to Clarify the Antioxidative Molecular Mechanism of Catechins Using 1,1-Diphenyl-2-picrylhydrazyl. *Journal of Agricultural and Food Chemistry*, 46, 111-114.
- 32 Selvi, A.T., Joseph, G.S., & Jayaprakasha, G.K. (2003). Inhibition of growth and aflatoxin production in *Aspergillus flavus* by *Garcinia indica* extract and its antioxidant activity. *Food Microbiology*, 20, 455-460.
- 14 White, T.J., Bruns, T.D., Lee, S.B., & Taylor, J.W. (1990). Amplification and direct sequencing of fungal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sninsky JJ, White TJ. (eds). *PCR protocols*. Academic; San Diego, 315-322.
- 3 Yadav, M., Yadav, A., & Yadav, J.P. (2014). In vitro antioxidant activity and total phenolic content of endophytic fungi isolated from *Eugenia jambolana* Lam. *Asian Pacific Journal of Tropical Medicine*, 7 (Suppl 1), S256-S261.
- 51 Yohandini, H., Julinar, J., & Muharni, M. (2015). Isolation and phylogenetic analysis of thermophile community within Tanjung Sakti hot spring, South Sumatera, Indonesia. *Hayati Journal of Biosciences*, 22, 143-148.

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