The potential source of natural antioxidant agent of *Casia alata* microgreen

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ABSTRACT

Commonly, plant was cultivated by microgreen have potentially source of natural antioxidant agents. This study was conducted to utilize Cassia alata (C. alata) as a microgreen and evaluated the potential of Microgreen gelinggang as the source of natural antioxidant agents. The seed of Cassia alata was cultivated in Rockwool at room temperature (27±1ºC). At the appearance of the first true leaves, about 21 days, microgreens were harvested from a triplicate of trays with sterilized scissors. The antioxidant activity assay using the DPPH (2,2-difenil-1-pikrilhidrazil) radical scavenging activity method. It was analyzed using spectrophotometry UV-VIS. The result showed that the IC_{50} *values of Microgreen gelinggang were* $1.789x10^3$ *±0.0 µg/mL. It was a weak category antioxidant. This study indicated that the extract of Microgreen gelinggang has a potential source of natural antioxidant agents.*

Keywords*: antioxidant activity, Cassia alata, DPPH radical scavenging activity, microgreen*

INTRODUCTION

In current years, microgreen has popular and is often used as a culinary (Ghoora et al., 2020) for consumers concerned about their health (Supapvanich et al., 2020). Microgreens is a new type of vegetable, immature plants, and harvested at the first true-leaf stage on 10-14 days (Tan et al., 2020); (Turner et al., 2020). It has been reported that microgreens are high in phytochemicals (Marton et al., 2010); (Xiao et al., 2012) and antioxidants (Senevirathne et al., 2019) The antioxidant has a critical role in preventing cell damage (Yadav et al., 2016) and contributes to health benefits (Grosso et al., 2013). According to the study by Ghoora et al., (2020), some microgreen plant, like onion, mustard, carrot, and fennel, contain DPPH antioxidant activity IC_{50} 452.4±51.3; 168.4±14.8; 97.6±2.1; 94.3±0.7 µg/mL, respectively. However, there has been no further research related to *C. alata* microgreens.

C. alata (*Casia alata*) is a native plant from Argentina. In Indonesia, it is known as “Ketepeng Cina” (Fatmawati et al., 2020). This plant can grow in the tropics, mainly in South Kalimantan. *C. alata* is a type of herb plant (Chatterjee, 2012). In South Kalimantan, the extract of *C. alata* leaves is commonly used as the traditional herb for skin disease. According to Oladeji et al. (2020), the extract of *C. alata* leaves is commonly used as the traditional herb for typhoid, diabetes, malaria, asthma, ringworms, tinea infections, scabies, blotch, herpes, and eczema. The seeds and leaves of *C. alata* can be used as an antimicrobial (Abdulwaliyu et al., 2013), anti-inflammatory (Wongkaew & Sinsiri, 2014), antidiabetic (Abdulwaliyu et al., 2013), and antifungal (Wongkaew & Sinsiri, 2014).

Several studies have shown that *C. alata* is rich in antioxidants (Fatmawati et al., 2020), such as ascorbic acid, flavonoid, tocopherol, anthraquinone, and carotene (Chatterjee, 2012). Thus, this study was conducted to the utilization of *Cassia alata* as a microgreen. The aim of this study evaluates potencial of Cassia alata microgreen as source of natural antioxidant agents.

MATERIAL AND METHODS

Plant Material

The seed of Cassia alata L. was obtained from PT. Sari Kaya Sega Utama, Banjarbaru, South Kalimantan. This study was adopted from Ghoora et al. (2020). The seed was cultivated in Rockwool for 21 days at room temperature (27±1 ºC). Before the seed was cultivated, it must be soaked in water for 5 hours. At the appearance of the first true leaves microgreens were harvested from a triplicate of trays with sterilized scissors. Microgreens were washed to remove extraneous dirt, washed with deionized water, and fan-dried for 5-10 min. Cleaned microgreens were frozen at -20±1oC before used.

Preparation of the extract

The extraction process was adapted from Sen et al. (2013). The frozen microgreens were air-dried prior to grinding. 250 g powdered microgreens were extracted with 100% methanol (Sigma-Aldrich) using maceration methods (sample-solvent ratio of 1:3) for 3 days. The extracts were stored at 4±1ºC.

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DPPH radical scavenging activity assay

The DPPH radical scavenging activity assay using spectrophotometry (Hitachi, U2900) was adopted from Senevirathne et al. (2019). 50 µl of samples with various concentrations (1.00; 1.33; 1.66; 1.99 and 2.33 mg/mL), 1.0 ml of DPPH 0.4 mM, and 3.950 ml of ethanol were homogenized using the vortex for 30 minutes. The control consisted of 1.0 ml of DPPH (Sigma-Aldrich) and 4.0 ml of ethanol (Sigma-Aldrich). The absorbance of samples was measured at 517 nm, and 50% inhibitory concentration (IC50) was calculated.

Statistical analysis

The data obtained was then analyzed using ANOVA with a p value of 5%. Results that showed the differences were further tested using the Duncan Multiple Range Test alpha 5%.

RESULT AND DISCUSSION

Figure 1 shows the C. alata microgreen leaves at 21 days. It will be extracted for an antioxidant activity assay. Based on this study, C. alata microgreen was classified as a weak antioxidant. Among factors contributing to weak antioxidant are extraction technique and stages of harvest maturity. According to Sultana et al. (2009), the extraction solvent and technique used can affect the antioxidant activity. Similar results were observed by and Hill et al. (2019). The study of El-Nakhel et al. (2020) conducted, the stage of harvest maturity can affect the antioxidant capacity of microgreens. Antioxidants are compounds that act to neutralize free radicals and prevent the damage of the normal cell. The performance of antioxidant activity is determined based on its ability to free radicals scavenging. Commonly, 2,2-diphenyl-1-picrylhydrazyl (DPPH) is a free radical that is used (Albaar, 2015).

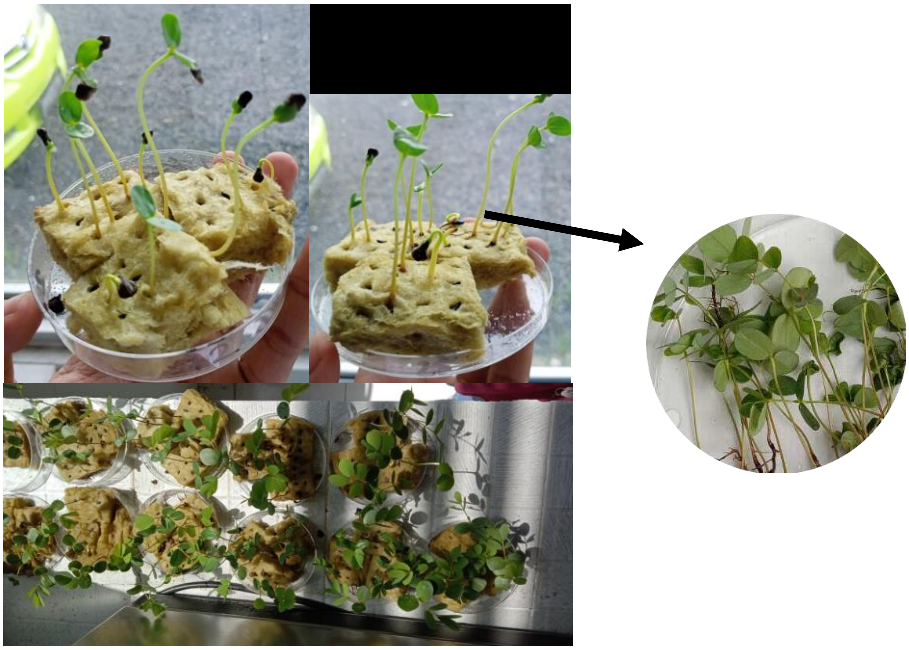

Figure 1. *C. alata* microgreen at 21 days

Table 1. Antioxidant activity of *C. alata* microgreen

Concentration (mg/ml)	Abs.			% Free radical scavenging (%)			Average
	1	2	3	1	2	3	
1.00	0.633	0.637	0.634	32.08	31.65	31.97	31.90
1.33	0.545	0.541	0.544	41.52	41.95	41.63	41.70
1.66	0.482	0.491	0.486	48.28	47.32	47.85	47.82
1.99	0.422	0.430	0.428	54.72	53.86	54.08	54.22
2.33	0.361	0.366	0.365	61.27	60.73	60.84	60.94

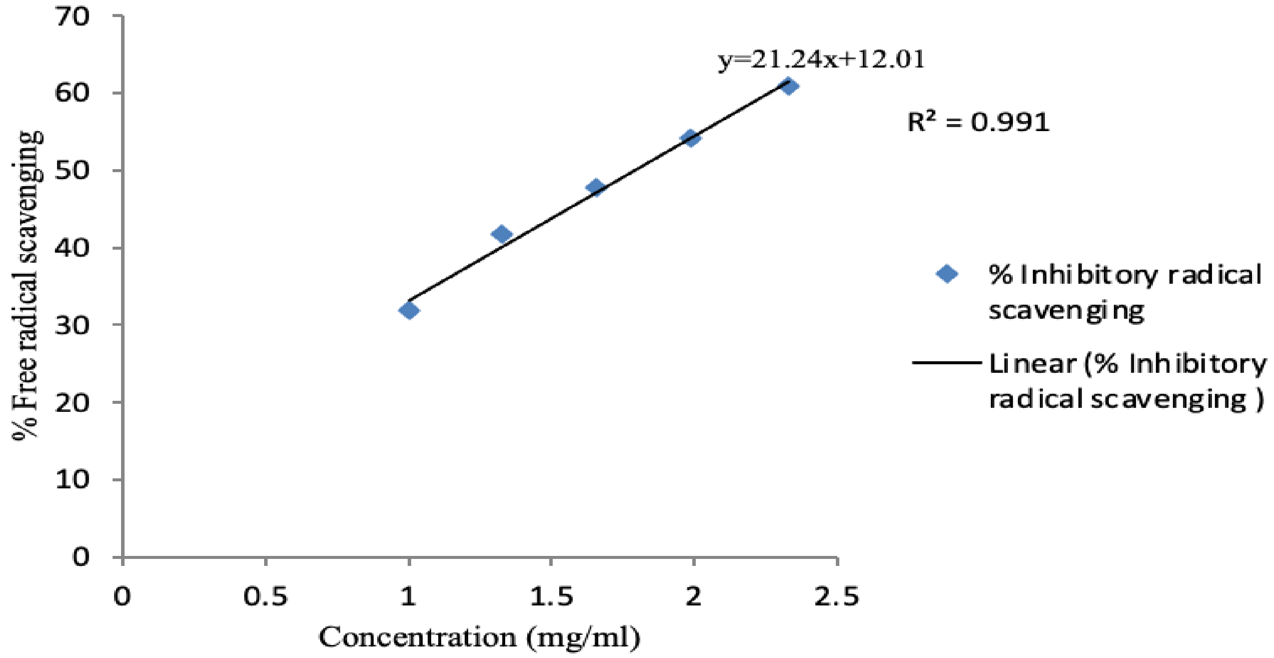

Figure 2. Correlation of concentration and % inhibitory radical scavenging

Table 1 shows the measuring antioxidant activity using the DPPH free radicals scavenging method. The results showed that the absorption formed a calibration curve with a concentration range of 1.00; 1.33; 1.66; 1.99, and 2.33 mg/mL at 517 nm.. The results gave a linear relationship within the concentration and % free radical scavenging (%), described in the form of a linear regression equation y = 21.24x + 12.01 with R2 0.991 (Figure 1). IC50 on microgreen C. alata was obtained by transforming the absorbance data (y) into %-free radical scavenging (%). The IC50 value of C. alata Microgreen was 1.789x103 ±0.01 µg/mL.

This result is higher than the study by Senevirathne et al. (2019), like finger millet and green peas, contain DPPH antioxidant activity IC50 4339 ± 86, and 1830 ± 109 µg/mL, respectively. The IC50 value is the parameter of antioxidant activity. The higher antioxidant activity, the lower IC50 (Rivero-cruz et al., 2020). According to (Qusti et al., 2010), the category of antioxidants is classified very strong (IC50<0.01 mg/mL), strong (0.01 mg/ml< IC50<1 mg/mL), moderate (1 mg/ml< IC50<7 mg/mL), and weak (IC50>7 mg/mL). Meanwhile, according to (Molyneux, 2018), the antioxidants are weak because of IC50 values 200-1000 µg/mL and it is considered a source of antioxidants..

Based on this research, gelinggang microgreen is classified as a weak antioxidant. The extraction technique and lack of stages of harvest maturity is one of factor that causing this. Sultana et al. (2009) showed that the solvent and technique extraction can influence antioxidant activity. Similar results were observed by Hill et al. (2019). El-Nakhel et al. (2020) showed that the level of harvest maturity can influence the antioxidant capacity of microgreens.

CONCLUSION

This paper utilized C. alata to microgreen and determined the potential of C. alata microgreen as the source of natural antioxidant agents. C. alata microgreen had the IC50 1.789x103 ±0.01 µg/mL and was classified as a weak antioxidant. Thus, it would be recommended to optimization the extraction technique and complement mature leafy by microgreen to derive maximum antioxidant activity.

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REFRENCES

Abdulwaliyu, I., Arekemase, S.O., Bala, S., Ibraheem, A., Dakare, A.M., Sangodare, R., & Gero, M. (2013). Nutritional Properties of Senna alata Linn Leaf and Flower. International Journal of Modern Biology and Medicine, 4(1), 1-11

Albaar, N.M. (2015). Aktivitas antioksidan jus rumput gandum (triticum aestivum) sebagai minuman kesehatan dengan metode DPPH. Jurnal Mkmi

1(September), 197–202.

Chatterjee, S. (2012). Study of Antioxidant Activity and Immune Stimulating Potency of the Ethnomedicinal Plant, Cassia alata (L.) Roxb. Medicinal & Aromatic Plants, 02(04), 2–7. DOI:10.4172/2167-0412.1000131.

El-Nakhel, C., Pannico, A., Graziani, G., Kyriacou, M.C., Giordano, M., Ritieni, A., Pascale, S. De, *et al*. (2020). Variation in macronutrient content, phytochemical constitution and in vitro antioxidant capacity of green and red butterhead lettuce dictated by different developmental stages of harvest maturity. Antioxidants, 9(4). DOI:10.3390/antiox9040300.

Fatmawati, S., Yuliana, Purnomo, A.S., & Abu Bakar, M.F. (2020). Chemical constituents, usage and pharmacological activity of Cassia alata. Heliyon, 6(7), e04396. Elsevier Ltd. DOI:10.1016/j.heliyon.2020.e04396.

Ghoora, M.D., Haldipur, A.C., & Srividya, N. (2020). Comparative evaluation of phytochemical content, antioxidant capacities and overall antioxidant potential of select culinary microgreens. Journal of Agriculture and Food Research 2(March): 100046. Elsevier Ltd. DOI:10.1016/j.jafr.2020.100046.

Grosso, G., Bei, R., Mistretta, A., Marventano, S., Calabrese, G., Masuelli, L., Giganti, M.G., *et al.* (2013). Effects of vitamin C on health: A review of evidence. Frontiers in Bioscience, 18(3), 1017–1029. DOI:10.2741/4160.

Hill, C.V.L., Horozić, E., Kolarević, L., Zukić, A., Bjelošević, D., & Mekić, L. (2019). Original Paper Effects of Extraction Solvent and Technique on the. DOI:10.5457/503.

Marton, M., Mandoki. Z., Caspo J., Caspo-Kiss, Z., Marton, M., Mándoki, Z., Marton, M., Mandoki. Z., Caspo J., & Caspo-Kiss, Z. (2010.) The role of sprouts in human nutrition. A review. Alimentaria, Hungarian University of Transylvania, 3, 81–117. Retrieved from http://www.acta.sapientia.ro/acta-alim/C3/alim3-5.pdf

Molyneux, P. (2018). The use of the stable free radical diphenylpicryl-hydrazyl (DPPH) for estimating antioxidant activity. (May):

Oladeji, O.S., Adelowo, F.E., Oluyori, A.P., Bankole, D.T. 2020. Ethnobotanical Description and Biological Activities of Senna alata. Evidence-based Complementary and Alternative Medicine 2020. DOI:10.1155/2020/2580259.

Qusti, S., Abo-khatwa, A.N., Lahwa, M.A. 2010. Screening of antioxidant activity and phenolic content of selected food items cited in the holly quran. European Journal of Biological Sciences. 2(1): 40-51

Rivero-cruz, J.F., Granados-pineda, J., Pedraza-chaverri, J., Rivero-cruz, B.E. 2020. And Antimicrobial Activities of the Ethanolic Extract of Mexican Brown Propolis. Antioxidants 9(70): 1–11.

Sen, S., De, B., Devanna, N., Chakraborty, R. 2013. Total phenolic, total flavonoid content, and antioxidant capacity of the leaves of Meyna spinosa Roxb., an Indian medicinal plant. Chinese Journal of Natural Medicines 11(2): 149–157. China Pharmaceutical University. DOI:10.1016/S1875-5364(13)60042-4.

Senevirathne, G.I., Gama-Arachchige, N.S., Karunaratne, A.M. 2019. Germination, harvesting stage, antioxidant activity and consumer acceptance of ten microgreens. Ceylon Journal of Science 48(1): 91. DOI:10.4038/cjs.v48i1.7593.

Sultana, B., Anwar, F., Ashraf, M. 2009. Effect of extraction solvent/technique on the antioxidant activity of selected medicinal plant extracts. Molecules 14(6): 2167–2180. DOI:10.3390/molecules14062167.

Supapvanich, S., Sangsuk, P., Sripumimas, S., Anuchai, J. 2020. Efficiency of low dose cyanocobalamin immersion on bioactive compounds contents of ready to eat sprouts (sunflower and daikon) and microgreens (red-amaranth) during storage. Postharvest Biology and Technology 160(July 2019): 111033. Elsevier. DOI:10.1016/j.postharvbio.2019.111033.

Tan, L., Nuffer, H., Feng, J., Kwan, S.H., Chen, H., Tong, X., Kong, L. 2020. Antioxidant properties and sensory evaluation of microgreens from commercial and local farms. Food Science and Human Wellness 9(1): 45–51. Beijing Academy of Food Sciences. DOI:10.1016/j.fshw.2019.12.002.

Turner, E.R., Luo, Y., Buchanan, R.L. 2020. Microgreen nutrition, food safety, and shelf life: A review. Journal of Food Science 85(4): 870–882. DOI:10.1111/1750-3841.15049.

Wongkaew, P., Sinsiri, W. 2014. Effectiveness of Ringworm Cassia and Turmeric Plant Extracts on Growth Inhibition against Some Important Plant Pathogenic Fungi. American Journal of Plant Sciences 05(05): 615–626. DOI:10.4236/ajps.2014.55076.

Xiao, Z., Lester, G.E., Luo, Y., Wang, Q. 2012. Assessment of vitamin and carotenoid concentrations of emerging food products: edible microgreens. Journal of agricultural and food chemistry 60(31): 7644–7651. United States. DOI:10.1021/jf300459b.

Yadav, A., Kumari, R., Yadav, A., Mishra, J.P., Seweta, S., Prabha, S. 2016. Antioxidants and its functions in human body. Research in Environment and Life Sciences 9(11): 1328–1331.