RINI PRASTIWI-Phytochemical Evaluation and Antioxidant Activity of Virginia tobacco Leaves (Nicotiana tabacum L. var virginia) Fractions with DPPH and FTC Methods

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Phytochemical Evaluation and Antioxidant Activity of Virginia tobacco Leaves (*Nicotiana tabacum* L. var virginia) Fractions with DPPH and FTC Methods

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ABSTRACT

Introduction: Virginia tobacco (*Nicotiana tabacum* var. Virginia) is one of the most extensive varieties of tobacco plants. *Nicotiana tabacum* var. Virginia leaves known to contain alkaloids, saponins, tannins, phenol, flavonoids, triterpenoids and glycosides. In addition to cigarette raw materials, tobacco is also able to be efficacious as an antibacterial, antifungal and bioinsecticide. **Objective:** The aim of this study to determine the total phenol and total flavonoids and also antioxidant activity of Virginia tobacco leaves fractions. The fraction with the largest compound content was evaluated further for its antioxidant activity. **Methods:** Virginia tobacco leaves was fractioned into dichloromethane (DCM), ethyl acetate, butanol and water fractions. Phenol levels were determined with Follin-Ciocalteu reagent using the UV-Vis spectrophotometer method measured at 743.50 nm and gallic acid as a reference compound. Total flavonoid levels were determined with $AlCl_3$ reagent using the UV-Vis spectrophotometer method measured at 434.50 nm and quercetin as a reference compound. Antioxidant activity was evaluated with DPPH and Ferric Thiocyanate (FTC) method and the standard used was quercetin. **Results:** Total phenol levels in Virginia tobacco leaves of DCM, ethyl acetate, butanol and water fractions respectively were 191.2386 mgGAE/g, 201.2913 mgGAE/g, 180.5714 mgGAE/g, 212.8692 mgGAE/g. Total Flavonoid levels respectively were 6.0927 mgQE/g, 6.9659 mgQE/g, 5.1112 mgQE/g, 8.3346 mgQE/g. Antioxidant of water fraction was evaluated further using DPPH and FTC method with IC50 respectively were 75.9148µg/ml and 67.8972 µg/ml. **Conclusion:** Overview of total phenol and flavonoid levels fractions and antioxidant can be used as an additional initial reference for Virginia tobacco leaves development as source of medicinal substances.

Key words: Antioxidant, DPPH, Phenol, Flavonoid, FTC, Virginia tobacco.

INTRODUCTION

Indonesia is one of the countries in Asia with abundant of biodiversity. It has the largest tropical rainforest in the world with high humidity that allows the growth of various types of plants. The plant has been used for various purposes, especially for traditional remedies. The utilization of traditional remedies in Indonesia is part of the national culture and has been widely used for centuries ago. However, the effectiveness and safety have not been supported by adequate research.[1]

In improving the quality, safety and usefulness of traditional medicines, it is necessary to standardize raw materials in the production of traditional medicines including the standardization of plant extracts and fractions. The extraction quality standardization parameters consist of non-specific parameters (drying losses and specific gravity, the total ash content, moisture content, residual solvent and also pesticide, heavy metal contamination and the microbial contamination) and specific parameters (identity, organoleptic, dissolved compounds in certain solvents, phytochemical screening, total class of chemical content, specific chemical content.[2]

Virginia tobacco (*Nicotiana tabacum* var. Virginia) is one of the world's widest varieties of tobacco plants. This variant is planted around 1.8-2.0 million hectares. Recently, 1.5-2 million hectares of tobacco growing areas worldwide. Many researches has reported that *Nicotiana tabacum* var. Virginia contains more than 4000 chemical compounds.[3] Previous research reported that Virginia tobacco leaves contained triterpenoids, alkaloids, glycosides, saponins, flavonoids, tannins and phenols. In addition as raw materials for cigarettes, tobacco is also efficacious as an antibacterial,[4] antifungal and bioinsecticide.[5] Andjani *et al.* (2019). proved that, *Nicotiana tabacum* L. bio-oil at dosage 5000 mg/kg body weight was toxic against female winstar rats. There is no mortality and also no significant change in behavior and the body weight.[6]

The chemical coumpound of *Nicotiana tabacum* are phenol and flavonoid which are responsible for their health benefits.[7] According to multiple reports in the literature, phenolic and its derivate exhibit free radical inhibition, peroxide decomposition, metal inactivation or oxygen scavenging in biological systems and prevent oxidative disease burden.[8] Considering the important role and function of phenolic and flavonoid compounds, it is necessary to conduct research on analysis of phenolic and flavonoid levels of Virginia tobacco leaves (*Nicotiana tabacum* var. Virginia) and assesses the antioxidant using two different methods with a different mechanisms. So that the data in the chemical content of the group will be preliminary information about the characteristics of the extract associated with pharmacological effects.

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MATERIAL AND METHODS

Plant extract

Ethanol extract of Virginia tobacco leaves was obtained from the Agency for the Assessment and Application of Technology (BPPT), Indonesia extracted with reflux method.[3]

Chemicals and standards

Quercetin, gallic acid, Folin Ciocalteu and $AlCl_3$ were purchased from Merck and other chemical reagent for analytical analysis.

Physicochemical parameter evaluation of extract

Physicochemical parameters i.e. moisture content, total ash, water soluble ash content, acid insoluble ash content were evaluated according to the official methods described in Indonesian Pharmacopeia.[2]

Moisture content

Determination of drying loss according to Indonesian Pharmacopeia.[2]

Total Ash

The extract was put into the crucible silicate that has been equalized and tamed, incinerate slowly until the charcoal runs out, cool and weigh. If the charcoal cannot be removed, add hot water, stir, filter through ash-free filter paper. Filter paper along with the rest of the filtering the same crucible is incinerated to a fixed weight. Total ash content is calculated against the weight of the test material, expressed in% w/w.

$$Total\ ash\ value = \frac{W2 - W0}{W1 - W0}\ x\ 100$$

W0 = empty crucible weight

W1 = weight of crucible and extract before flattened

W2 = weight of crucible and dried extracts

Water-soluble ash content

Ash obtained from the determination of ash content boil with 25 ml of water for 5 minutes. Collect insoluble parts then filter with ash-free filter paper, wash with hot water, and incinerate for 15 minutes at a temperature of no more than 450 °C, until the weight remains then weigh.

$$Water\ soluble\ ash\ value = \frac{\text{total weight} - \text{Empty crucible weight}}{\text{Extract weight}}\ x\ 100$$

Acid insoluble ash content

Ash obtained from the determination of ash content, boiled with 25 ml dilute hydrochloric acid for 5 minutes, collect acid-insoluble parts, filtered through ash-free filter paper, then wash with water, incinerate to a fixed weight. Acid insoluble ash content was calculated against the weight of the test material, expressed in% w/w.

$$Acid\ insoluble\ ash\ value = \frac{\text{total weight} - \text{Empty crucible weight}}{\text{Extract weight}}\ x\ 100$$

Determination of phenolic level

The analysis was carried out according to the procedure performed by Lim & Murtijaya (2007) with a few modifications and gallic acid was used as a reference compound. 50.0 mg gallic acid was dissolved in 0.5 ml ethanol, then diluted with distilled water to a volume of 100.0 ml. 300 µL of gallic acid solution added 1.5 ml of the Folin-Ciocalteau reagent (1:10), then shaken and allowed to stand for 3 minutes. 1.2 ml of 7.5% Na_2CO_3 solution was added and shaken until homogeneous, and allowed to stand at room temperature in the operating time ra The absorbance of gallic acid was read at maximum wavelength. The series concen tion of gallic acid was made and recorded at maximum wavelength. The standard calibration curve was determined from the linear regression equatio etween series concentrations of gallic acid (x) and the absorbance of the gallic acid reaction with the Folin-Ciocalteu reagent (y). 10.0 mg of Virginia tobacco leaves fractions were dissolved to 10.0 ml with a mixture of methanol: distilled water (1: 1). 300 µl sample solution was allowed the procedure previously described, read at maximu velength and the procedure was carried out triplicate. Phenol levels of the samples obtained were expressed as equivalence to gallic acid (GAE).[9]

Determination of flavonoid level

analysis was carried out according to the procedure performed by Chang *et al.* (2002) with a few modifications used $AlCl_3$ and quercetin as used as a reference compound. The s dard solution was made at a concentration of 5 µg / ml, then added 3 ml of p.a ethanol, 0.2 ml of 10% $AlCl_3$, 0.2 ml of 1M sodium acetate, and aquadest to a volume of 10 ml. Absorbance was read at maximum wavelength. 100 mg Virginia tobacco leaves fractions were dissolved in a volumetric flask with 10 ethanol. 1 ml solution added with 3 ml ethanol, then added with 0.2 ml AlC 0% and 0.2 ml natrium asetat 1M reagent. The solution was added with distilled water up to 10 ml. The mixture was incubated at room temperature according to the operating time range. Absorbances were measured at the maximum wavelength and analysis was carried out in triplicate. Flavonoid content was obtained as equivalent to querc The equation of the standard calibration curve was obtained from linear regression between the concentration of quercetin (x) and absorbance (y). Flavonoid levels of the samples obtained were expressed as equivalence to quercetin (QE).[10]

Determination antioxidant activity

The fraction with a high level of phenolic and flavonoid content was analyzed further for its antioxidant activity with DPPH and FTC methods.

1,1-Diphenyl-2-Picrylhidrazil (DPPH) method

Virginia tobacco leaves fraction antioxidant capacity was determined according to a method performed by Molyneux (2004). 10 mg DPPH dissolved in 10 ml methanol. 2.0 ml of the stan d solution was pipetted then dissolved in a 100 ml volumetric flask. 0.2 ml of methanol mixed with 3.8 ml of DPPH then the maximum wavelength of DPPH solution was determined in a range of 400−800 nm. 10 mg of tobacco leaves fraction dissolved in 100 ml volu ric flask with methanol then prepared with various concentrations of 20 ppm, 40 ppm, 60 ppm, 80 ppm and 100 ppm. The absorbance of the sample was measured at λ_{max} for 30 minutes. The data obtained were processed to get the percentage inhibition value. Tests were carried out in triplicate.[11]

Ferric Thiocyanate (FTC) method

The capacity of Virginia tobacco leaves fraction to inhibit the peroxides formation from linoleic acid wa termined with the FTC methods described by Zahin *et al.* ( ). 1 ml of 50 mM linoleic acid in 100% ethanol was added with 1 ml of phosphate buffer pH 7 0.1 M, then incubated for 24 hours at 40 °C in a k room. The incubated solution was taken as much as 100 µL then added with 2.35 ml of 75% ethanol and 50 µL of ammonium thiocyanate 30%. Then add 50 µL $FeCl_3$ 0.02 M in 3.5% HCl. The maximum wavelength was determ d the range of 380-780 nm. The fraction was prepared with various concentrations (20, 4 , 80 and 100 ppm). The mixture was incubated according to the operating time range that has been determined before in a dark room temperature. Absorbances were measured at the maximum wavelength.[12]

RESULTS

Extraction and fractionation

Ethanol extract of Virginia tobacco leaves were obtained from the Agency for the Assessment and Application of Technology (BPPT) then fractioned with DCM, ethyl acetate, butanol and water solvents. All fractions then evaporated *in vacuo*. DCM, ethyl acetate, butanol and water fraction were analyzed further to determine phenolic, and flavonoid content.

Physicochemical evaluation

The non-specific parameters of the extract were determined including water content, total ash content, water-soluble ash content and acid insoluble ash content in the extract. Results are shown in Table 1.

Determination of phenolic content

Quantitative analysis of Phenolic levels was determined by the UV-Vis spectrophotometry method whose absorbance was measured at 743.50 nm. The phenolic level of Virginia tobacco leaves fractions is shown in table 2.

Determination of flavonoid content

Quantitative analysis of flavonoid levels was performed using UV-Vis spectrophotometry method. The maximum wavelength is obtained 443.50 nm. The flavonoid level of Virginia tobacco leaves fractions is shown in table 3.

Determination of antioxidant activity

DPPH method

Based on the analysis of phenolic and flavonoid content, the water fraction gave the highest value than the other fraction on both compounds analyzed. Therefore, the ability of water fractions to counteract free radicals was further evaluated by DPPH method. In this study, the maximum wavelength was obtained at 515 nm. The antioxidant activity of water fraction compared to the standard is shown in Table 4. Based on the research, antioxidant activity was increased according to water fraction concentration with the IC_{50} of 75.9148 µg/ml compared to quercetin was 6.7918 µg/ml. In this study, the results of quercetin gave a value that was close to the research of Rini *et al.* of 5.631 µg/ml.

Table 1: Physicochemical Evaluation of ethanol extract of Virginia tobacco.

Parameters	Result (%w/w)
Water content	16.43%
Total ash content	7.10 %
Acid insoluble ash content	0.36 %
Water-soluble ash content	3.47 %

Table 2: Phenolic content analysis from ethanol extract of Virgina tobacco.

Fractions	Absorbance	Phenolic Content (mgGAE/g)	Avarage (mgGAE/g)
DCM	0.661	194.3708	191.2386
	0.656	188.9604	
	0.666	190.3846	
Ethyl acetate	0.695	201.7234	201.2913
	0.697	200.4320	
	0.689	201.7185	
Buthanol	0.626	180.8988	180.5714
	0.632	184.6921	
	0.622	176.1234	
Water	0.729	210.0786	212.8692
	0.734	216.5896	
	0.724	211.2141	

Table 3: Flavonoid content analysis from ethanol extract of Virgina tobacco.

Fractions	Absorbance	Flavonoid content (mgQE/g)	Average (mgQE/g)
DCM	0.490	6.0897	6.0927 ± 0,089
	0.485	6.0043	
	0.487	6.1841	
Ethyl acetat	0.552	6.9187	6.9659 ±0,060
	0.554	6.9456	
	0.560	7.0335	
Buthanol	0.412	5.0281	5.1112 ±0,086
	0.417	5.1056	
	0.424	5.1999	
Water	0.653	8.2707	8.3346 ±0,088
	0.655	8.2976	
	0.664	8.4355	

Table 4: Antioxidant activity of water fraction with DPPH method.

Sample	Concentration (ppm)	Absorbance	%Inhibition	IC_{50} (µg/mL)
Quercetin	2	0.5248	22.6643%	
	4	0.4483	33.9357%	
	6	0.3658	46.0949%	6.7918µ/mL
	8	0.2946	56.4397%	
	10	0.2149	68.3318%	
Water fraction	20	0.5046	26.5822%	
	40	0.4304	37.3781%	
	60	0.3707	46.0643%	75.9148µ/mL
	80	0.3304	51.9278%	
	100	0.2915	67.5876%	

Table 5: Antioxidant activity of water fraction with FTC method.

Sample	Concentration (ppm)	Absorbance	%Inhibition	IC_{50} (µ/mL)
Quercetin	2	0.4331	35.5697%	
	4	0.3833	42.9782%	
	6	0.3296	50.9669%	6.0528µg/mL
	8	0.2866	57.3638%	
	10	0.2539	62.2285%	
Water fraction	20	0.4614	31.3597%	
	40	0.3925	41.6096%	
	60	0.3373	49.8215%	67.8972 µg/mL
	80	0.3060	54.4779%	
	100	0.2743	59.1936%	

FTC method

Furthermore, the capacity of water fraction in reducing the formation of peroxide in fat peroxidation was measured and the Ferric thiocyanate complex formed was read at 484 nm. The antioxidant activity of water fraction compared to the standard shown in Table 5. This study showed that water fraction Virginia tobacco leaves can prevent the formation of fat peroxidation from linoleic acid with IC_{50} was 67.8972 µg/ml compared to standard was 6.0528 µg/ml.

DISCUSSION

Virginia tobacco (*Nicotiana tabacum* var. Virginia) is one of a variety of tobacco that is mostly used as the main raw material for cigarettes

proven to be efficacious for many important pharmacological activities. The development of this variety for other purposes besides cigarettes is expected to increase its economic value. The present study, ethanol extract of Virginia tobacco leaves was obtained from the Agency for the Assessment and Application of Technology (BPPT), Indonesia which is extracted with the reflux method. the use of the extraction method is the main point that must be considered. The use of the reflux method provides several benefits to the compound extraction process. Hidayat *et al.* (2019) proved that the antioxidant activity possessed by black sumatran incense resin which is extracted using the reflux method is greater than *Jatropha curcas* L. and smaller than *Sargassum serratifolium* extract.[13]

Determination of physicochemical parameters of the extract is useful to determine the quality of extract as one of raw materials of traditional medicine. Water content is essential to provide a minimum limit or range of the amount of water content in the extract. Based on the result, the water content of the ethanol extract of Virginia tobacco meets the requirements of Indonesian Pharmacopoeia. Determination of total ash content gives an overview of extract mineral content, starting from the initial process to extract so that the total ash content parameters are related to the purity and contamination of an extract. The results showed that the level of minerals in an extract is still low.[2]

The chemical compound was one of the important parameters that related to the activity. In previous research ethanol extract of Virginia tobacco showed a positive result to the alkaloid, phenolic, flavonoid and saponin.[4] Phenolic and its derivatives are compounds that have been widely studied for various pharmacological activities that are important and beneficial. Many studies have reported the advantages of phenolic compounds, such as anti-aging, anti-inflammatory, antioxidant and anti-proliferative agents.[14] Phenolic compounds have received increasing interest in human health due to their beneficial effects against several diseases like cancers attributed in particular to their antioxidant activity.[15] *Nicotiana tabacum* L. var. Virginia leaf extract obtained from Ethanolic Heat Reflux Extraction (EHRE) had shown insecticidal activity against *Gryllus bimaculatus imago* and *Galleria mellonella* larvae. The values of LC_{50} were 38.5 mg/ml for *Gryllus bimaculatus* and 36.6 mg/ml for *Galleria mellonella*.[16]

Flavonoids are the largest group of phenolic compounds containing two aromatic rings linked by three atoms of carbon bridge (C_6–C_3–C_6). They include mainly flavones, isoflavones, flavonols, flavans, flavanones and anthocyanidins.[17] This compound is one of the polyphenol compounds which has the various property that is important for human health in inhibiting alpha-amylase enzyme which is a key enzyme that is responsible to digest carbohydrates in the digestive tract.[14] Besides that, the flavonoid is phenolic compounds that are highlighted for their antioxidant activity and the most widely distributed phenolic compounds in plant foods and also the most studied ones.[18]

Analysis of phenolic and flavonoid levels in the present study showed that ethanol extract of Virginia tobacco contained 153.7414 ± 0.281 mg GAE/g of phenolic compound and 3.5337 ± 0.1029 mg QE/g of flavonoid compound. It was an initial report that can be useful in the development of Virginia tobacco as a source of phenolic compound. Virginia tobacco (*Nicotiana tabacum* var Virginia) with high production capacity in various regions are more widely used to produce the white cigarette with high production capacity in various regions which may harm health. But this research indicated that Virginia tobacco still has pharmacological activities that are beneficial to our health. Interestingly, this plant can inhibit the oxidation process which can be caused by oxidants that can come from cigarette smoke.

CONCLUSION

Tobacco leaf is a plant that is widely found in Indonesia and is used as a commodity for export to various countries due to Indonesia has tobacco leaves which was one of the best qualities in the world. Virginia tobacco is one of the primary commodities in the production of cigarettes that provides a large income for the government but unfortunately, it harms human health. Determining the levels of phenols and flavonoids can be an opportunity in the search for sources of natural antioxidants. Besides that, analysis of phenol and flavonoid content from plant extracts can expand the utilization of Virginia tobacco plants for other uses as a source of traditional medicinal plants.

CONFLICTS OF INTEREST

All authors declare that they have no conflicts of interest.

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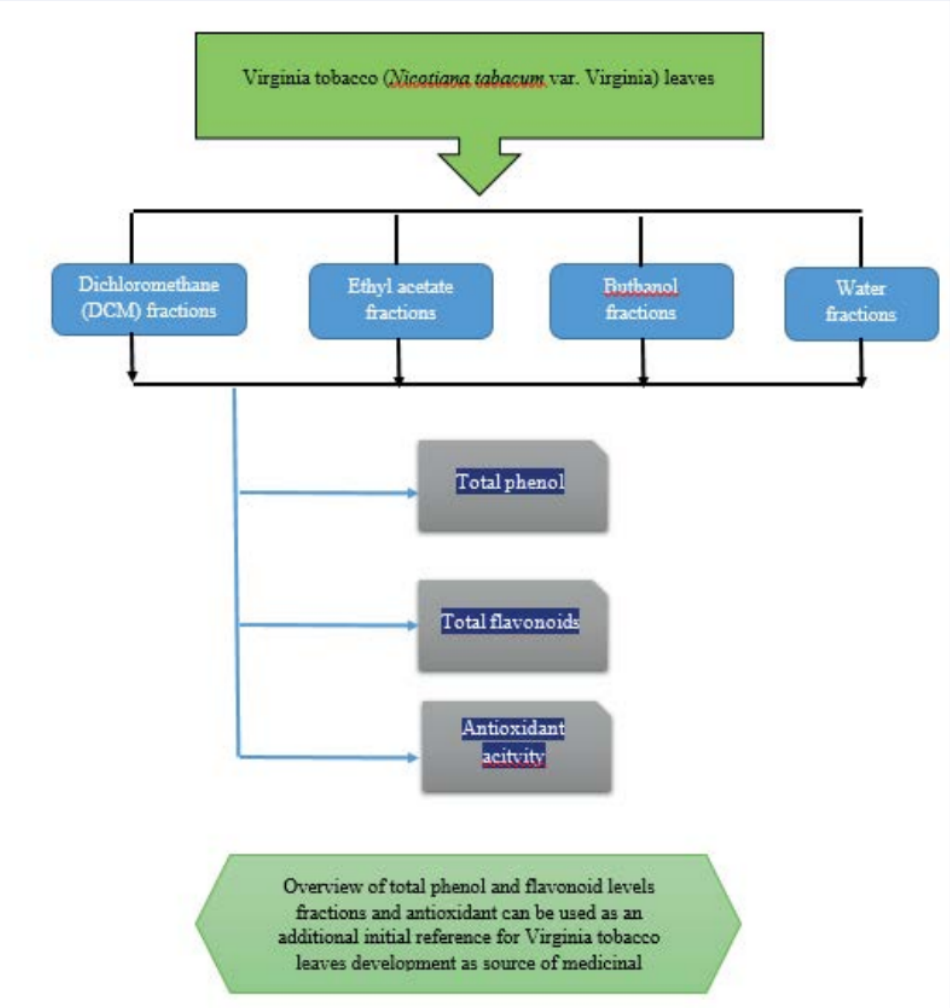
REFERENCES

1. Kementerian Kesehatan Republik Indonesia. Kebijakan obat tradisional nasional. Keputusan Menteri Kesehatan Republik Indonesia Nomor: 381/Menkes/SK/III/2007 Tanggal 27 Maret 2007. Jakarta: Departemen Kesehatan Republik Indonesia. 2007.
2. Kementerian Kesehatan RI. 2000. Parameter Standar Umum Ekstrak Tumbuhan Obat. Direktorat Jenderal Pengawasan Obat dan Makanan. Jakarta.
3. Fauzantoro A, Muharam Y, Gozan M. Improvement of Nicotine Yield by Ethanolic Heat Reflux Extraction of *Nicotiana tabacum* Var .Virginia Origin of Ponorogo. International Journal of Applied Engineering Research. 2017;12(23):13891-13897.
4. Yati K, Mahdi J, Misri G, Lusi PD. The Effect of Hidroxy Propyl Methyl Cellulose (HPMC) Concentration Variation on Physical Stability of Tobacco (*Nicotiana tabaccum* L.) Extract Gel and Its Activity Against Streptococcus mutans. Pharmaceutical Sciences and Research (PSR). 2018;5(3):133-141.
5. Pramono A, Fauzantoro A, Hidayati IR, Hygea A, Puspita OS, Muktamiroh H, *et al. In Vitro* Assay of Ethanolic Heat Reflux Extract of *Nicotiana tabacum* L. Var. Virginia Against Nosocomial Bacteria Pathogen. Journal of Physics: Conference Series. 2018;970(1).
6. Andjani HN, Sentosa Y, Yati K, Fauzantoro A, Gozana M, Yoo YJ. Acute Oral Toxicity Test of *Nicotiana tabacum* L. Bio-Oil Against Female Winstar Rats IOP Conf. Ser. Earth Environ Sci. 2019;353:012047.
7. Bocco A, Cuvelier ME, Richard H, Berset C. Antioxidant activity and phenolic composition of citrus peel and seed extracts. J Agric Food Chem. 1998;46:2123-2129.
8. Babbar N, Oberoi HS, Sandhu SK. Therapeutic and Nutraceutical Potential of Bioactive Compounds Extracted from Fruit Residues. Crit Rev Food Sci Nutr. 2015;55(3):319-337.
9. Lim YY, Murtijaya J. Antioxidant properties of *Phyllanthus amarus* extracts as affected by different drying methods. Swiss Society of Food Science and Technology. LWT. 2007;40:1664-1669.
10. Chang CC, Yang MH, Wen HM, Chern JC. Estimation of Total Flavonoid Content in Propolis by Two Complementary Colorimetric Methods. Journal of Food and Drug Analysis. 2002;10(3):178-182.
11. Molyneux P. The use of the stable free radical diphenylpicrylhydrazyl (DPPH) for estimating antioxidant activity. Songklanakarin J Sci Technol. 2004;26(2):211-219.
12. Zahin, Maryam, Aqil F, Ahmad I. The *in Vitro* Antioxidant Activity and Total Phenolic Content of Four Indian Medicinal Plants. International Journal of Pharmacy and Pharmaceutical Sciences. 2009;88-95.

13. Hidayat N, Yati K, Krisanti EA, Gozan M. Extraction and Antioxidant Activity Test of Black Sumatran Incense. The 4th Biomedical Engineering's Recent Progress in Biomaterials, Drugs Development, Health, and Medical Devices AIP Conf Proc. 2019;2193:030017-1-030017-6.
14. Lin D, Xiao M, Zhao J, Li Z, Xing B, Li X, *et al.* An Overview of Plant Phenolic Compounds and their Importance in Human Nutrition and Management of Type 2 Diabetes. Molecules. 2016;21(1374):1-19.
15. Basli A, Belkacem N, Amrani I. Health Benefits of Phenolic Compound Against Cancer. Intech. 2017;10.
16. Andjani HN, Sentosa Y, Yati K, Jufri M, Fauzantoro A, Gozan M. Determination of LC50 Value of *Nicotiana tabacum* L. Extract against *Gryllus bimaculatus* Imago and *Galleria mellonella* Larvae. The 4th Biomedical Engineering's Recent Progress in Biomaterials, Drugs Development, Health, and Medical Devices AIP Conf Proc. 2019;2193:1-7.
17. Giada MDLR. Food Phenolic Compounds: Main classes, source and Their Antioxidant Power. Intech. 2013;4.
18. Bravo L. Polyphenols: chemistry, dietary sources, metabolism and nutritional significance. Nutrition Reviews. 1998;56(11):317-333.
19. Prastiwi R, Elya B, Hanafi M, Sauriasari R, Desmiaty Y, Dewanti E, *et al.* The chemical constituents of *Sterculia comosa* (wall) Roxb woods for arginase inhibitory, antioxidant activity and molecular docking against SARS CoV-2 protein. 2022;8(2):1-19.

GRAPHICAL ABSTRACT



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