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## DETERMINATION OF 3,4 DIHYDROXY BENZOIC ACID FROM “BAWANG HUTAN” *Scorodocarpus borneensis* Becc. AS AN ANTIOXIDANT

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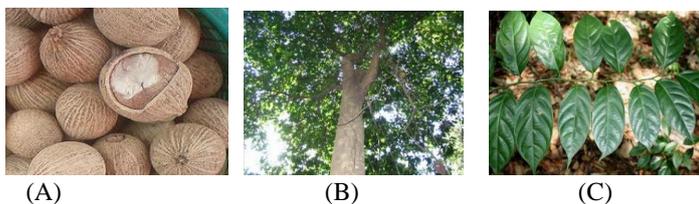
### ABSTRACT

Forest onion (Bawang hutan, local name), *Scorodocarpus borneensis* Becc is one of the medicinal ingredients considered to have properties that are useful as antioxidants, anticancer, antibacterial, antifungal, etc. This plant is widely found in the forest areas of Kalimantan, Indonesia. While its cultivation is still very lacking. In this research, the chopped bark of bawang hutan was refluxed with methanol solvent, then the methanol extract was partitioned to ethyl-acetate: water = 1:1. The ethyl acetate phase obtained was fractionated by column chromatography (SiO<sub>2</sub>; *i*). *n*-hexan : ethyl acetate = 20:1~2:1; *ii*). *n*-hexane-ethylacetate = 5: 1; and preparative thin layer chromatography (*n*-hexane-ethylacetate = 2:1). Each fraction was tested for antioxidant activity by the free radical scavenging method using 1,1-diphenyl-2-picrylhydrazyl (DPPH) and obtained pure isolates that have antioxidant activity in fraction SbEA 8-5. Based on the interpretation of Ultra-violet-visible (UV-Vis), Fourier Transform Infra Red (FT-IR) spectra, Nuclear Magnetic Resonance (NMR) 1D <sup>1</sup>H, <sup>13</sup>C-NMR, DEPT, and NMR 2D (HMQC, COSY, HMBC), the chemical compound is 3,5-dihydroxybenzoic acid (protocatechuic acid) which has an IC<sub>50</sub> of 12.05 ppm.

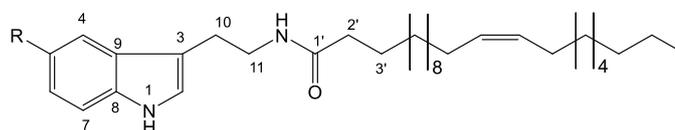
**Keywords:** Bawang hutan; 3,5 dihydroxybenzoic acid; protocatechuic acid; dihydroxy benzoic derivative compound; *Scorodocarpus borneensis* Becc, Olacaceae.

## INTRODUCTION

Indonesia is a country rich in natural resources. One of them is various types of plants which are very useful both in the food and health sectors. The plant can be used as a medicine in phytotherapy and is expected to be used in formal health services. One of the medicinal ingredients that is considered to have useful properties as medicines is "forest onion" (bawang hutan, local name), *Scorodocarpus borneensis* Becc is a type of wood tree that is the only member of the genus *Scorodocarpus*, in family Olacaceae. The name of bawang hutan is because the trunk, bark, and leaves emit a strong onion smell. Wood and its trunks, commonly called Kulim, are a type of tree that has the potential to be made into wooden door frames for houses and ships, especially walls, hatches, and ship masts. Now, the wood of bawang hutan is difficult to obtain due to massive illegal exploitation for the community, meanwhile, its cultivation is still very lacking.<sup>1</sup> The coarse extract of the bark of bawang hutan contains sesquiterpenes, saponins, steroids, flavonoids, and the compound methyl ditiliomethyl (methyl sulfonyl), methyl disulfide which causes a smell like garlic and has antioxidant, antimicrobial, antitoxicity and anticancer activities.<sup>2,3</sup> The research of Kartika *et al.*, (2014) reported the isolated compound of the fruit part in *n*-hexane extract gives dehydroxy scorodocarpine B (1) and scorodocarpine B (2) which have activity as antioxidants and anticancer leukemia.<sup>4</sup> In ethylacetate extract give cadalene-5- $\beta$ -carboxylic acid (3) and stigmaterol (4) which are cytotoxic to *Artemia salina* (BSLT, *Brine Shrimp Lethality Test*).<sup>5</sup> The purpose of this research is to determine the chemical structure of the dihydroxy benzoate derivative from ethyl acetate extract of stem bark bawang hutan, *Scorodocarpus borneensis* which has activity as an antioxidant.

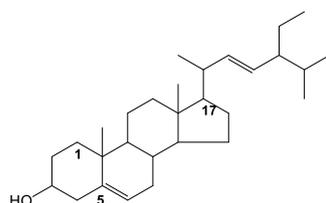
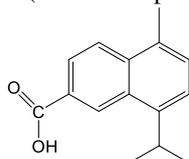


**Fig. 1.** Plant parts bawang hutan, *Scorodocarpus borneensis* Becc. the fruit (A). the bark (B). the leaves (C)



R = H (dehydroxy scorodocarpine B (1))

R = OH (Scorodocarpine B (2))



Cadalene-5β-carboxylic acid (3)

Stigmasterol (4)

**Figure 2.** Some chemical structures from the fruit of bawang hutan, collected from the forests of Samarinda, Indonesia

## EXPERIMENTAL

### Materials and Methods

The bark of bawang hutan, is collected in the forests of Samarinda, East Kalimantan, Indonesia. The identity of *Scorodocarpus borneensis* has been determined by the Research Center for Biology, Herbarium Bogoriense, Indonesian Institute of Sciences (LIPI), Bogor, Indonesia.

Methanol, ethyl acetate, 1,1-diphenyl-2-picrylhydrazil, distilled water, acetone, cerium sulfate, vitamin C, silica gel 60, silica gel GF<sub>254</sub>, KBr powder.

The instruments used in this research include chemical glassware, reflux apparatus, Ultra Violet – Visible spectrophotometer (Beckman DU 650), Fourier Transform Infra-Red (FT-IR) 8400S Shimadzu spectrophotometer), Nuclear Magnetic Resonance spectrometry (JEOL Nuclear Magnetic ECA 500 MHz), Mass Spectra (Liquid Chromatography-Mass Spectroscopy (Model-G6550 A of Agilent Tech.), rotavapour vacuum (Janke & Kunkel RV 05-ST), ultra-violet lamp.

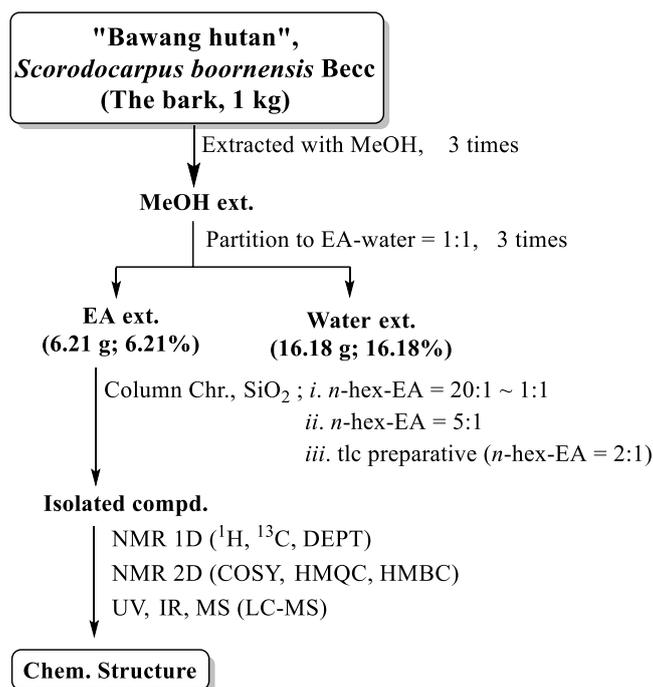
### General Procedure

#### Preparation of Plant Extract and Column Chromatography

One Kg of stem bark bawang hutan was extracted with methanol by reflux, and then partitioned with ethyl acetate-water (1:1). The ethyl acetate extract was subjected to column chromatography (SiO<sub>2</sub>; *i*). *n*-hexane: ethyl acetate = 20:1 ~ 2:1; *ii*). *n*-hexane-ethylacetate = 5 : 1; and preparative thin layer chromatography (*n*-hexane-ethylacetate = 2 :1).

#### Antioxidant Assay

The antioxidant activity of the ethyl acetate extract, the fractions of column chromatography, and the pure compound was determined by 1,1-Diphenyl-2-Picrylhydrazyl (DPPH) method according to Madhuranga & Samarakoon (2023) with small modifications.<sup>6</sup>



**Fig. 3.** The scheme for isolation of 3,4-dihydroxy benzoic acid (protocatechuic acid, PCA)

## RESULTS AND DISCUSSION

The results of the extraction of 1 kg of the stem bark, bawang hutan that have been partitioned to ethyl acetate and water gave 6.21 g (0.62%) of ethyl-acetate extract and 16.18 g (0.16%) of water extract.

### The Antioxidant Assay

The results of the antioxidant assay based on the free radical scavenging activity method of ethyl acetate extract and column chromatography fractions can be shown in Table 1.

**Table 1.** The antioxidant activity of ethylacetate and some fractions of column chromatography

No	Sample	IC <sub>50</sub> (ppm)	No	Sample	IC <sub>50</sub> (ppm)
1	SBEA-1	439.52	10	SBEA 8-1	92.15
2	SBEA-2	335.67	11	SBEA 8-2	80.45
3	SBEA-3	228.82	12	SBEA 8-3	81.23
4	SBEA-4	293.30	13	SBEA 8-4	82.98
5	SBEA-5	87.60	14	SBEA 8-5	<b>31.23</b>
6	SBEA-6	143.87	15	SBEA 8-6	85.73
7	SBEA-7	72.35	16	Pure compd. SBEA-851	<b>12.05</b>
8	SBEA-8	<b>52.42</b>	17	EA extract	134.95
9	SBEA-9	106.79	18	Vit. C	<b>8.73</b>

SBE : *Scorodocarpus borneensis*; EA : Ethyl acetate

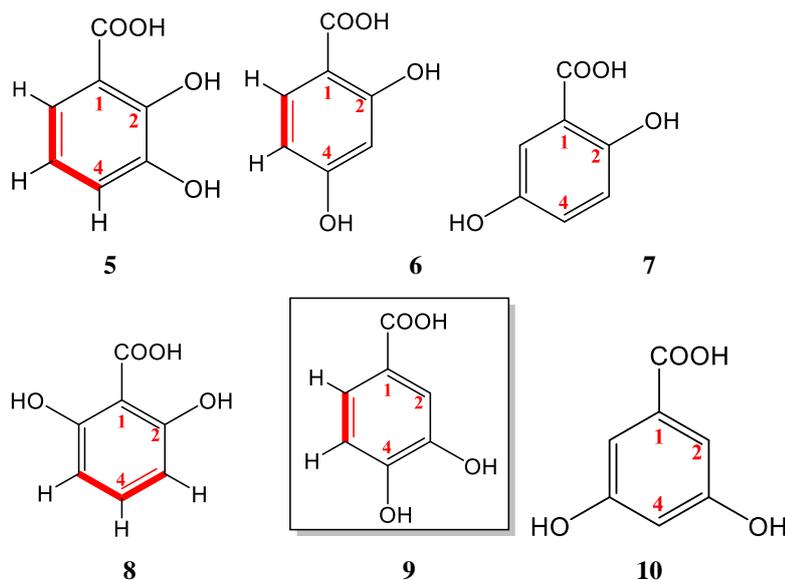
The antioxidant activities showed that ethyl acetate extract, SBEA-8 fraction, SBEA 8-5, and pure compound, IC<sub>50</sub> are 52.42; 31.23, and 12.05 ppm respectively, while vitamin C as standard, IC<sub>50</sub> is 8.73 ppm.

### Determination chemical structure of isolated compounds

The white powder of the isolate showed the maximum UV absorbance at  $\lambda$  259 and 294 nm. The Infrared (IR) absorption band at wave number (cm<sup>-1</sup>) is 3238.26; 3668.06 (hydroxyl groups) and 1676.03 (carbonyl group).

An investigation based on the <sup>1</sup>H-NMR spectrum showed that the chemical shift in the low-field area at  $\delta$ H 7.52 (H-2); 6.90 (H-5) and 7.47 (H-6). Investigation based on <sup>13</sup>C-NMR and DEPT (Distortionless Enhancement by Polarization Transfer) spectra showed that there were seven carbon atoms consisting of 3 methine groups at  $\delta$ C 117.48 (C-2); 115.75 (C-5); 123.15 (C-6) and four quaternary carbon at  $\delta$ C 123.67 (C-1); 145.53 (C-3); 150.74 (C-4); and 167.61 (C=O). The mass spectra (MS) analysis (by LC-MS spectra) gave  $m/z$  154.01 (M)<sup>+</sup> for the C<sub>7</sub>H<sub>6</sub>O<sub>4</sub> molecule.

In the interpretation of UV, IR,  $^1\text{H}$ -,  $^{13}\text{C}$ -NMR, and mass spectra, six possible chemical structures that have similarities from the group of dihydroxy-derived compounds of benzoic acid<sup>7</sup>. (Fig. 4)



— : H-H COSY

2,3-dihydroxybenzoic acid (Pyrocatechuic acid, **5**)

2,4-dihydroxybenzoic acid ( $\beta$ -resorcylic acid, **6**)

2,5-dihydroxybenzoic acid (Gentisic acid, **7**)

2,6-dihydroxybenzoic acid ( $\gamma$ -resorcylic acid, **8**)

3,4-dihydroxybenzoic acid ((protocatechuic acid, **9**)

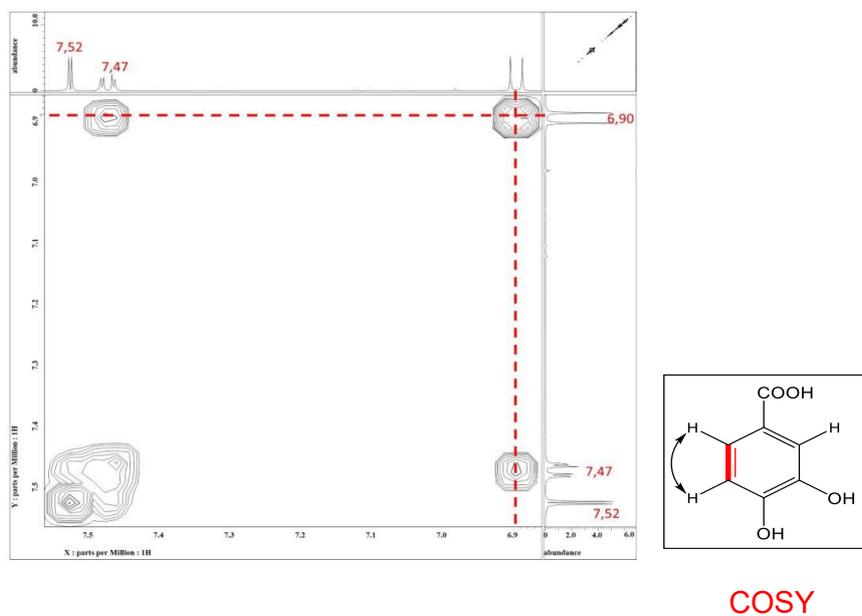
3,5-dihydroxybenzoic acid ( $\alpha$ -resorcylic acid, **10**)

**Fig. 4.** Possible Chemical structure of benzoate dihydroxy derivatives (differentiated by H-H COSY correlation)

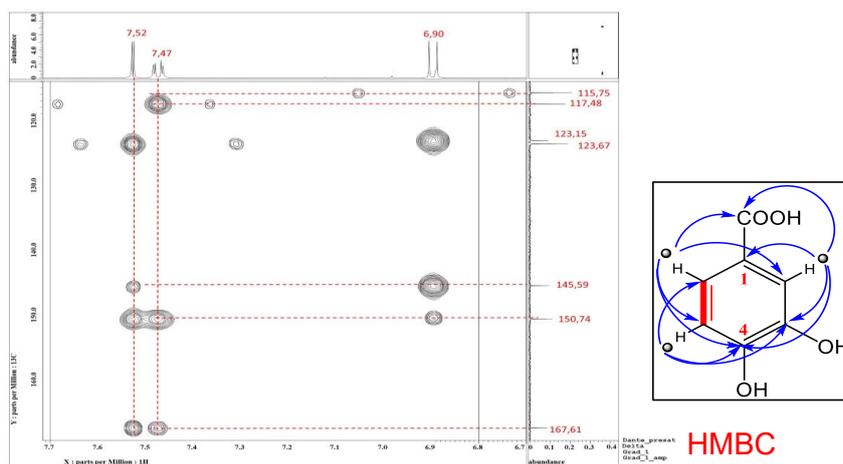
To ascertain the actual chemical structure, it can be analyzed by 2-dimensional NMR such as COSY (*Correlation Spectroscopy*) and HMBC (*Hetero Multiple Bond Connectivity*). COSY analysis showed that  $\delta\text{H}$  6.90 (H-5) only correlated to  $\delta\text{H}$  7.47 (H-6). Based on this COSY correlation, it provides two possible chemical structures of compounds **6** and **9**.

Based on the HMBC analysis shows that there is a proton and carbon relationship that is more than one bond. The proton signal at  $\delta\text{H}$  6.90 (H-5) correlates with carbon at  $\delta\text{C}$  123.15 (C-6); 123.67 (C-1); 145.59 (C-3); and 150.74

(C-4). Proton signal at  $\delta$ H 7.47 (H-6) with  $\delta$ C (117.48 (C-2); 117.48 (C-6); 150.74 (C-4); 167.61 (C=O)); and protons at  $\delta$ H 7.52 (H-2) with  $\delta$ C 123.15 (C-6); 123.67 (C-1); 145.59 (C-3); 150.74 (C-4); 167.61 (C=O).



**Fig. 5.** COSY correlation for **9**



**Fig. 6.** HMBC correlation for **9**

It is clear that the chemical structure of 3,4 dihydroxy benzoate acid (Protocatechuic acid, PCA, **9**) is an isolated compound from the bark of the stem of bark (*Scorodocarpus borneensis*). The comparison of the chemical shifts ( $^1\text{H}$  and  $^{13}\text{C}$ -NMR) can be seen in Table 2.

**Table 2.** Comparison of chemical shifts of compound 9 with isolated by Gurial *et al* 2013.<sup>8</sup>

No	$^{13}\text{C}$ -NMR ( $\text{CDCl}_3$ , 125 MHz, DEPT) ( <b>Compd. 9</b> )	$^{13}\text{C}$ -NMR (DMSO-d6, 75 MHz) <b>Gurial, et al. 2013</b>	$^1\text{H}$ -NMR ( $\text{CDCl}_3$ , 500 MHz, <i>J</i> in Hz) ( <b>Compd. 9</b> )	$^1\text{H}$ -NMR (DMSO-d6, 300 MHz). <b>Gurial, et al. 2013</b>
1	123.67 (s)	122.3 (s)	-	-
2	117.48 (d)	116.9 (d)	7.52 (d, <i>J</i> =2.0)	7.36 (d, <i>J</i> =2.1)
3	145.53 (s)	145.2 (s)	-	-
4	150.74 (s)	150.3 (s)	-	-
5	115.75 (d)	115.5 (d)	6.90 (d, <i>J</i> =8.5)	6.80 (d, <i>J</i> =8.1)
6	123.15 (d)	122.0 (d)	7.47 (dd, <i>J</i> =2.0;8.5)	7.31 (dd, <i>J</i> =2.1; 8.1;
7	167.61 (s)	167.7 (s)	-	-

3,4-dihydroxy benzoic acid (Protocatechuic acid, PCA, **9**) is the first compound to be isolated from bawang hutan, *Scorodocarpus boornensis* Becc). Song J. *et al.*, 2020 reported that protocatechuic acid, PCA displays extensive pharmacological properties that may act as antioxidant, anti-inflammatory, neuroprotective, antitumor, antibacterial, antidiabetic, and antiapoptotic agents. PCA may also be valuable in the prevention and treatment of cancer, diabetes, Alzheimer's disease (AD), atherosclerosis, and other diseases.<sup>9</sup>

## REFERENCES

1. LSL. Chua, "Scorodocarpus" Becc. In : MSM. Siseff, LT. Hong and S. Prawirohatmodjo (Eds.). Plant Resources of South-East Asia (Prosea) no 5 (3), timber tree: Lesser-known timbers : 1998. 514-6. Bogor Prosea Foundation
2. C. Wiart, A.A. Shorna, M. Rahmatullah, V. Nissapatorn, J.S.S. Seelan, H. Rahman, N.A.Rusdi, N. Mustafa, L. Elbehairy, and M. Sulaiman. The Garlic Tree of Borneo, *Scorodocarpus borneensis* (Baill.) Becc. (Olacaceae): Potential Utilization in Pharmaceutical, Nutraceutical, and Functional Cosmetic Industries. . *Molecules* 2023, 28, 5717. <https://doi.org/10.3390/molecules28155717>

3. K. Kubota, Kobayashi, A., Sulfur Compounds in Wood Garlic (*Scorodocarpus boornensis* Becc) As Versatile Food Components. In Sulfur Compounds in Foods; ACS Symposium Series, No 564; American Chemical Society: Washington, DC, USA, 1994, p. 238-246. AGR: [IND20507205](#)
4. R. Kartika, T. Barus, R. Surbakti, P. Simanjuntak, Structure Characterization of Alkaloid Scorodocarpine Derivative from fruits of *Scorodocarpus boornensis* Becc. (Olacaceae). Asian J. Chem. 2014, 26, 6047 – 6049. <http://dx.doi.org/10.14233/ajchem.2014.16562>
5. R. Kartika, B. Bustanussalam, P. Simanjuntak, Identification of Cadalena-b-carboxylic Acid from Barks of Bawang Hutan (*Scorodocarpus boornensis* Becc). Annales Bogor.2012. 16. 19-22. ISBN/ISSN 01578452
6. H.D.T. Madhuranga and D.N.A.W. Samarakoon. Advancing In vitro Antioxidant Activity Assessment: A Comprehensive Methodological Review and Improved Approaches for DPPH, FRAP and H<sub>2</sub>O<sub>2</sub> Assays. J. of Nat. and Ayurvedic Medicine vol. 7 (4), 2023. 1-7. DOI: [10.23880/jonam-16000431](https://doi.org/10.23880/jonam-16000431)
7. B.H.J. Juurlink, H.J Azouz, A.MZ Aldalati, B.M.H. AlTinawi and P. Ganguly. Hydroxybenzoic acid isomers and the cardiovascular system. A review. Nutrition Journal 2014, 13:63 <http://www.nutritionj.com/content/13/1/6>
8. M. Gurial, P. Mitra, T. Ghosh, S. Gupta, B. Basu, P. K. Mitra. 3, 4 dihydroxybenzoic acid Isolated from the Leaves of *Ageratum conyzoides* L. European Journal of Biotechnology and Bioscience 2013;1 (4): 25-28. ISSN 2320-7078 EJBB 2013; 1 (1): 25-28
9. J. Songa, Y. Hea, C.Luo, B. Fenga, F. Rana, H.X. Xub, Z. Cia, R. Xua, L. Hana, D. Zhanga. 2020. New progress in the pharmacology of protocatechuic acid: A compound ingested in daily foods and herbs frequently and heavily. Pharmacological Research Vol. 161, 2020, 105109. <https://doi.org/10.1016/j.phrs.2020.105109>