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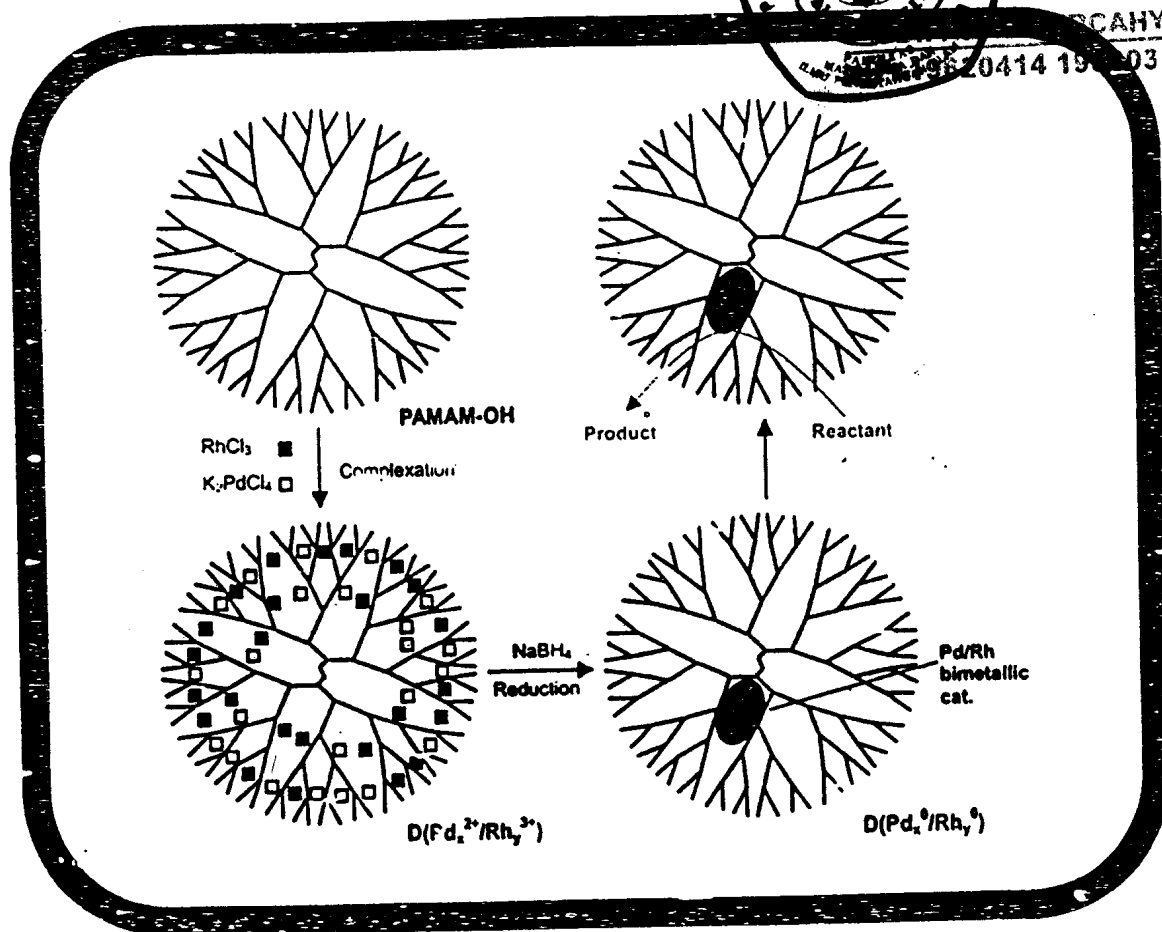
ISSN 1411-9420

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# Indonesian Journal of Chemistry

Vol. 6, No. 1, March, 2006

PENGESAHAN  
TELAH DIPERIKSA KEBENARANYA  
DAN SESUAI DENGAN ASLINYA



Published by  
Department of Chemistry, Gadjah Mada University  
Yogyakarta, Indonesia

Indo. J. Chem.	Vol. 6	No. 1	PP 1 - 110	Yogyakarta March 2006	ISSN 1411-942
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Indonesian Journal of Chemistry (ISSN 1411-9420) is published three times annually, in March, July and November by the Department of Chemistry, Faculty of Mathematics and Natural Sciences, Gadjah Mada University, Sekip Utara, Yogyakarta, Indonesia. All orders accompanied by payment should be sent directly to The department of Chemistry, Gadjah Mada University. Annual subscription rate: Rp 125.000,00 (including postages paid). Customers may make payments by transfer on BNI 46 Gadjah Mada University Billing (Iqmal Tahir cq IJC, Account Nr. 38422282) or by cheque.

## CONTENTS

<b>Review: Preparations And Application of Metal Nanoparticles</b> By Adlim	1 - 10
<b>Synthesis of Zeolite Beta Directly from Rice Husk Ash: Effect of Reaction Composition on Crystallinity of Zeolite Beta</b> by Didik Prasetyoko, Zainab Ramli, Salasiah Endud, Halimatun Hamdan and Bogdan Sulikowski	11 - 15
<b>Growth Rate Distribution of Borax Single Crystals on The (001) Face under Various Flow Rates</b> by Suharso	16 - 19
<b>Radiation Grafting of Styrene onto PTFE Film</b> ( <i>Pencangkakan Radiasi Stirena pada Film PTFE</i> ) by Yohan, Rifaid M. Nur, Lilik Hendrajaya and E. S. Siradj	20 - 26
<b>The Effect of PVC-Based Membrane Composition and Zn(II), Cd(II) and Pb(II) Interfering Ions To Hg(II) Ion Selective Electrode (ISE) Performance By Using DBA218C6 Ionophore</b> ( <i>Pengaruh Komposisi Membran Berbasis PVC dan Ion Pengganggu Zn(II), Cd(II) dan Pb(II) terhadap Kinerja Elektroda Selektif Ion (ESI)-Hg(II) Menggunakan Ionofor DBA218C6</i> ) by Abd. Wahid Wahab	27 - 31
<b>Photodegradation of Alizarin S Dye using TiO<sub>2</sub>-Zeolite and UV Radiation</b> ( <i>Fotodegradasi Zat Warna Alizarin S Menggunakan TiO<sub>2</sub>-Zeolit dan Sinar UV</i> ) by Karna Wijaya, Eko Sugiharto, Is Fatimah, Iqmal Tahir and Rudatiningsih	32 - 37
<b>Titanium Oxide Dispersed on Natural Zeolite (TiO<sub>2</sub>/Zeolite) and Its Application for Congo Red Photodegradation</b> ( <i>Titan Dioksida Terdispersi pada Zeolit Alam (TiO<sub>2</sub>/Zeolit) dan Aplikasinya Untuk Fotodegradasi Congo Red</i> ) by Is Fatimah, Eko Sugiharto, Karna Wijaya, Iqmal Tahir and Kamalia	38 - 42
<b>The Influence of pH towards Multiple Metal Ion Adsorption of Cu(II), Zn(II), Mn(II), and Fe(II) on Humic Acid</b> ( <i>Pengaruh Ph terhadap Adsorpsi Ion Logam Gabungan Cu(II), Zn(II), Mn(II) dan Fe(II) pada Asam Humat</i> ) by Buhani and Suharso	43 - 46
<b>Sorption-Desorption Mechanism of Zn(II) and Cd(II) on Chitin</b> by Sri Juari Santosa, Narsito and Aldes Lesbani	47 - 51
<b>Adsorption Thermodynamics of Cd(II), Ni(II), and Mg(II) on 3-Mercapto-1,2,4-Triazole Immobilized Silica Gel</b> ( <i>Termodinamika Adsorpsi Cd(II), Ni(II) Dan Mg(II) Pada Silika Gel Yang Terimobilisasi Dengan 3-Merkapto-1,2,4-Triazol</i> ) by Fahmiati, Nuryono and Narsito	52 - 55
<b>Determination of pH Effect and Capacity of Heavy Metals Adsorption by Water Hyacinth (<i>Eichhornia crassipes</i>) Biomass</b> ( <i>Pengaruh Ph Dan Penentuan Kapasitas Adsorpsi Logam Berat Pada Biomassa Eceng Gondok (Eichhornia Crassipes)</i> ) by Anis Shofiyani and Gusrizal	56 - 60
<b>Bioaccumulation Dynamics of Heavy Metals in <i>Oreochromis niloticus</i>: Predicted through A Bioaccumulation Model Constructed Based on Biotic Ligand Model (BLM)</b> by Sri Noegrohati	61 - 69
<b>Callus Induction and Phytochemical Characterization of <i>Cannabis sativa</i> Cell Suspension Cultures</b> by Tri J. Raharjo, Ogbuadike Eucharia, Wen-Te Chang and Robert Verpoorte	70 - 74
<b>Balanocarpol and Heimiol A, Two Resveratrols Dimer from Stem Bark <i>Hopea mengarawan</i> (<i>Dipterocarpaceae</i>)</b> by Sri Atun, Nurfina Aznam, Retno Arianingrum and Masatake Niwa	75 - 78
<b>(-)-Ampelopsin A : Dimer Resveratrol from <i>Dryobalanops oblongifolia</i> (<i>dipterocarpaceae</i>)</b> by Nanik Siti Aminah, Sjamsul Arifin Achmad, Masatake Niwa, Yana Maolana Syah and Euis Holisotan Hakim	79 - 81
<b>Non Phenolic Compounds Isolated from The Leaves of Fern <i>Chingia sakayensis</i> (Zeiller) Holtt</b> by Suyatno, Noor Cholies Zaini, Gunawan Indrayanto and Motoo Tori	82 - 84

# BALANOCARPOL AND HEIMIOL A, TWO RESVERATROLS DIMER FROM STEM BARK *Hopea mengarawan* (Dipterocarpaceae)

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Received 2 January 2006; Accepted 16 January 2006

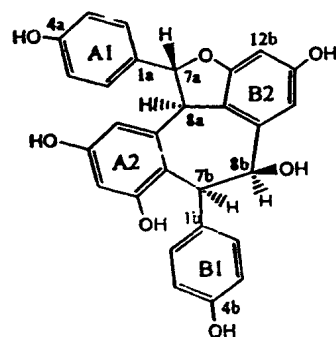
## ABSTRACT

Isolation and structure elucidation of two resveratrols dimer, namely balanocarpol (1) and heimiol A (2) from stem bark of *Hopea mengarawan* had been done. The isolation of those compounds was carried out by chromatographic method and structure elucidation was performed by interpretation of spectroscopic data, including UV, IR, <sup>1</sup>H and <sup>13</sup>C NMR 1D and 2D, and FABMS.

**Keywords:** Balanocarpol; Heimiol A; Dipterocarpaceae.

## INTRODUCTION

*Hopea* is one the main genus of Dipterocarpaceae, consisting of approximately 100 species and widely distributed in Indonesia specially in Kalimantan [1,2] and until now only few species have been investigated. This family of plant is known to produce a variety of resveratrol oligomer [3-16]. These structures are very interesting and showed interesting biological activity, such as antibacterial, anticancer, antihepatotoxic, and anti-HIV [3-16]. Thus Dipterocarpaceae plants are very potential for chemical research in natural product and pharmaceutical industry. In our continuing phytochemical study of the Dipterocarpaceae family occurring in Indonesia, we have examined resveratrol oligomer constituents of *H. mengarawan* Miq. This plant is widely distributed in tropical rain forest of Sumatra, Malaysia; until Andaman islands, and it is locally known as "merawan hitam" or "pengarawan" [3]. This paper will report our first investigation of two resveratrols dimer from stem bark of *H. mengarawan*, namely balanocarpol (1) and heimiol A (2). The structure of this compound based on the analysis spectrum of UV, IR, MS and NMR included 1D and 2D NMR (<sup>1</sup>H-<sup>1</sup>H COSY, HMQC, HMBC and NOESY).



Balanocarpol (1)

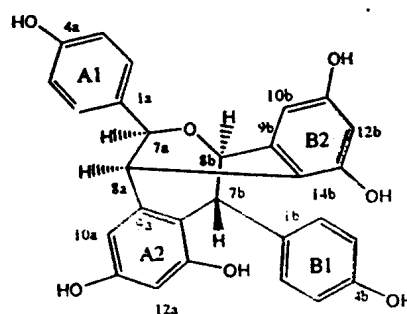
## EXPERIMENTAL SECTION

### General Experimental Procedure

UV and IR spectra were measured with varian Cary 100 Conc and Shimadzu 8300 FTIR respectively. <sup>1</sup>H and <sup>13</sup>C NMR spectra were recorded with Jeol JNM A-5000 spectrophotometers, operating at 600.0 MHz (<sup>1</sup>H) and 150.0 MHz (<sup>13</sup>C) using residual and deuterated solvent peaks as internal standards. MS spectra were obtained with a JMS-AM 20 spectrometer, using the mode FAB. Vacuum liquid chromatography (VLC) was carried out using Merck Si gel Merk 60 GF<sub>254</sub> (230-400 mesh), column chromatography using Si-gel Merk 60 (200-400 mesh), and TLC analysis on precoated Si gel plates Si-gel Merk Kieselgel 60 F<sub>254</sub> 0.25 mm, 20 x 20 cm.

### Plant Material

Samples of the stem bark of *H. mengarawan* were collected in Desember 2003 from the Experimental Garden in Carita, Banten, Indonesia. The plant was identified by the staff at the Herbarium Bogoriense, Kebun Raya Bogor, Bogor, and a voucher specimen had been deposited at the Herbarium.



Heimiol A (2)

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Table 1 <sup>1</sup>H and <sup>13</sup>C NMR data of compound 1 in acetone-d<sub>6</sub>

No	δ H (mult., J in Hz)	δ C	HMBC (H → C)
1a	-	133.7	-
2a,6a	7.48 (d, 8.8)	131.5	C-1a; C-4a; C-7a; C-3a
3a,5a	6.95 (d, 8.8)	116.4	C-1a; C-4a;
4a	-	159.2	-
7a	5.70 (d, 9.5)	93.5	C-1a; C-2a; C-9a; C-8a; C-11b;
8a	5.16 (d, 9.5)	52.5	C-9a; C-11b; C-10b; C-1a; C-7a; C-14a; C-10a
9a	-	142.8	-
10a	-	120.5	-
11a	-	157.4	-
12a	6.09 (d, 2.2)	102.0	C-11a; C-10a; C-14a; C-13a
13a	-	156.9	-
14a	5.96 (d, 2.2)	106.8	C-13a; C-12a; C-10a;
1b	-	133.4	-
2b,6b	6.75 (d, 9.5)	132.0	C-1b; C-4b; C-3b
3b,5b	6.42 (d, 9.5)	114.1	C-4b; C-1b;
4b	-	155.8	-
7b	4.89 (br. s)	50.2	C-1b; C-2b; C-8b; C-11a; C-10a; C-10b; C-9b;
8b	5.39 (br s)	73.2	C-7b; C-10a; C-9b; C-10b; C-9b; C-10b; C-7b
OH	4.32 (d, 4.4)	-	-
9b	-	140.8	-
10b	-	113.9	-
11b	-	159.2	-
12b	6.20 (d, 2.2)	95.1	C-11b; C-10b; C-13b; C-14b
13b	-	159.7	-
14b	6.25 (d, 2.2)	104.5	C-12b; C-13b; C-10b

Table 2 <sup>1</sup>H and <sup>13</sup>C NMR data of compound 2 in acetone-d<sub>6</sub>

No	δ H (mult., J in Hz)	δ C	HMBC (H → C)
1a	-	136.8	-
2a,6a	6.90 (d, 8.4)	127.9	C-7a
3a,5a	6.69 (d, 8.4)	115.3	C-7a; C-1a
4a	-	157.2	-
7a	5.57 (br s)	81.5	C-8a; C-1a; C-2a; C-9a
8a	4.24 (br. s)	46.9	C-7a; C-14a; C-10a; C-9b; C-9a; C-13b
9a	-	147.4	-
10a	6.41 (d, 2.6)	107.4	C-12a
11a	-	157.1	-
12a	6.16 (d, 2.6)	102.0	C-11a; C-10a; C-13a
13a	-	154.6	-
14a	-	116.0	-
1b	-	136.9	-
2b,6b	7.14 (d, 8.4)	130.0	C-7b; C-4b
3b,5b	6.72 (d, 8.4)	115.5	C-1b; C-4b
4b	-	157.2	-
7b	4.32 (d, 3.3)	50.9	C-8b; C-14a; C-2b; C-1b; C-9a; C-13a
8b	4.97 (d, 3.3)	81.4	C-7a; C-14b; C-10b; C-9b
9b	-	142.6	-
10b	6.48 (d, 2.2)	104.8	C-8b; C-12b
11b	-	158.1	-
12b	6.21 (d, 2.2)	102.1	C-14b; C-10b; C-11b
13b	-	156.2	-
14b	-	117.0	-

### Extraction and Isolation

The milled dried stem bark of *H. mengarawan* (5 kg) was extracted exhaustively with acetone. The acetone extract on removal of the solvent under reduced pressure gave a brown residue (400 g). A portion (40 g) was then subjected to fractionated by VLC (silica gel GF 60 Merk 250 g; φ: 10 cm, t = 10 cm), using n-hexane, n-hexane-EtOAc, EtOAc, Me<sub>2</sub>CO, and MeOH of increasing polarity as eluents to give twenty fractions. These fractions were combined to give two major fractions A (31.8 g) and B (7.05 g). Fraction A (31.8 g) was repeatedly separated and purified by column chromatography. From this method we obtained two resveratrols dimer, nameiy balanocarpol (1) (300 mg) and heimiol A (2) (200 mg).

Balanocarpol (1) was obtained as a pale yellow powder, m.p. 230 °C, UV (MeOH) λ<sub>max</sub> (log ε): 227 (5.6); 283 (3.76) nm, IR (KBr) ν<sub>max</sub>: 3384; 1608; 1405; 1350; 1240; 1132; 1037; 995; 833 cm<sup>-1</sup>, <sup>1</sup>H and <sup>13</sup>C NMR (Me<sub>2</sub>CO-d<sub>6</sub>, 600.0 and 150 MHz) see Table 1. FABMS m/z 470 [M<sup>+</sup>] (C<sub>28</sub>H<sub>22</sub>O<sub>7</sub>).

Heimiol A (2) was obtained as a pale yellow powder, m.p. 240 °C, UV (MeOH) λ<sub>max</sub> (log ε): 225 (6.01); 230 (sh 4.83); 282 (3.65) nm, IR (KBr) ν<sub>max</sub>: 3352; 1606; 1512; 1450; 1234; 1141; 1068; 954; 835 cm<sup>-1</sup>, <sup>1</sup>H and <sup>13</sup>C NMR (Me<sub>2</sub>CO-d<sub>6</sub>, 600.0 and 150 MHz) see Table 2. FABMS m/z 471 [M+H]<sup>+</sup> (C<sub>28</sub>H<sub>22</sub>O<sub>7</sub>).

### DISCUSSION

From the acetone extract non polar fraction of stem bark *H. mengarawan*, after separated and repeatedly purification by extensive chromatography resulted two compounds. Balanocarpol (1) was obtained as a pale yellow powder, m.p. 230 °C. Its UV spectrum showed absorption maxima at 283 nm suggesting the presence of unconjugated phenolic chromophore. The IR spectrum exhibited hydroxyl group (3384 cm<sup>-1</sup>), C=C aromatic (1608; 1405; 1350 cm<sup>-1</sup>), and monosubstituen benzene (833 cm<sup>-1</sup>), these spectra characteristic absorptions for supporting 1 to be an oligoresveratrol. The positive ion FABMS exhibited an [M]<sup>+</sup> ion at m/z 470 consistent with a molecular formula C<sub>28</sub>H<sub>22</sub>O<sub>7</sub> for a resveratrol dimer and supported by the NMR data. <sup>13</sup>C NMR spectra showed six signals for oxyaryl carbon at 159.2 (C-4a), 157.4 (C-11a), 154.6 (C-13a), 157.2 (C-4b), 159.2 (C-11b), and 159.7 (C-13b) ppm, characteristics for resveratrol dimer. Additionally, the <sup>13</sup>C NMR also exhibited one oxyalkyl carbon at 73.2 (C-8b) indicating that C-8b attached with hydroxyl functional group. The <sup>1</sup>H NMR spectrum of 1 in acetone-d<sub>6</sub> exhibited signals for two sets of 4-hydroxybenzene at 7.48 (d, J = 8.8 Hz) and 6.95 (d, J = 8.8 Hz) ppm, each 2H (ring A1) and at δ 6.75 (d, J = 9.5 Hz) and 6.42 (d, J = 9.5 Hz) ppm, each

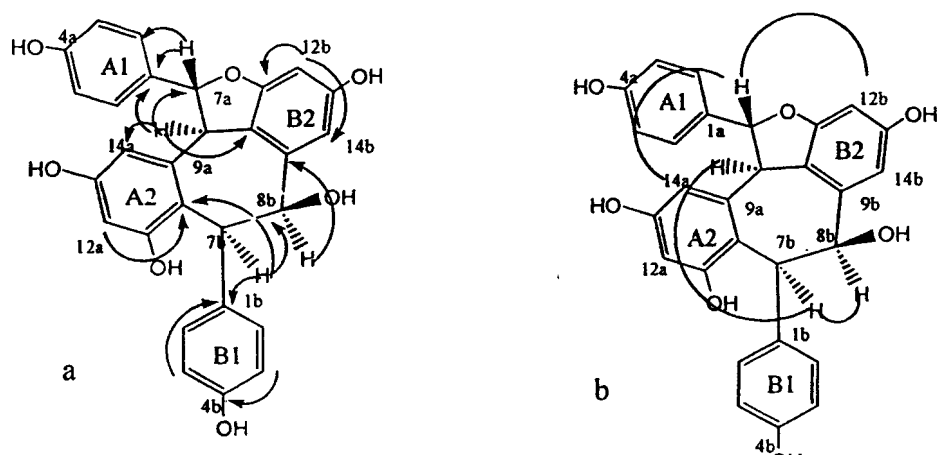


Fig 1 Significant HMBC (a) dan NOESY (b) correlation of 1

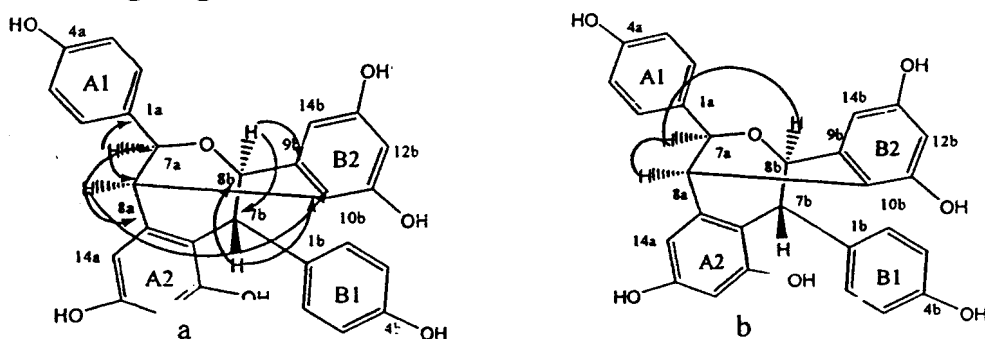


Fig 2 Significant HMBC (a) and NOESY (b) correlations of 2

2H (ring B1). The  $^1\text{H}$  NMR spectrum also showed two sets of meta-coupled aromatic protons signals at  $\delta$  6.09 ( $d$ ,  $J = 2.2$  Hz) and 5.96 ( $d$ ,  $J = 2.2$  Hz) ppm, each 1H (ring A2), and at  $\delta$  6.20 ( $d$ ,  $J = 2.2$  Hz) and 6.25 ( $d$ ,  $J = 2.2$  Hz) ppm, each 1H (ring B2). Additionally, the  $^1\text{H}$  NMR spectrum exhibited signals for a set of aliphatic proton at  $\delta$  5.70 ( $d$ ,  $J = 9.5$  Hz) and 5.16 ( $d$ ,  $J = 9.5$  Hz), each 1H, characteristic for *trans*-2,3-diaryl-dihydrobenzofuran moiety, and signals assignable two coupled aliphatic protons at  $\delta$  4.89 (*br. s*) and 5.39 (*br. s*) ppm, each 1H. These spectral data indicated that compound 1 has a dimeric stilbene skeleton as part of its structure.

The HMQC spectrum supported complete assignment of all proton-bearing carbon signals of compound 1 (Table 1). Further support for the structure 1 was obtained from HMBC measurement (Figure 1). The HMBC spectrum of 1 showed long-range correlations between H-7a/C-1a, H-7a/C-2a, H-8a/C-7a, H-8a/C-1a, H-8a/C-10a, and H-8a/C-10b, indicating the presence of *trans*-2,3-diaryl-dihydrobenzofuran moiety. Long-range correlation were also observed for the methine proton between H-7b/C-10a, H-7b/C-1b, H-

7b/C-8b, and H-8b/C-9b, indicating that A2 and B2 ring attached with sikloheksan ring. The relative stereochemistry of 1 was identified by means of NOESY spectrum. The NOE interactions between H-7a/H-14a indicated that the relative configuration of both methine protons of the dihydrofuran ring were *trans*-configuration. The NOESY experiments also showed the interaction between H-8a/H-7b and H-7b/H-8b, indicated that the relative configuration of both methine proton H-7b and H-8b were *cis*-configurations. Further evidence for the structure assigned to compound 1 came from comparison of the NMR data to that reported for balanocarpol. Therefore, it may be conclude that 1 is balanocarpol [14].

Compound 2 was obtained as a light brown powder, maxima of absorption were observed at 225; 230 sh; 282 nm in the UV spectrum attributable to the phenol rings. The IR spectrum exhibited hydroxyl group ( $3352\text{ cm}^{-1}$ ), C=C aromatic ( $1606$ ;  $1512$ ;  $1450\text{ cm}^{-1}$ ), and monosubstituen benzene ( $835\text{ cm}^{-1}$ ). Its molecular formula of  $\text{C}_{28}\text{H}_{22}\text{O}_7$  was established by FABMS, showing a  $[\text{M}+\text{H}]^+$  ion at  $m/z$  471, together with its NMR spectral data, suggesting that 2 was

resveratrol dimer. The  $^1\text{H}$  NMR (Table 2) and  $^1\text{H}$ - $^1\text{H}$  COSY spectra showed two sets of AA'BB' system of aromatic protons assignable to two independent 4-hydroxyphenyl groups at 6.90 (2H, *d*,  $J = 8.4$  Hz) and 6.69 (2H, *d*,  $J = 8.4$  Hz) (ring A1), and 7.14 (2H, *d*,  $J = 8.4$  Hz) and 6.72 (2H, *d*,  $J = 8.4$  Hz) (ring B2), two sets of *meta*-coupled aromatic protons at 6.41 (1H, *d*,  $J = 2.6$  Hz) and 6.16 (1H, *d*,  $J = 2.6$  Hz) (ring A2), 6.48 (1H, *d*,  $J = 2.2$  Hz) and 6.21 (1H, *d*,  $J = 2.2$  Hz) (ring B2) assignable to two units 1,2,3,5-tetrasubstituted benzene group. They also displayed two set of copuled benzyl methine protons at 5.57 (1H, *br s*) (7a), 4.24 (1H, *br. s*) (8a), 4.32 (1H, *d*,  $J = 3.3$  Hz) (7b), 4.97 (1H, *d*,  $J = 3.3$  Hz) (8b). The  $^{13}\text{C}$  NMR spectrum showed that C-7a (81.5 ppm) and C-8b (81.4 ppm) indicated that they might both be attached to benzylic carbons bearing an oxygen atom. The connection between protons and their corresponding carbons was established by HMQC. Further support for the structure 2 was obtained form HMBC measurement (Fig 2). The HMBC spectrum (Fig 2) of 2 showed long-range correlations between H-2a with C-7a (81,5 ppm) confirmed that a 4-hydroxyphenyl group is attached to an oxygen bearing carbon. Long-range correlation were also observed for the methine proton between H-8b/C-7b, H-7b/C-10b, and H-8a/C-10b showed to a fused benzopyran-benzo-oxepane structure, in the same pattern with those of heimiol A<sup>[18]</sup>. The relatif configuration of 2 was established on the basis of the NOESY spectra (Figure 2). The NOE correlation showed that the H-8a and H-8b are in a *syn* configuration, deduced from the NOE correlations between H-8b/H-7a/H-8a, as well as H-7b does not show any correlations. Therefore, it may be concluded that the 2 is heimiol A, a resveratrol dimer.

## CONCLUSION

From the non polar fraction extract acetone stem bark of *H. mengarawan* can be isolated two resveratrols dimmer, namely balanocarpol (1) and heimiol A (2).

## ACKNOWLEDGEMENT

This work was supported by competitive grant XII-2004, Directorate General Higher Education, Republic of Indonesia. The authors are grateful to the experimental Garden in Carita, Pandeglang, Banten, Indonesia and Herbarium Bogoriensis for supported the sample and identification of the plant specimen.

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