TWO NEW DIMERIC XANTHONES IN *MESUA FERREA*

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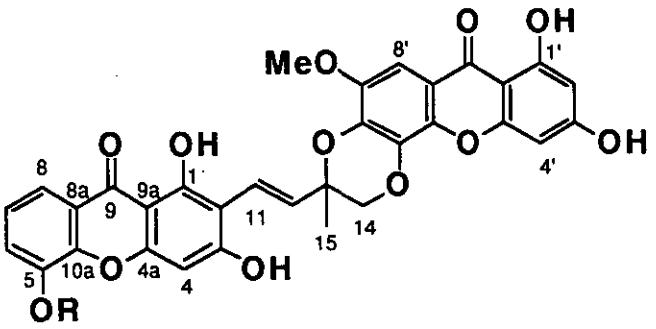
Abstract — Two new dimeric xanthones linked through a C_5 unit, mesuferrols A and B, were isolated from the bark of *Mesua ferrea* (Guttiferae), in addition to two known xanthones (1,7-dihydroxy- and 5-hydroxy-1-methoxyxanthone) and a flavonoid ((-)-epicatechin). The structures of these compounds were established by the aids of spectroscopic analysis including 2D nmr technique.

Mesua ferrea L. (Guttiferae) is an indigenous plant to tropical Asia. The seed is used as a traditional medicine against eczema and rheumatism, and the flowers are also supplied to aroma and expectorant in mental disturbance in Indonesia.[1] The occurrence of simply oxygenated xanthones[2] and coumarin derivatives[3] in *M. ferrea* has been reported. In continuation of our studies oriented to search for biologically active substances in Guttiferaeous plants,[4-8] chemical constituents in the bark of *M. ferrea* were examined. We report here the isolation and structural characterization of two new xanthone dimers. By chromatographic purification, compounds (**1**-**5**) were isolated from an acetone extract of the bark of *M. ferrea*.

Compound (**1**), mesuferrol A, a pale yellow amorphous powder, reacted positively to $FeCl_3$ and Gibbs tests. High-resolution (HR) FABms showed $[M-H]^-$ at *m/z* 597.1005, indicating the molecular formula of $C_{32}H_{22}O_{12}$. In the ^1H nmr spectrum, four hydroxyls [δ 9.60 (2H, br s), 13.08 and 14.02 (1H each, s, chelated)] and a methoxyl group [δ 3.98 (3H, s)] were observed in addition to two protons in *meta*-coupling [δ 6.23 and 6.44 (1H each, d, J = 2.0 Hz)], three protons in an ABC system based on a 1,2,3-trisubstituted benzene ring [δ 7.23

(1H, t, J = 7.8 Hz), 7.33 and 7.66 (1H each, dd, J = 7.8, 1.0 Hz)] and aromatic protons in singlet [δ 6.58 and 7.23 (1H each)]. The chemical shifts of all carbons with hydrogen were assigned by the CH COSY spectrum



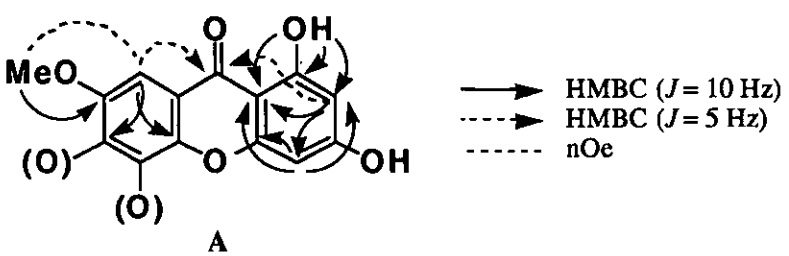
1: R = H
2: R = Me

(Table 1). In the HMBC (J = 10 Hz) spectrum (Figure 1), the chelated hydroxyl group (δ 13.08) caused cross peaks to three aromatic carbons (δ 98.9, 103.5 and 164.3), the former (δ 98.9) was further correlated to one of the *meta*-coupled protons at δ 6.23 in the CH COSY spectrum. The proton at δ 6.23 was correlated to the carbonyl carbon at δ 180.4 in the HMBC (J = 5 Hz) through ^4J. An nOe was observed between the methoxyl group and the aromatic proton at δ 7.23 which was correlated to the carbonyl carbon (δ 180.4) through ^3J in the HMBC (J = 5 Hz) spectrum (Figure 1). Furthermore, the proton at δ 7.23 caused cross peak to the aromatic carbons with an oxygen-function at δ 140.2 and 142.5 in the HMBC (J = 10 Hz) spectrum. The chemical shift of the carbonyl carbon at δ 180.4 showed a characteristic feature of a 1-hydroxyxanthone (a xanthone with a chelated hydroxyl group).[9] These data led a plausible partial structure of a 1-hydroxy-7-methoxy-3,5,6-trioxygenated xanthone (**A**) (Figure 1). The ^{1}H and ^{13}C nmr spectral data based on the partial structure **A** closely resembled those of 1,3,5,6-tetrahydroxy-7-methoxyxanthone (caloxanthone E) previously isolated from *Calophyllum inophyllum*[5] and the other correlations in the HMBC spectrum (Figure 1) also supported the partial structure. The proton (δ 7.66) in an ABC system was correlated to the aromatic carbon at δ 145.8 which caused a cross peak to the proton at δ 7.33, and the proton at δ 7.23 was also correlated to the carbon at δ 146.8 in the HMBC (J = 10 Hz) spectrum (Figure 2). The chemical shifts of the aromatic carbons with an oxygen-function suggested the substitution to be a catechol type such as **B**. The other partial structure **C** was determined as follows. In the ^{1}H nmr spectrum, the signals due to *trans*-olefinic protons [δ 6.99 and 7.11 (1H each d, J = 16.6 Hz)], a methyl group [δ 1.64 (1H, s)] and *gem*-methylene protons [δ 4.25 and 4.43 (1H each d, J = 11.5 Hz)]

Table 1. ^{1}H (400 MHz) and ^{13}C (100 MHz) nmr spectral data of **1** and **2**

	1 (acetone-d_6)		**2**		
No.	δ_C	δ_H J (Hz)	δ_C	δ_H (DMSO-d_6) J (Hz)	δ_H (acetone-d_6) J (Hz)
1	162.8		160.9		
2	107.5		106.0		
3	165.7[a]		165.0[a]		
4	94.6[b]	6.58 (1H, s)	96.4	6.45 (1H, s)	6.59 (1H, s)
5	146.8		147.9		
6	121.5	7.66 (1H, dd, 7.8, 1.0)	120.4[b]	7.48 (1H, dd, 7.8, 1.0)	7.46 (1H, dd, 7.3, 1.0)
7	124.9	7.23 (1H, t, 7.8)	124.1	7.37 (1H, t, 7.8)	7.37 (1H, t, 7.3)
8	116.3	7.33 (1H, dd, 7.8, 1.0)	115.6	7.66 (1H, dd, 7.8, 1.0)	7.74 (1H, dd, 7.3, 1.0)
9	181.9		179.9		
4a	157.1		155.6		
8a	122.1		120.4		
9a	103.5[c]		101.7[c]		
10a	145.8		145.2		
11	121.2	7.11 (1H, d, 16.6)	119.8	6.90 (1H, d, 16.6)	7.12 (1H, d, 16.6)
12	131.7	6.99 (1H, d, 16.6)	130.3	6.88 (1H, d, 16.6)	7.02 (1H, d, 16.6)
13	77.4		76.5		
14	71.5	4.25, 4.43 (1H each, d, 11.5)	71.0	4.21, 4.42 (1H each, d, 11.2)	4.25, 4.45 (1H each, d, 11.5)
15	23.0	1.64 (3H, s)	22.4		
1'	164.3		162.5		
2'	98.9	6.23 (1H, d, 2.0)	98.0	6.18 (1H, d, 2.0)	6.24 (1H, d, 2.4)
3'	164.5[a]		164.1[a]		
4'	94.6[b]	6.44 (1H, d, 2.0)	93.7	6.38 (1H, d, 2.0)	6.45 (1H, d, 2.4)
5'	132.6		131.2		
6'	140.2		138.7		
7'	147.7		146.1		
8'	97.8	7.23 (1H, s)	95.4	7.16 (1H, s)	7.24 (1H, s)
9'	180.4		178.8		
4a'	158.7		157.0		
8a'	113.6		112.0		
9a'	103.5[c]		101.7[c]		
10a'	142.5		140.8		
OMe-C-5			56.2	3.95 (3H, s)	4.02 (3H, s)
OMe-C-7'	56.6	3.98 (3H, s)	55.9	3.93 (3H, s)	3.99 (3H, s)
OH-C-1		14.02 (1H, s)		13.90 (1H, s)	14.00 (1H, s)
OH-C-1'		13.08 (1H, s)		12.99 (1H, s)	13.11 (1H, s)
OH		9.60 (2H, br s)		10.90 (2H, br s)	9.80 (2H, br s)

All carbons were assigned by CH COSY and HMBC spectrum. a: interchangeable. b, c: overlapping.



A

Figure 1 Plausible partial structure (A) in **1**, and HMBC and nOe results

were observed. On the other hand, in the HMBC (J = 10 Hz) spectrum (Figure 2), the chelated hydroxyl group at δ 14.02 was correlated to three aromatic carbons (δ 103.5, 107.5 and 162.8), the latter of which (δ 162.8) was further correlated to one of the *trans*-olefinic protons at δ 7.11. The quaternary aromatic carbon at δ 107.5 caused cross peaks to another *trans*-olefinic proton (δ 6.99) and the aromatic proton at δ 6.58. In the ^{13}C nmr spectrum, the remaining aromatic carbons with an oxygen-function were observed at δ 157.1 and 165.7, which indicated the presence of a benzene ring with a phloroglucinol type. Furthermore, nOes were observed in the methyl group (δ 1.64), *trans*-olefinic protons (δ 6.99 and 7.11) and the *gem*-methylene proton (δ 4.23) (Figure 2) when the methyl was irradiated. The methyl proton (δ 1.64) caused a cross peak to the quaternary carbon with an oxygen-function (δ 77.4) which was correlated to the *trans*-olefinic protons (δ 6.99 and 7.11) in the HMBC spectrum (Figure 2). These results substantiated the presence of the partial structure **C**. A connective moiety of the partial structures (**A**-**C**) was characterized as follows. In the HMBC (J = 5 Hz) spectrum, the *gem*-methylene protons (δ 4.25 and 4.43) were correlated to the aromatic carbon at δ 132.6 through ^3J shown as **D** in Figure 2. The other carbonyl carbon was observed at δ 181.9 in the ^{13}C nmr spectrum, which supported the presence of a 1-hydroxyxanthone.[9] Taking the above data and the degrees of unsaturation into consideration, the structure of **1** is a dimeric xanthone linked through a C_5 unit. Thus the total structure of mesuferrol A was concluded to be **1**.

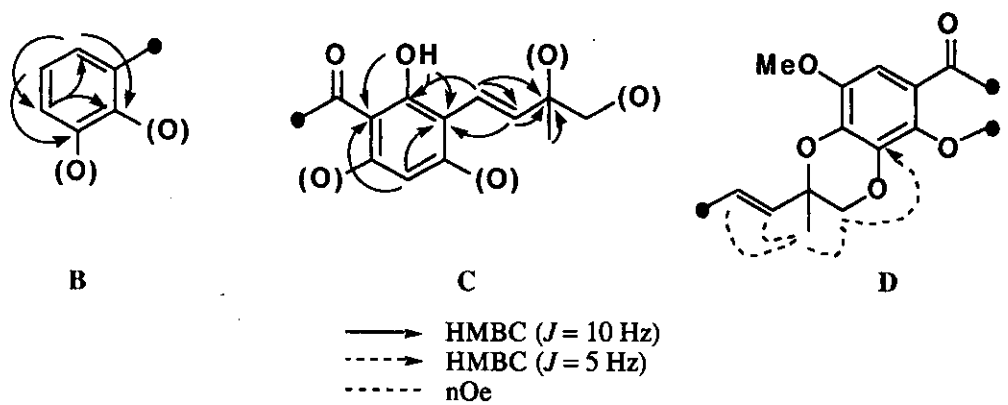


Figure 2 Plausible partial structures (**B**-**D**), and HMBC and nOe results

Compound (2), mesuferrol B, was obtained as a pale yellow amorphous powder, and its uv absorptions were closely similar to those of **1**. The molecular formula of $C_{33}H_{24}O_{12}$ was deduced by the HR-FABms. Analysis of the ^1H and ^{13}C nmr spectral data (Table 1) revealed that **2** was a 5-methyl ether of **1**. The structure (2) was supported by the HMBC spectrum and nOe experiment (Figure 3).

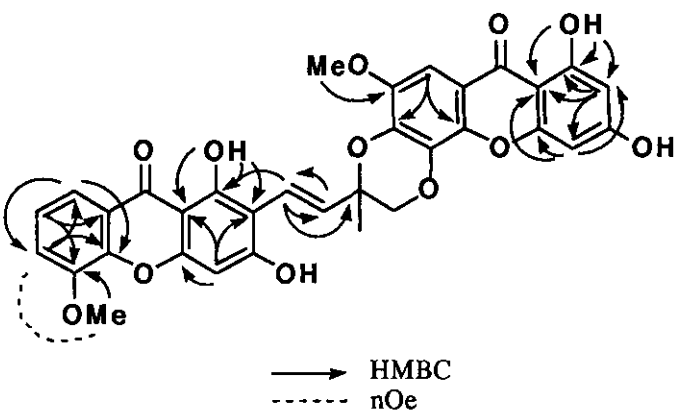


Figure 3 HMBC (J = 10 Hz) and nOe results in **2**

Compounds (**3**-**5**) were identified as 1,7-dihydroxy- (**3**), 5-hydroxy-1-methoxyxanthone (**4**) and (-)-epicatechin (**5**) by spectroscopic analysis including 2D nmr technique.

A xanthone dimer linked *via* a C_5 unit is very rare natural product, and the occurrence has been reported only in the family Guttiferae (*Garcinia livingstonei*,[10] *Mesua ferrea*,[11] *Cratoxylum cochinchinense*[12]).

EXPERIMENTAL

General. The following instruments were used: Ms spectra, JEOL JMS-D300 (70 eV) instrument; ^1H and ^{13}C nmr spectra, JEOL JNM EX-400 (TMS as internal standard), ir spectra (on KBr pellet), JASCO IR-AI spectrophotometer; Polarimeter, JASCO DIP-370 digital polarimeter; uv (in methanol solution), Shimadzu UV-2200 spectrophotometer. The following adsorbents were used for purification: analytical tlc, Merck Kieselgel 60 F_{254}; column chromatography, Merck Kieselgel 60.

Plant material. Bark of *Mesua ferrea* was collected at North Pasuruhan in East Java, Indonesia, January 1995. The voucher specimen is deposited in the herbarium of Gifu Pharmaceutical University.

Extraction and isolation. The dried and ground bark (400 g) of *M. ferrea* was extracted successively with benzene, acetone and 70% MeOH. The acetone extract (20 g) was subjected to silica gel column chromatography eluted with a $CHCl_3$-MeOH system to give 15 fractions. The second fraction ($CHCl_3$-MeOH = 7 : 1) was further purified by ptlc (*n*-hexane-EtOAc-MeOH = 8 : 2 : 1) to give **3** (2 mg) and **4** (2 mg). An amorphous mixture (**1** and **2**) was obtained from the fourth fraction ($CHCl_3$-MeOH = 7 : 1) after filtration. The mixture was separated by ptlc ($CHCl_3$-MeOH = 10 : 1) to give **1** (5 mg) and **2** (3 mg). Compound **5** (5 mg) was obtained from the seventh fraction ($CHCl_3$-MeOH = 5 : 1).

Compound 1 (mesuferrol A): A pale yellow amorphous powder; $[\alpha]_D^{24}$: -4° (*c* 0.05, MeOH); HR-FABms: $[M-H]^+$ *m/z* 597.1005 (Calcd 597.1033 for $C_{32}H_{21}O_{12}$); uv λ (nm): 206, 258, 285, 319, 356sh; ir ν (cm^{-1}): 3400, 1640, 1605, 1580. The ^1H and ^{13}C nmr spectral data are shown in Table 1.

Compound 2 (mesuferrol B): A pale yellow amorphous powder; $[\alpha]_D^{24}$: -4° (*c* 0.05, MeOH); HR-FABms: $[M-H]^+$ *m/z* 611.1163 (Calcd 611.1189 for $C_{33}H_{23}O_{12}$); uv λ (nm): 204, 258, 285, 316, 355sh; ir ν (cm^{-1}): 3240, 1640, 1605, 1580. The ^1H and ^{13}C nmr spectral data are shown in Table 1.

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