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Beccaridiol, an unusual 28-nortriterpenoid from the leaves of *Diplectria beccariana*

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Dedicated to Professor Rodney B. Croteau on the occasion of his 60th birthday

Abstract

A C_{29}-triterpene, beccaridiol (**1**), a dihydrochalcone natural product, 2′,4′-dihydroxy-3-(4-methoxyphenyl)-propiophenone (**2**), as well as three known compounds, 4′-hydroxy-1′,2′-dihydro-β-ionone, 4′-*O*-methyldavidigenin (**3**), and ursolic acid, have been isolated from an EtOAc-soluble extract of the leaves of *Diplectria beccariana*. Beccaridiol (**1**) was characterized as an ursane-type 28-nortriterpene possessing an unusual aromatic E-ring by spectroscopic data interpretation. The relative configuration of this unusual isolate was established by analyzing the observed NOESY NMR correlations, and the absolute stereochemistry of **1** was then determined based on the circular dichroism (CD) spectrum of its 2,3-di-*p*-bromobenzoate (**1b**) derivative. All isolates were evaluated for their potential cancer chemopreventive properties utilizing a cell culture assay to determine quinone reductase induction.

Keywords: *Diplectria beccariana*; Melastomataceae; Nortriterpenoid; Beccaridiol; Dihydrochalcone; Circular dichroism; Quinone reductase

1. Introduction

The family Melastomataceae comprises about 4570 species in 150–156 genera, and is represented by herbs, shrubs, and trees (Renner, 1993; Clausing and Renner, 2001a,b). Several hundred species of this family grow epiphytically, and therefore, Melastomataceae is a significant contributor to the world's epiphytes and climbers (Clausing and Renner, 2001b). The major chemical constituents of species of the Melastomataceae have been found to be flavonoids (Herrera and Bain, 1991; Santos and Salatino, 2000), triterpenoids (Chan et al., 1992; Chaturvedula et al., 2004), and hydrolysable tannins (Yoshida et al., 1991; Isaza et al., 2004). *Diplectria* is a small genus of the Melastomataceae, and contains only two species, *D. barbata* and *D. beccariana*. These two species are mainly distributed in Southeast Asia. A literature survey revealed that no previous phytochemical or biological studies have been conducted on this genus. As part of a research program directed towards the discovery of new cancer chemopreventive agents from plants (Pezzuto, 1997; Kinghorn

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et al., 2003, 2004), an EtOAc-soluble extract of the leaves of *D. beccariana* was found to induce quinone reductase (QR) activity in cultured Hepa lclc7 mouse hepatoma cells (Prochaska and Santamaria, 1988; Misico et al., 2002). Bioassay-directed fractionation of this extract led to the isolation of an ursane C_{29}-triterpene, beccaridiol (**1**), a dihydrochalcone natural product, 2′,4′-dihydroxy-3-(4-methoxyphenyl)-propiophenone (**2**), and three known compounds. We report herein the isolation and structure determination of compounds **1** and **2**, as well as the evaluation in the QR induction assay of all isolates obtained in this study.

2. Results and discussion

Compound **1** was obtained as a white amorphous powder (petroleum ether–acetone; 3:1), m.p. 126–128 °C, $[\alpha]_D^{23}$ +58.0° (*c* 0.4, $CHCl_3$). A molecular formula of $C_{29}H_{42}O_2$ was determined for this compound from the molecular ion peak at *m/z* 422.3213 $[M]^+$ (Calc. for $C_{29}H_{42}O_2$, 422.3185) obtained by HREIMS, consistent with nine degrees of unsaturation. The ^1H NMR spectrum (Table 1) of compound **1** displayed the characteristic signals of seven methyl groups as singlets, two oxygenated methines at δ_H 3.03 (*d*, $J = 9.5$ Hz, H-3) and 3.70 (*ddd*, $J = 11.4$, 9.5, 4.4 Hz, H-2), an olefinic proton at δ_H 5.50 (*dd*, $J = 4.6$, 2.9 Hz, H-12), and two aromatic protons at 6.78 (*d*, $J = 7.4$, H-21), and 6.92 (*d*, $J = 7.4$ Hz, H-22). The ^{13}C and DEPT NMR spectra (Table 1) of compound **1** showed 28 carbon signals, including characteristic signals of six methyls (δ_c 16.8, 16.9, 17.3, 20.8, 27.3, and 28.6), two oxygenated methines (δ_c 69.1 and 84.0), and eight olefinic and/or aromatic carbons (δ_c 122.9, 127.3, 125.1, 133.8, 135.1, 138.4, 138.6, and 138.9). However, in the HMQC spectrum of **1**, the relatively more intense methyl signal at δ_c 16.9 was correlated with two methyl singlets at δ_H 1.03 (*s*, CH_3-26) and 2.27 (*s*, CH_3-29). This indicated that two methyls (CH_3-26 and CH_3-29) have the same chemical shifts and their carbon signals were overlapped at δ_c 16.9 in the ^{13}C NMR spectrum of **1**. Therefore, 29 carbon atoms are present in the molecule of compound **1**, which is consistent with the determined molecular formula and the above-mentioned ^1H NMR data analysis. Interpretation of the observed correlations in the ^1H–^1H COSY, HMQC, HMBC, and NOESY spectra of compound **1** suggested that this isolate is a triterpenoid, whose rings A, B and C are the same as those of many widely occurring ursane and oleanane triterpenoids (Connolly and Hill, 2005). In the HMBC spectrum of **1**, the proton signals of CH_3-23 and CH_3-24 were correlated with one of two oxygenated methines at δ_c 84.0. This, in combination with the observed ^1H–^1H COSY correlation between two oxygenated methines at δ_H 3.03 (H-3) and 3.70 (H-2), indicated the presence of two hydroxyl groups at C-2 and C-3. The location of the double bond was assigned between C-12 and C-13 based on the HMBC correlations from CH_3-27 to C-8, C-13, C-14, and C-15, and from H-12 to C-9, C-11, C-13, and C-14. Thus, an aromatic ring must be present as ring E in the molecule of **1**. On further analysis of the HMBC spectrum of **1**, correlations from CH_3-29 to C-l8, C-19, and C-20, and from CH_3-30 to C-19, C-20, and C-21 were observed. These data were used to determine that the two aromatic methyls were at C-19 and C-20, respectively. Generally, a methyl, a hydroxymethyl, or a carboxylic acid group is present at C-l7 of triterpenoids (Bhandari et al., 1990; Lee, 1998). However, this carbon is absent in the molecule of **1**. Therefore, compound **1** (beccaridiol) is an ursane 28-nortriterpene containing an unusual aromatic E-ring. Only a few 28-nortriterpenes with an aromatic E-ring have been isolated previously such as from *Dillenia indica* (Dilleniaceae) (Bhattacharjee and Chatterjee, 1962) and mussel shale (Schaeffer et al., 1995).

The H-3 signal was displayed in the ^1H NMR spectrum of **1** as a doublet with a coupling constant of 9.5 Hz, which suggested both H-2 and H-3 are axial protons and that they are *trans*-oriented. In the NOESY spectrum of **1**, correlations from H-3 to H-5 and CH_3-23, and from H-3 to CH_3-24 and CH_3-25 were observed. These correlations indicated the relative configurations of OH-2 and OH-3 to be α and β, respectively. This assignment was confirmed

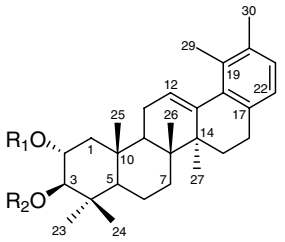
Table 1
NMR spectroscopic data of compound **1**[a]

Position	δ_H	δ_C
1	2.09–2.18, *m*; 1.00–1.07, *m*	46.8 *t*
2	3.70, *ddd* (11.4, 9.5, 4.4)	69.1 *d*
3	3.03, *d* (9.5)	84.0 *d*
4		39.2 *s*
5	0.99, *m*	55.6 *d*
6	1.47–1.66, *m*	18.2 *t*
7	1.47–1.66, *m*	33.8 *t*
8		40.1 *s*
9	1.71, *dd* (11.2, 6.1)	47.2 *d*
10		38.3 *s*
11	2.09–2.18, *m*	23.5 *t*
12	5.50, *dd* (4.6, 2.9)	125.1 *d*
13		138.9 *s*
14		44.2 *s*
15	2.23, *m*; 0.89–0.99, *m*	32.1 *t*
16	2.38–2.42, *m*	31.0 *t*
17		138.4 *s*
18		138.6 *s*
19		135.1 *s*
20		133.8 *s*
21	6.78, *d* (7.4)	127.3 *d*
22	6.92, *d* (7.4)	122.9 *d*
23	1.07, *s*	28.6 *q*
24	0.88, *s*	16.8 *q*[b]
25	1.05, *s*	17.3 *q*
26	1.03, *s*	16.9 *q*[b]
27	0.94, *s*	27.3 *q*
29	2.27, *s*	16.9 *q*[b]
30	2.27, *s*	20.8 *q*

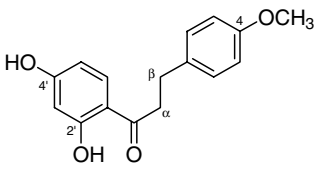
[a] ^1H and ^{13}C NMR spectra were acquired in $CDCl_3$ at 300 and 75 MHz, respectively; TMS was used as internal standard; chemical shifts are shown in the δ scale with *J* values (Hz) in parentheses. Assignments are based on ^1H–^1H COSY, HMQC, HMBC, and NOESY spectra.

[b] Assignments are interchangeable in the same column.

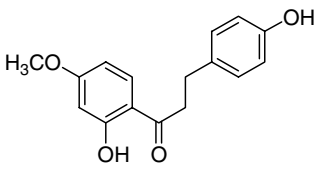
by comparison of the chemical shifts, splitting patterns and coupling constants of H-2 (δ_H 3.70, *ddd*, $J = 11.4$, 9.5, 4.4 Hz) and H-3 (δ_H 3.03, *d*, $J = 9.5$ Hz, H-3) of **1** with those of the literature values (H-2: δ_H 3.70, *ddd*, $J = 11.2$, 9.6, 4.6 Hz; H-3: δ_H 3.00, *d*, $J = 9.3$ Hz) reported for similar compounds (De Sousa Menezes et al., 1998). The absolute stereochemistry of beccaridiol (**1**) was determined from its circular dichroism (CD) spectrum, since the CD exciton chirality method is a well-established procedure for the absolute configuration determination of organic compounds possessing a 1,2-diol functionality (Harada and Nakanishi, 1972). Beccaridiol (**1**) was treated with *p*-bromobenzoyl chloride in pyridine, to give its 2-mono-*p*-bromobenzoate (**1a**) and 2,3-di-*p*-bromobenzoate (**1b**) derivatives. A negative chirality (Harada and Nakanishi, 1972) was observed in the CD spectrum of **1b**. This, in combination with the determined relative configuration of **1**, was used to determine the absolute stereochemistry of beccaridiol (**1**) as 2*R* and 3*R*, respectively. Therefore, the structure of beccaridiol (**1**) was characterized as (2*R*,3*R*)-28-norursa-12,17,19,21-tetraen-2,3-diol.



1 $R_1 = R_2 = H$
1a $R_1 = COC_6H_4(p)Br$ $R_2 = H$
1b $R_1 = R_2 = COC_6H_4(p)Br$



2



3

Compounds **2** and **3** were initially isolated as a mixture by repeated silica gel chromatography. The ^{1}H NMR spectrum of the mixture suggested the presence of two similar phenolic compounds in a ratio of about 5 to 4. This mixture was then separated by preparative TLC using benzene–EtOAc (14:1, run three times) as developing solvent, to yield pure compounds **2** and **3**. A molecular formula of $C_{16}H_{16}O_4$ was assigned for **2** based on the molecular ion peak at *m/z* 272.1047 $[M]^+$ (Calc. for $C_{16}H_{16}O_4$, 272.1049) in its HREIMS. The ^{1}H NMR spectrum (Table 2) of compound **2** displayed signals of two methylene triplets at δ_H 2.98 (2H, *t*, $J = 7.2$ Hz, H_2-β) and 3.19 (2H, *t*, $J = 7.2$ Hz, H_2-α), which were coupled to each other, a methoxy group at δ_H 3.83 (3H, *s*, OMe-4), a *para*-substituted aromatic ring at δ_H 7.11 (2H, *d*, $J = 8.5$ Hz, H-2 and H-6) and 6.78 (2H, *d*, $J = 8.5$ Hz, H-3 and H-5), and a 1,2,4-trisubstituted aromatic ring at δ_H 6.43 (1H, *d*, $J = 2.5$ Hz, H-3′), 6.41 (1H, *dd*, $J = 9.6$, 2.5 Hz, H-5′), and 7.63 (1H, *d*, $J = 9.6$ Hz, H-6′). Also observed was a downfield exchangeable singlet at δ_H 12.81 (1H, *s*, OH-2′), which is generally characteristic for the chelated hydroxyl group of flavonoids. Consistent with the ^{1}H NMR spectroscopic analysis, the ^{13}C and DEPT NMR spectra (Table 2) of compound **2** also displayed two methylenes, one methoxy group, the signals of two aromatic rings, and a conjugated ketone at δ_c 203.7 (C=O). Accordingly, this suggested that compound **2** is a dihydrochalcone (Yao et al., 2005). Thus, two hydroxy groups in **2** could be inferred from the molecular formula of $C_{16}H_{16}O_4$, since only one methoxy group was evident in both the ^{1}H and ^{13}C NMR spectra. Based on the ^{1}H NMR spectroscopic data, the methoxy and the two hydroxyl groups should be attached at C-4, C-2′ and C-4′, respectively. In the NOESY spectrum of **2**, the correlation from the methoxy group at δ_H 3.83 to one of the doublets of the *para*-substituted aromatic ring at δ_H 6.78 (H-3 and H-5) was observed clearly, which determined the location of the methoxy group at C-4. Thus, the two hydroxyl groups were assigned at C-2′ and C-4′, respectively, and were confirmed by the observed correlations in the HMBC spectrum of **2**. This is the first isolation of compound **2** from a natural source, although the synthesis of this dihydrochalcone has been reported (Jain and Mehta, 1985; Narender et al., 2004).

Both the ^{1}H and ^{13}C NMR spectra (Table 2) of compound **3** were very similar to those of **2**. The HREIMS of **3** was used to establish a molecular formula of $C_{16}H_{16}O_4$, which is also the same as that of **2**. Interpretation of the 2D NMR spectra of **3** suggested that the difference between these two dihydrochalcones is the location of the methoxy group. In the NOESY spectrum of **3**, the methoxy group at δ_H 3.79 correlated to both H-3′ and H-5′, which determined the position of the methoxy group to be at H-4′. This dihydrochalcone, 4′-*O*-methyldavidigenin (**3**), was previously isolated from the leaves of *Viburnum davidii* (Jensen et al., 1977). Its complete NMR data assignments based on 2D NMR spectroscopic correlations are given in Table 2.

The structures of two other known compounds, 4′-hydroxy-1,2′-dihydro-β-ionone (Wirth et al., 2001) and ursolic acid (Numata et al., 1989), were identified by physical and spectroscopic data measurement ($[\alpha]_D$, ^{1}H NMR, ^{13}C NMR, DEPT, 2D NMR, and MS) and by comparing the data obtained with those of published values.

All five compounds isolated from *D. beccariana* leaves in the present study were evaluated for their potential to

Table 2
NMR spectroscopic data of compounds **2** and **3**[a]

Position	**2**		**3**	
	δ_H	δ_C	δ_H	δ_C
1		133.0 *s*		132.9 *s*
2	7.11, *d* (8.5)	129.5 *d*	7.16, *d* (8.6)	129.3 *d*
3	6.78, *d* (8.5)	115.4 *d*	6.84, *d* (8.6)	114.0 *d*
4		154.0 *s*		158.1 *s*
5	6.78, *d* (8.5)	115.4 *d*	6.84, *d* (8.6)	114.0 *d*
6	7.11, *d* (8.5)	129.5 *d*	7.16, *d* (8.6)	129.3 *d*
1′		113.5 *s*		113.9 *s*
2′		165.4 *s*		165.2 *s*
3′	6.43, *d* (2.5)	101.0 *d*	6.37, *d* (2.5)	103.6 *d*
4′		166.1 *s*		162.5 *s*
5′	6.41, *dd* (9.6, 2.5)	107.7 *d*	6.34, *dd* (9.0, 2.5)	107.7 *d*
6′	7.63, *d* (9.6)	131.5 *d*	7.63, *d* (9.0)	132.2 *d*
C=O		203.7 *s*		203.8 *s*
α	3.19, *t* (7.2)	39.9 *t*	3.20, *t* (7.1)	40.0 *t*
β	2.98, *t* (7.2)	29.6 *t*	2.99, *t* (7.1)	29.5 *t*
OMe-4	3.83, *s*	55.6 *q*		
OMe-4′			3.79, *s*	56.3 *q*
OH-2′	12.81, *s*		12.75, *s*	

[a] ^1H and ^{13}C NMR spectra were acquired in $CDCl_3$ at 300 and 75 MHz, respectively; TMS was used as internal standard; chemical shifts are shown in the δ scale with J values (Hz) in parentheses. Assignments are based on ^1H–^1H COSY, HMQC, HMBC, and NOESY spectra.

induce QR (Prochaska and Santamaria, 1988; Misico et al., 2002). Among these isolates, only one of the dihydrochalcones, 4′-*O*-methyldavidigenin (**3**), was found to be active with CD (concentration to double activity), IC_{50}, and chemoprevention index (IC_{50}/CD) values of 8.0 μg/mL, 28.9 μg/mL, and 3.6, respectively. Compounds **1, 2**, 4′-hydroxy-1,2′-dihydro-β-ionone and ursolic acid were inactive in this test system (CD > 10 μg/mL). The difference between the structures of **2** and **3** is the location of the methoxy group, with the active regioisomer, 4′-*O*-methyldavidigenin (**3**), possessing a methoxy group at C-4′. This is consistent with the functional group arrangement in pinostrobin (5-hydroxy-7-methoxyflavanone), another QR inducer identified in our program on potential natural product cancer chemopreventives (Gu et al., 2002; Su et al., 2003). These results suggest that the presence of a methoxy group in ring A at a *para*-position to the carbonyl group (e.g., C-4′ for dihydrochalcones and C-7 for flavanones) may augment the QR induction activity of such flavonoids.

3. Experimental

3.1. General

Melting points were determined on a Fisher–Johns melting-point apparatus and are uncorrected. Optical rotations were obtained using a Perkin–Elmer 241 polarimeter. UV spectra were recorded with a Beckman DU-7 spectrometer. CD spectra were measured with a JASCO J-810 spectropolarimeter. IR spectra were run on an ATI Mattson Genesis Series FT-IR spectrometer. NMR experiments were conducted on Bruker DPX-300 and DRX-500 MHz spectrometers with tetramethylsilane (TMS) as internal standard. EIMS and HREIMS were obtained on a Finnigan MAT 95 sector-field mass spectrometer or a JEOL GCmate II mass spectrometer, operating at 70 eV. Thin-layer chromatographic (TLC) analysis was performed on Kieselgel 60 F_{254} (Merck) plates (silica gel, 0.25 mm layer thickness), with compounds visualized by dipping plates into 10% (v/v) H_2SO_4 reagent (Aldrich, Milwaukee, WI) followed by charring at 110 °C for 5–10 min. Silica gel (Merck 60A, 70–230 or 200–400 mesh ASTM) was used for column chromatography. Preparative TLC was performed on Kieselgel 60 F_{254} (Merck) plates (silica gel, 0.25 mm layer thickness). All solvents used for chromatographic separations were purchased from Fisher Scientific (Fair Lawn, NJ) and distilled before use.

3.2. Plant material

The leaves of *Diplectria becariana* were collected in Bulungan, East Borneo, Indonesia, in October 2000, and identified by S. R. A voucher specimen has been deposited at the Herbarium Bogoriense, Indonesia Institute of Science, Bogor, Indonesia.

3.3. Quinone reductase (QR) induction assay

For the evaluation of plant extracts, fractions, and pure isolates as inducers of QR, cultured mouse Hepa 1c1c7 cells were used as described previously (Chang et al., 1997; Misico et al., 2002). Enzyme activity was expressed as CD, the concentration required to double the specific activity of QR. IC_{50} (half-maximal inhibitory concentration of cell viability) and CI (chemoprevention index, IC_{50}/CD) values were also determined. In the QR assay, plant extracts and chromatographic fractions are considered as inactive when the CD value is >20 μg/mL, while the pure compounds are considered as inactive when the CD value is >10 μg/mL.

3.4. Extraction and isolation

The dried and milled plant material (2.9 kg) was extracted with MeOH (3 × 12 L) by maceration. The extracts were combined and concentrated in vacuo at 40 °C. The concentrated extract was suspended in 90% MeOH and then partitioned with petroleum ether (3 × 2 L) to afford a petroleum ether-soluble syrup (D001) on drying. Next, the aqueous MeOH extract was concentrated and suspended in H_2O (2 L) and partitioned with EtOAc (3 × 2 L) to give an EtOAc-soluble extract (D002) and an aqueous residue (D003). The CD values (μg/mL) of the solvent partitions, D001, D002, and D003 were >20, 9.4, and >20, respectively.

Based on the above activity results, the EtOAc-soluble extract (D002, 60.0 g) was subjected to silica gel CC as stationary phase using a $CHCl_3$–MeOH gradient (from

100:0 to 0:100 v/v) as mobile phase to afford 13 pooled fractions (fractions F004-16). Of these, fractions F010, 11, and 12 showed the most potent QR-inducing activity (CD values 13.1, 13.7, and 11.1 μg/mL, respectively). Fractions F010-12 [eluted with $CHCl_3$–MeOH (19:1 v/v), 5.2 g], were combined, and then applied to a silica gel column (*n*–hexane–EtOAc–MeOH 7:3:0 → 5:4.5:0.5 v/v) resulting in nine subfractions (fractions 17–25). Ursolic acid (37 mg, 0.0013%) was isolated from fraction F018 [eluted with *n*-hexane–EtOAc (7:3 v/v), 310 mg] by recrystallization from MeOH. Fraction F019 [eluted with *n*-hexane–EtOAc (3:2 v/v), 76 mg] was further purified by preparative TLC, developed with benzene–EtOAc (14:1, run three times), to give compounds **3** (5.5 mg, 0.00019%, $R_f = 0.69$) and **2** (2.8 mg, 0.000097%, $R_f = 0.63$), in turn. Fraction F021 [eluted with *n*-hexane–EtOAc (1:1 v/v), 496 mg] was purified over a further silica gel column, with petroleum ether–acetone (7:3 → 1:1 v/v) as solvent system, yielding the new compound beccaridiol (**1**) (5.0 mg, 0.00017%) and 4′-hydroxy-1′,2′-dihydro-β-ionone (10.4 mg, 0.00036%) {$[\alpha]_D^{20}$ −15.8° (*c* 1.0, $CHCl_3$)}.

3.5. Beccaridiol (*1*)

White powder; m.p. 126–128 °C; $[\alpha]_D^{23}$ +58.0° (*c* 0.4, $CHCl_3$); UV ($CHCl_3$) λ_{max} (log ε) 246 (3.78) nm; IR ν_{max} (film) 3381, 1456, 1374, 1216, 1048, 757 cm^{-1}; ^1H and ^{13}C NMR data, see Table 1; LREIMS *m/z* 422 ($[M]^+$, 84), 407 (31), 404 (15), 389 (8), 251 (11), 237 (33), 198 (100), 186 (66), 185 (67), 184 (54), 183 (77); HREIMS *m/z* 422.3213 ($[M]^+$, Calc. for $C_{29}H_{42}O_2$, 422.3185).

3.6. 2-Mono-p-bromobenzoate (*1a*) and 2,3-di-p-bromobenzoate (*1b*) of beccaridiol (*1*)

Beccaridiol (**1**, 1.8 mg) was dissolved in 0.3 mL anhydrous pyridine in a 4 mL vial, and *p*-bromobenzoyl chloride (5.5 mg) was added. After standing at r.t. overnight, the reaction mixture was evaporated to dryness under reduced pressure at ca. 45 °C. The products were then purified by preparative TLC developed with *n*-hexane–EtOAc (3:1), to give the 2-mono-*p*-bromobenzoate (**1a**, 1.1 mg) and the 2,3-di-*p*-bromobenzoate (**1b**, 1.5 mg) derivatives of beccaridiol. **1a**: $[\alpha]_D^{23}$ +5.0° (*c* 0.11, $CHCl_3$); UV ($CHCl_3$) λ_{max} (log ε) 247 (3.83) nm; CD ($CHCl_3$; 23 °C; *c* 0.025 mg/mL) (Δε) 232 (−83.61), 247 (+7.91) nm; ^1H NMR (300 MHz, in $CDCl_3$, TMS) δ 7.92 (2H, *d*, *J* = 8.6 Hz, COC_6H_4-*p*-Br), 7.60 (2H, *d*, *J* = 8.6 Hz, COC_6H_4-*p*-Br), 6.93 (1H, *d*, *J* = 7.4 Hz, H-22), 6.79 (1H, *d*, *J* = 7.4 Hz, H-21), 5.49 (1H, *br t*, *J* = 3.6 Hz, H-12), 5.26 (1H, *ddd*, *J* = 11.0, 10.0, 4.4 Hz, H-2), 3.41 (1H, *d*, *J* = 10.0 Hz, H-2), 2.26, 2.24, 1.16, 1.13, 1.05, 0.96, 0.95 (each 3H, singlet, methyls); **1b**: $[\alpha]_D^{23}$ −60.7° (*c* 0.15, $CHCl_3$); UV ($CHCl_3$) λ_{max} (log ε) 249 (3.88) nm; CD ($CHCl_3$; 23 °C; *c* 0.025 mg/mL) (Δε) 232 (−83.66), 244 (+5.77), 257 (−8.08) nm; ^1H NMR (300 MHz, in $CDCl_3$, TMS) δ 7.84 (2H, *d*, *J* = 8.6 Hz, COC_6H_4-*p*-Br), 7.76 (2H, *d*, *J* = 8.6 Hz, COC_6H_4-*p*-Br), 7.55 (2H, *d*, *J* = 8.6 Hz, COC_6H_4-*p*-Br), 7.50 (2H, *d*, *J* = 8.6 Hz, COC_6H_4-*p*-Br), 6.96 (1H, *d*, *J* = 7.4 Hz, H-22), 6.81 (1 H, *d*, *J* = 7.4 Hz, H-21), 5.51 (2H, *m*, H-2 and H-12), 5.24 (1H, *d*, *J* = 10.4 Hz, H-2), 2.28, 2.27, 1.27, 1.14, 1.09, 1.05, 0.98 (each 3H, singlet, methyls).

3.7. 2′,4′-Dihydroxy-3-(4-methoxyphenyl)-propiophenone (*2*)

Colorless oil; UV (MeOH) λ_{max} (log ε) 316 (3.85), 276 (4.18), 226 (4.17), 215 (4.31) nm; IR ν_{max} (film) 3365, 1631 cm^{-1}; ^1H and ^{13}C NMR data, see Table 1; LREIMS *m/z* 272 ($[M]^+$, 55), 253 (9), 137 (73), 134 (20), 121 (100), 91 (6), 77 (6); HREIMS *m/z* 272.1047 ($[M]^+$, Calc. for $C_{16}H_{16}O_4$, 272.1049).

3.8. 4′-O-Methyldavidigenin (*3*)

Colorless oil; UV (MeOH) λ_{max} (log ε) 316 (3.81), 275 (4.17), 225 (4.16), 216 (4.30) nm; IR ν_{max} (film) 3378, 1630 cm^{-1}; ^1H and ^{13}C NMR data, see Table 1; LREIMS *m/z* 272 ($[M]^+$, 58), 253 (12), 166 (13), 151 (100), 120 (28), 107 (26), 95 (5), 77 (8); HREIMS *m/z* 272.1050 ($[M]^+$, Calc. for $C_{16}H_{16}O_4$, 272.1049).

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