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Two new flavonoids from Derris laxiflora Benth

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ABSTRACT

Two new compounds, derriscoumaronochromone (1) and *cis*-3,4'-dihydroxy- 5,7-dimethoxyflavan (2), as well as *trans*-4'-O-methylcatechin (3) were isolated from *Derris laxiflora*, The structures of these compounds were determined by analysis of their spectroscopic data.

1. Introduction

Derris laxiflora Bentham is a native plant, which grows at the lowlands of Taiwan. The aborigines lived in Taiwan had used the whole plants of D. laxiflora as pesticide (Brooks and Watson, 1985). In addition, the juice of root of D. laxiflora is used as a nonselective piscicide since ancient time. As regard to the phytochemistry study, seven flavonoids, including 3'-methoxylupinifonin, laxifolin, isolaxifolin, laxichalcone, derrichalcone, derriflavanone, and epi-derriflavanone have been isolated and identified from ethanolic extract of D. laxiflora roots (Lin et al., 1991, 1992). Four oleanane-type and one glutinanetype triperpenoids isolated from whole plants of D. laxiflora had been reported by our group (Chiu et al., 2008). To complete understand the chemical constituents, we are continuing to investigate the new compounds from D. laxiflora. Currently, we obtained two new flavonoids, which were namely derriscoumaronochromone (1) and cis-3,4'dihydroxy-5,7-dimethoxyflavan (2), and one known trans-4'-O-methylcatechin (3) (Cren-Olivé et al., 2002) from D. laxiflora (Fig. 1). The structures of these compounds were determined by analysis of their spectroscopic data.

2. Results and discussion

The molecular formula ($C_{17}H_{12}O_7$), UV (λ_{max} 224, 284, and 335 nm) and IR (3375, 1652 and 1500 cm⁻¹) spectra of compound 1 suggested that it contains benzoyl and hydroxyl groups. The absorption at 1652 cm⁻¹ in IR spectrum is a feature of conjugated carbonyl in flavonoid. The molecular formula $C_{17}H_{12}O_7$ of 1 was determined from the molecular ion peaks observed in the EI-MS and by high-resolution EI-MS measurements as well as ¹³C NMR data. From EI-MS spectra, we can observe fragment ion peaks (Scheme 1) at m/z 310 (M⁺-H₂O), 178 (M^+ -150), and 151 (M^+ -177). The above ion peaks in EI-MS are all the features of flavanoid type compounds, and the m/z 151 is the characteristic fragment of A ring (Monache et al., 1995). The ¹H and 13 C NMR (Table 1) spectra of 1 showed signals assignable to a methoxy group δ H 3.81 (s, CH₃O-7), a methylenedioxyl group δ H 5.88 and 5.90 (1H each, d, J = 1.6 Hz, -OCH₂O-), and five aromatic protons including two *para* singlet protons and a set of ABX protons ($\delta_{\rm H}$ 6.50) (s, H-3'), 6.64 (s, H-6'), 6.45 (d, J = 2.4 Hz, H-8), 6.61 (dd, J = 8.8, 2.4 Hz, H-6), 7.73 (d, J = 8.8 Hz, H-5). The lowest-shift of latter phenyl proton discerned that is peri to carbonyl group. Ascribing to two phenoxyl groups crossing position, H-2 exhibited very low field at δ 6.29 (s) in ¹H NMR (δ 110.2 in ¹³C NMR), which is a typical NMR data

¹ Equal contribution to first author.

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Fig. 1. Structures of compounds 1-3

in coumaronochromone skeleton (Zhao et al., 2007). The HMBC (Fig. 2) experiment showed correlations OMe/C-7 confirmed the position of 7-OMe, and another correlations H-2/C-4, C-9, C-1'; H-5/C-4, C-9 established the location of 4-oxo and connections among C-ring, D-ring and B-ring. The signal at $\delta_{\rm C}$ 143.4 in 13 C NMR data was assigned at C-5' (higher field than C-4' at δ 150.4) due to receiving the resonance effect from *para*-position of C-2' O-atom. On the basis of this evidence, the structure of 1 was elucidated as shown (Fig. 1) and named derriscoumaronochromone.

Compound **2** was obtained as a light-yellow solid (mp: 176–177 °C) with a molecular formula of C17H18O5 based on HREIMS analysis. The IR spectrum of **2** showed absorption band at 3356 cnm⁻¹ ascribable to hydroxyl group. The ¹H NMR spectrum of 2 did not show intramolecular hydrogen bonding signal as well as no carbonyl signal in its ¹³C NMR spectrum. Seventeen ¹³C NMR signals, including twelve aromatic carbons, two oxygenated methine carbons and one methylene carbon as well as two methoxy carbons indicated that compound 2 possessed flavan skeleton. The ¹H NMR showed a typical meta-coupled pattern for δ H 6.11 (d, J = 2.0 Hz, H-6) and 6.07 (d, J = 2.0 Hz, H-8) in the A-ring and an A_2X_2 system for the δ H 7.35 (2H, d, J = 8.4 Hz, H-2' and H-6') and 6.80 (2H, d, J = 8.4 Hz, H-3' and H-5') protons in the B-ring. Also two methoxy groups at δ H 3.78 (OMe) and 3.74 (OMe) are unambiguous from the ¹H NMR data. The other four signals were δ H 4.93, 4.21, 2.86, and 2.74, which were proposed the protons of C-ring in flavanoid. From the ¹H-¹H COSY spectrum (Fig. 3) showed the correlations of H-2/H-3; H-3/H-4 and the HMBC signals (Fig. 3) showed H-2/C-1', C-9; H-3/C-10; H-4/C-9. Above evidences pinpointed the δH 4.93 (bs, H-2), 4.21 (bs, H-3), and 2.86 (dd, J = 16.4, 4.8 Hz, H-4), 2.74 (dd, J = 16.4, 2.4 Hz, H-4). The HMBC signals also established the location of 5-OMe and 7-OMe. From the ¹³C NMR spectra showed a hydroxyl group at $\delta_{\rm C}$ 66.5 (C-3). The NOESY (Fig. 4) signals showed the correlations of H-2/H-3; H-2/8H 2.86 (H-4); H-3/8H 2.86 (H-4), 8 2.74(H-4) as well as the coupling constant confirmed that H-2 and δ 2.86 (H-4) were axial and H-3 and δH 2.74 (H-4) were equatorial configuration. On the basis of these data, compound 2 is assigned the structure cis-3,4'-dihydroxy-5,7-dimethoxyflavan, and it is a new flavonoid and is reported here as a new natural product. There are two chiral centers in this compound and 2,3-cis configuration are the same with (+)-epifisetinidol, (2S,3S)-2,3-cis-flavan-3,3',4',7-tetraol

Table 1 ¹H- and ¹³C NMR Data of Compounds 1 and 2 (1 in CDCl₃ and 2 in CD₃COCD₃). δ in ppm, *J* in Hz.

Position	1		2	
	$\delta_{\rm H}$	δ_{C}	$\delta_{\rm H}$	δ_{C}
2	6.29 s	110.2	4.93 bs	79.5
3	-	80.3	4.21 bs	66.5
4	-	188.6	2.86 dd (16.4,4.8), 2.74 dd	29.0
			(16.4,2.4)	
5	7.73 d (8.8)	129.1	-	160.0
6	6.61 dd (8.8,2.4)	111.6 ^a	6.11 d (2.0)	91.9
7	-	167.1	-	160.4
8	6.45 d (2.4)	101.4	6.07 d (2.0)	94.1
9	-	161.2	-	156.8
10	-	111.5 ^a	-	101.8
1'	-	117.7	-	131.1
2′	-	155.4	7.35 d (8.4)	129.0
3′	6.50 s	94.2	6.80 d (8.4)	115.4
4′	-	150.4	-	157.7
5′	-	143.4	6.80 d (8.4)	115.4
6′	6.64 s	103.5	7.35 d (8.4)	129.0
$5-OCH_3$	-		3.78 s	55.7
7-OCH3	-		3.74 s	55.4
OCH_3	3.81 s	55.8		
OCH ₂ O	5.88, 5.90 d (1.6)	101.9		

^a Signals maybe exchange.



Fig. 2. Key HMB correlations $(H \rightarrow C)$ of 1.



Scheme 1. Proposed fragments in MS of compound 1.



Fig. 3. Key HMB ($H \rightarrow C$) and COSY correlations (H — H) of 2.



Fig. 4. Key NOESY (H +++ H) correlations of 2.

(Steenkamp et al., 1988). The specific rotation $[\alpha]_D^{25} - 23.9^\circ$ of **2** means that this compound has 2*R*,3*R* absolutely configuration.

3. Experimental part

3.1. General experimental procedures

Melting points were determined with a Yanagimoto micromelting point apparatus and are uncorrected. IR spectra were recorded on a Perkin-Elmer 983G spectrophotometer. ¹H, ¹³C, DEPT, and two-dimensional NMR spectra were acquired on a Varian Unity Plus 400 MHz (¹H) and 100 MHz (¹³C) spectrometer. EIMS, UV, and specific rotations were determined using a JEOL JMS-HX 300, Hitachi S-3200 spectrometer, and JASCO DIP-180 digital polarimeter, respectively. Extracts were initially fractionated on silica gel (Merck 70–230 mesh, 230–400 mesh, ASTM) and then purified with a semi-preparative normal-phase HPLC column (250 × 10 mm, 7 µm, LiChrosorb Si 60) or a semi-preparative reverse-phase HPLC column (250 × 10 mm, 5 µm, Phenomenex Luna C18 (2)) on an LDC Analytical-III system.

3.2. Plant material

The whole plant of *D. laxiflora* was collected in Taitong County, Taiwan, in December 2001. The plant material was identified by Prof. Shang-Tzen Chang (School of Forestry and Resource Conservation, National Taiwan University), and a voucher specimen was deposited at the herbarium of School of Forestry and Resource Conservation, National Taiwan University, Taipei, Taiwan.

3.3. Extraction and isolation

Air-dried pieces of the whole plant of *D. laxiflora* (11.7 kg) were extracted with MeOH (140 L) by soaking for 1 week each at room temperature two times. The extract was evaporated under vacuum and concentrated in a rotary evaporator to a residue (400 g). The residue was suspended in H_2O and partitioned successively with EtOAc and *n*-

BuOH to yield EtOAc (100 g), *n*-BuOH (83 g), and H_2O (217 g) soluble fractions. The EtOAc-soluble fraction was subjected to chromatography using a Geduran Si-60 (Merck, Darmstadt, Germany) column eluted with EtOAc-*n*-hexane (gradient elution by changing from 5:95 to 100:0) and then acetone to give fractions A (8.7 g), B (10.1 g), C (11.2 g), D (9.3 g), E (8.7 g), F (9.3 g), G (7.5 g), H (4.5 g), I (2.2 g), and J (20.2 g). The fractions were further purified by repeated HPLC using a hexane-EtOAc solvent system (normal phase on LiChrosorb Si 60) and **1** (8.2 mg) and **2** (7.9 mg) were eluted from fraction G with 60% EtOAc in *n*-hexane. And **3** (10.2 mg) was eluted from fraction J using a 90% MeOH in H₂O solvent system on the reverse phase of Phenomenex Luna C18.

3.4. Spectral data

Derriscoumaronochromone, **1:** amorphous solid; $[\alpha]_D^{25}$ - 121..3° (*c* 0.28, CH₃OH); UV MeOH λ max (log ε) nm: 224 (3.90), 284 (3.58), 335 (3.00); IR (film) ν_{max} 3375, 2946, 1652, 1619, 1603, 1500, 1469, 1159, 1128, 1041, 963 cm⁻¹; for ¹H and ¹³C NMR (CDCl₃) spectra, see Table; EI-MS: 328 (30, M⁺), 310 (34), 178 (42), 151 (100); HR-EI-MS: 328.0585 (M⁺, C₁₇H₁₂O₇; calc. 328.0579).

cis-3,4′-dihydroxy-5,7-dimethoxyflavan, **2:** pale yellow solid; mp 176–177 °C; $[\alpha]_{D}^{25}$ -23.9° (*c* 0.43, CH₃OH); UV (MeOH) $\lambda_{max}(\log \varepsilon)$ nm: 226 (4.01), 273 (2.80); IR (film) ν_{max} 3356, 2927, 1623, 1597, 1525, 1499, 1458, 1209, 1147, 1116 cm⁻¹; for ¹H and ¹³C NMR (CDCl₃) spectra, see Table; EI-MS: 302 (53, M⁺), 284 (13), 283 (10), 167 (100); HR-EI-MS: 302.1155 (M⁺, C₁₇H₁₈O₅; calc. 302.1149).

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.phytol.2017.05.004.

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