

Communication

Two New Flavonol Glycosides from *Sarcopyramis bodinieri* var. *delicate*

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Abstract: Detailed chemical investigation of the herb *Sarcopyramis bodinieri* var. *delicate* resulted in the isolation of two new flavonol glycosides, namely, isorhamnetin-3-*O*-(6''-*O*-*E*-feruloyl)- β -D-glucopyranoside (**1**) and isorhamnetin-3-*O*-(6''-*O*-*E*-feruloyl)- β -D-galactopyranoside (**2**). In addition, four known compounds, quercetin-3-*O*-(6''-acetyl)- β -D-glucopyranoside (**3**), isorhamnetin-3-*O*-(6''-acetyl)- β -D-glucopyranoside (**4**), quercetin-3-*O*-(6''-*O*-*E*-*p*-coumaroyl)- β -D-glucopyranoside (**5**), and isorhamnetin-3-*O*-(6''-*O*-*E*-*p*-coumaroyl)- β -D-glucopyranoside (**6**) were obtained. The structures of the new isolates were determined by extensive spectroscopic analysis.

Keywords: *Sarcopyramis bodinieri* var. *delicate*, flavonol glycoside.

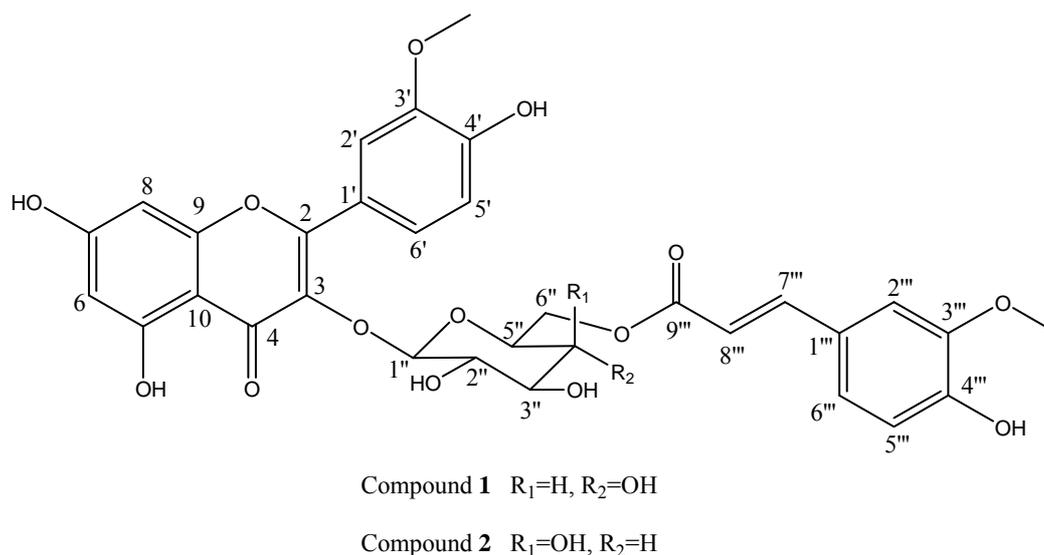
Introduction

Members of the Melastomataceae family are widespread in many regions of the world and particularly in tropical and subtropical regions, mainly in South America and South China. Many species of this family are known by their different use in folk medicine as antioxidant [1],

antihypertensive [2], antihyperglycemic [3], hemostatic [4] and antihepatitis drugs [5]. Antiinflammatory [4], antimicrobial [6] and cytotoxic effects [1] have also been investigated. Cumulative phytochemical studies of Melastomataceous plants have indicated an abundance of tannins [7], polyphenols [7], flavonoids [8], fatty acids, steroids, and free triterpenoids [9]. The great variety of natural compounds found in this family, as well as their pharmacological properties prompted us to study the chemical constituents and bioactivities of *Sarcopyramis bodinieri* var. *delicate*, an endemic and chemically uninvestigated plant distributed widely in South China.

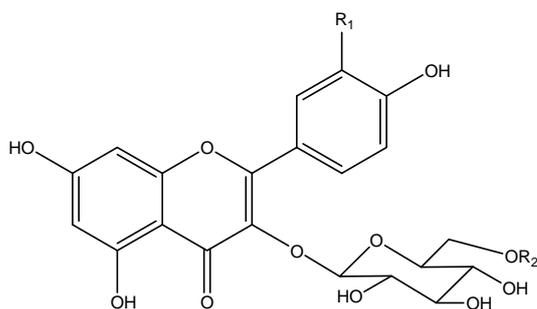
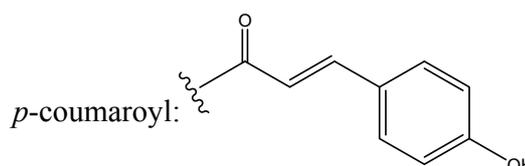
As a rare species, *Sarcopyramis bodinieri* var. *delicate* was widely used as hepatoprotective drug in Fujian province, China. The water extract of this dried herb could reduce aminotransferase and cure choloplania and hepatoma. Detailed fractionation led to the isolation of two new flavonol glycosides, namely, isorhamnetin-3-*O*-(6''-*O*-*E*-feruloyl)- β -D-glucopyranoside (**1**) and isorhamnetin-3-*O*-(6''-*O*-*E*-feruloyl)- β -D-galactopyranoside (**2**), along with four known compounds. The structures of the new isolates were determined by extensive spectroscopic analysis.

Figure 1. The structures of new compounds **1** and **2**.



Results and Discussion

The known compounds **3-6** corresponded to quercetin-3-*O*-(6''-acetyl)- β -D-glucopyranoside (**3**) [10], isorhamnetin-3-*O*-(6''-acetyl)- β -D-glucopyranoside (**4**) [11], quercetin-3-*O*-(6''-*O*-*E*-*p*-coumaroyl)- β -D-glucopyranoside (**5**) [12], and isorhamnetin-3-*O*-(6''-*O*-*E*-*p*-coumaroyl)- β -D-glucopyranoside (**6**) [13, 14]. The structures of these known flavonol glycosides were identified on the basis of extensive spectroscopic data analysis and by comparison of their spectral data with those reported in the literature.

Figure 2. The structures of compounds 3-6.Compound 3 $R_1=OH$, $R_2=COCH_3$ Compound 4 $R_1=OCH_3$, $R_2=COCH_3$ Compound 5 $R_1=OH$, $R_2=p$ -coumaroylCompound 6 $R_1=OCH_3$, $R_2=p$ -coumaroyl

Compound **1** was isolated as a yellow powder. The molecular formula $C_{32}H_{30}O_{15}$ was suggested by a mass spectrum with a $[M+Na]^+$ peak at m/z 677, further confirmed by the adduct ions: 693 $[M+K]^+$, and 1331 $[2M+Na]^+$, combined with the ^{13}C -NMR and DEPT spectra. The IR spectrum of compound **1** revealed the aliphatic and aromatic hydroxyl signals at 3200 – 3550 cm^{-1} . A conjugated carbonyl group (1656 cm^{-1}) and an additional α,β -unsaturated carbonyl ester group (1724 cm^{-1}) were observed in the same region. The absorptions at 3310 and 1057 cm^{-1} indicated the presence of a glycosidic moiety [15].

The 1H -NMR spectrum confirmed many of the above features and revealed a set of isorhamnetin signals, a feruloyl group and a glucopyranose moiety. The presence of isorhamnetin was suggested by the following signals: two doublets at δ_H 6.14 (d, $J = 1.8$ Hz, H-6) and 6.37 (d, $J = 1.8$ Hz, H-8); an ABX spin system due to the aromatic ring at δ_H 6.90 (d, $J = 8.6$ Hz, H-5'), 7.52 (dd, $J = 8.6, 1.9$ Hz, H-6') and 7.87 (d, $J = 1.9$ Hz, H-2'); a methoxyl group singlet at δ_H 3.86 (s, 3'-OMe) with a HMBC correlation with δ_C 147.1 (C-3') [16]. In the 1H -NMR spectrum another set of ABX-type aromatic proton signals at δ_H 6.75 (d, $J = 8.1$ Hz, H-5'''), 6.88 (dd, $J = 8.1, 1.2$ Hz, H-6''') and 7.20 (d, $J = 1.2$ Hz, H-2''') were observed, together with an additional HMBC crosspeak between δ_H 3.82 (s, 3'''-OMe) and δ_C 147.9 (C-3'''). Two olefinic protons with a *trans* coupling constant ($J = 15.8$ Hz) at δ_H 6.22 (d, H-8'''), 7.34 (d, H-7'''), which confirmed the presence of an α,β -unsaturated carbonyl ester group. The feruloyl structure was then deduced from the HMBC correlation from δ_H 6.22 (H-8''') to δ_C 126.4 (C-1''') and the crosspeaks between H-2'''/C-7''' and H-6'''/C-7'''. Detailed analyses of the 1H - (δ 5.53, d, $J = 7.8$ Hz, H-1'') and ^{13}C -NMR (δ 102.8, 77.8, 76.1, 76.0, 71.8, 64.2) suggested glucopyranose as the sugar moiety. A downfield shift of C-6'' was from δ_C 61.8 to 64.2, and an upfield shift of C-5'' was from δ_C 76.8 to 76.0, which were in accordance with the acylation of C-6'' of the glucose moiety [17]. Moreover, the downfield shift of H-2-6'' to 4.11 (dd, $J = 6.8, 11.5$ Hz) and 4.19 (dd, $J = 2.1, 11.5$ Hz) further confirmed the presence of a C-6'' feruloyl in compound **1** [18]. Consequently, the structure of compound **1** was established as isorhamnetin-3-*O*-(6''-*O*-*E*-feruloyl)- β -D-glucopyranoside.

Compound **2**, a yellow powder, shared the same molecular formula $C_{32}H_{30}O_{15}$ with **1**, according to the $[M+Na]^+$ peak at m/z 677 and $[M+K]^+$ peak at m/z 693. Moreover, its NMR data is very similar to those of **1**. Detailed comparison of the ^{13}C -NMR and HMQC spectra between the two compounds indicated that the major difference was in the glycoside moiety. The carbon signals at δ_C 103.4 (C-1''), 71.2 (C-2''), 74.6 (C-3''), 69.5 (C-4''), 74.3 (C-5''), and 62.9 (C-6'') revealed a galactopyranoside

moiety [16]. The structure of compound **2** was therefore assigned as isorhamnetin-3-*O*-(6''-*O*-*E*-feruloyl)- β -D-galactopyranoside.

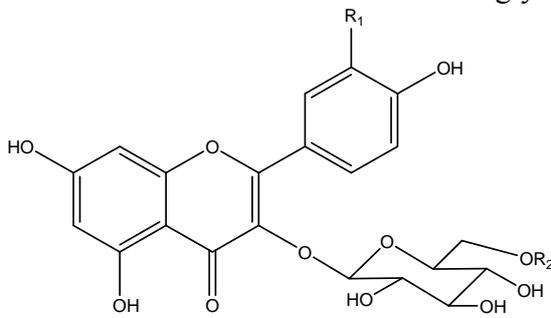
Table 1. The ^1H and ^{13}C -NMR data of compounds **1** and **2** (DMSO- d_6).

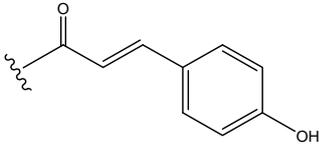
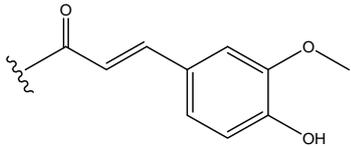
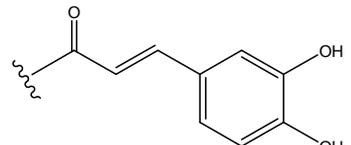
No.	Compound 1		Compound 2	
	H (J_{Hz})	C	H (J_{Hz})	C
2		157.6		157.5
3		134.3		134.3
4		178.6		178.5
5		162.3		162.2
6	6.14 d (1.8)	99.6	6.15 d (1.8)	99.5
7		165.1		165.1
8	6.37 d (1.8)	94.9	6.37 d (1.8)	94.7
9		157.6		157.5
10		104.9		104.9
1'		122.3		122.2
2'	7.87 d (1.9)	114.2	7.99 d (1.9)	114.5
3'		147.1		147.1
4'		150.7		150.6
5'	6.90 d (8.6)	116.1	6.89 d (8.6)	116.2
6'	7.52 dd (8.6, 1.9)	122.9	7.50 dd (8.6, 1.9)	122.9
1''	5.53 d (7.8)	102.8	5.52 d (7.8)	103.4
2''	3.26 m	76.1	3.20 m	71.2
3''	3.28 m	77.8	3.48 m	74.6
4''	3.62 m	71.8	3.70 m	69.5
5''	3.43 m	76.0	3.74 m	74.3
6''	4.11 dd (6.8, 11.5) 4.19 dd (2.1, 11.5)	64.2	4.12 dd (7.0, 11.8) 4.24 dd (2.1, 11.8)	62.9
1'''		126.4		126.4
2'''	7.20 d (1.2)	112.1	7.19 d (1.2)	111.7
3'''		147.9		147.8
4'''		150.3		150.4
5'''	6.75 d (8.1)	116.4	6.78 d (8.0)	116.4
6'''	6.88 dd (8.1, 1.2)	123.8	6.88 dd (8.0, 1.2)	124.0
7'''	7.34 d (15.8)	145.9	7.34 d (15.8)	146.0
8'''	6.22 d (15.8)	115.1	6.26 d (15.8)	113.8
9'''		167.5		167.2
3'-OMe	3.86	56.9	3.87	56.8
3'''-OMe	3.82	56.6	3.78	56.2

^1H and ^{13}C -NMR spectra were obtained at 600 and 150 MHz, respectively.

The UV spectra of the different flavonol glycosides showed an interesting phenomenon (see Table 2) according to our experiments. The substituted fraction on the glycosidic moiety could be characterized by the absorption over 300 nm. For example, the absorption maxima at 315 nm means a *p*-substituted aromatic ring in R₂, while the absorption at 330-336 nm indicates a tri-substituted aromatic ring in the same position.

Table 2. The UV data for different flavonol glycosides.



Compound		UV (MeOH) λ_{\max}	
R ₁	R ₂		
OH	H	356	258
OCH ₃	H	354	256
OH	COCH ₃	357	256
OCH ₃	COCH ₃	355	256
H		315	266
OH	the same as above	315	263
OCH ₃	the same as above	315	259
OH		333	254
OCH ₃		336	252

Experimental

General

The IR spectra were determined on a Thermo Nicolet Nexus 470 FT-IR spectrometer. Optical rotations were measured with a Perkin-Elmer 243 B polarimeter using a 1 dm microcell. The ¹H-NMR and ¹³C-NMR spectra were recorded on a Bruker Avance-600 FT NMR spectrometer. ESI-MS were recorded on a PE Q-STAR ESI-TOF-MS/MS spectrometer. Column chromatography was carried with silica gel (200-300 mesh), and HF₂₅₄ silica gel for TLC was obtained from Qingdao Marine Chemistry

Co. Ltd., Qingdao, People's Republic of China. ODS and Sephadex LH-20 (18-110 μm) were obtained from Pharmacia Co.

Extraction and Isolation

The specimen of *Sarcopyramis bodinieri* var. *delicate* was collected from Fujian Province, P.R. China, in April 2007. A voucher specimen (RSC07) is deposited at the Department of Pharmacy, School of Medical, Xiamen University. The air-dried plant material (5 kg) was ground and extracted exhaustively by maceration at room temperature with EtOH-H₂O (70:30, 20 L \times 3). The concentrated total extract (1.8 kg) was extracted with petroleum ether, CHCl₃, EtOAc and *n*-BuOH, respectively. Part of the EtOAc portion (SBC, 45 g) was suspended in H₂O (2 L), and the filter layer was then subjected to D101 macroporous adsorption resin column, eluted with an equivalent H₂O-EtOH stepwise gradient to obtain 5 fractions. Fraction 3 (SBC-C, 13.76 g) was subjected to a Sephadex LH-20 eluted with MeOH-H₂O (3:1) to give 6 fractions. Fr. SBC-C3 was subjected to Sephadex LH-20 column and then the repeated silica gel column to give compounds **3** (16.7 mg), **4** (33.4 mg), **5** (18.2 mg), **6** (7.5 mg). Compounds **1** and **2** were repurified from SBC-C3C by semi-preparative reversed-phased HPLC (45% MeOH-H₂O, YMC-Pack Pro C-18, 20 \times 150 mm) to give 4.6 mg and 5.8 mg, respectively.

Compound 1: $[\alpha]_D^{25} = -48.6$ (*c* 0.67, MeOH); UV (MeOH) λ_{max} (log ϵ) 251 (4.53), 333 (3.21) nm; IR (neat) ν_{max} 3550, 1724, 1656, 1057 cm^{-1} ; for ¹H and ¹³C-NMR see Table 1; ESI-TOF MS (*m/z*): 677 [M+Na]⁺, 693 [M+K]⁺, 1331 [2M+Na]⁺.

Compound 2: $[\alpha]_D^{25} = -64.2$ (*c* 0.24, MeOH); UV (MeOH) λ_{max} (log ϵ) 249 (4.47), 334 (3.20) nm; for ¹H and ¹³C-NMR see Table 1; ESI-TOF MS (*m/z*): 677 [M+Na]⁺, 693 [M+K]⁺.

Acknowledgements

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Sample Availability: Samples of the compounds are available from the authors.