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Two New Iridoid Glucosides from *Paederia scandens* (Lour.) Merr. var. *mairei* (Léveillé) Hara

Hideaki OTSUKA

Institute of Pharmaceutical Sciences, Hiroshima University Faculty of Medicine, 1-2-3 Kasumi, Minami-ku, Hiroshima 734-8551, Japan

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From aerial parts of *Paederia scandens* var. *maeirei*, an iridoid (6-β-*O*-synapoyl scandoside methyl ester) and a dimeric iridoid (dimer of paederoside) glucosides were isolated along with known iridoid glucosides, paederoside and scandoside methyl ester. Their structures were elucidated by spectroscopic analyses.

Key words: *Paederia scandens* var. *mairei*; Rubiaceae; iridoid glucoside; paederoscandoside

It is well known that *Paederia scandens* (Lour.) Merr. var. *mairei* (Léveillé) Hara produces a sulfur containing iridoid glucoside, namely paederoside and the sulfur form in the iridoid was discussed previously.[1,2] Reinvestigation of the same plant afforded new iridoid glucoside (**1**) and dimeric (**2**) iridoid glucosided, named as paederoscandoside, together with known iridoid glucosides (**3**,**4**).

From a *n*-BuOH-soluble fraction of a methanol extract, two new (**1**,**2**) and two known (**3**,**4**) iridoid glucosides were isolated using various kinds of chromatography. Structures of known compounds were found to be paederoside (**3**)[2] and scandoside methyl ester (**4**).[3] This paper deals with the structural elucidation of the new compounds.

Compound **1** was isolated as an amorphous powder and formulated as $C_{28}H_{34}O_{15}$ on the basis of negative-ion high resolution (HR) FAB-MS. The ^{13}C-NMR spectrum of **1** showed a close resemblance to those of (**4**) except for the presence of sinapinic acid moiety (Table 1). The aromatic ring of the acyl portion was substituted symmetrically with one hydroxyl and two methoxyl groups and a *trans* double bond was in the side chain. Thus, the structure of the acyl group was elucidated to be sinapinic acid. The place of ester linkage of the acyl group was presumed to be the hydroxyl group at the 6-position of **4**, since H-6 proton was obviously shifted downfield (δ_H 5.86), when compared with that of **4**. When compared the ^{13}C-NMR chemical shifts of the C-6 positions for **4** (δ_C 82.5) and its 6-epimer, namely deacetylasperulosidic acid methyl ester (δ_C 75.4),[3] compound **1** was clearly shown to have the hydroxyl group in the β-position on C-6 (δ_C 83.8, with a slight downfield shift by acylation from δ_C 82.5. Therefore, the structure of **1** was elucidated to be 6-*O*-sinapinoyl scandoside methyl ester.

Paederoscandoside (**2**) was also isolated as an amorphous powder and the elemental composition was analyzed to be $C_{36}H_{44}O_{22}S_2$ by negative-ion HR-FAB-MS. The IR spectrum showed absorption bands due to hydroxyl groups and conjugated ester moieties. The ^{13}C-NMR spectrum showed the presence of 36 signals. Two glucopyranoses were presumed to be in the molecule and one of which must have an ester linkage at the 6-position (δ_C 64.5). Of the remaining signals, 12 were reasonably assigned to those of the iridoid moiety of co-occurring paederoside (Part A). From the results of HR-FAB-MS, the other part (Part B) must have the same

elemental composition of non-hydrogen atoms as Part A and from the ^{13}C-NMR spectrum, Part B has almost the same functional groups as those of Part A. These results indicated that **2** was presumed to be a dimer of paederoside. The carboxyl group of Part B must formed an ester linkage with the 6'-OH of Part A. This was confirmed by HMBC spectroscopy in which carbonyl carbon (δ_C 168.6) crossed the H_2-6' protons and this was also supported by the upfield shift of the H-6 proton (δ_H 4.86) of Part B from that (δ_H 5.57) in Part A. With a combination of other two dimensional NMR spectra, the structure was elucidated to be **2** in Fig. 1.

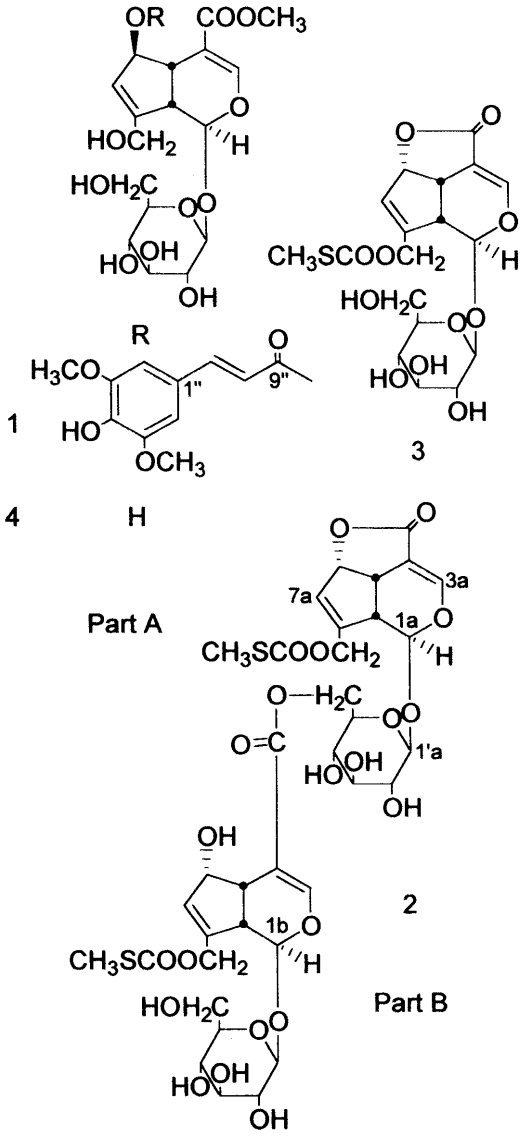

Fig. 1. Structures of **1**-**4**.

Table 1. ^{13}C-NMR Data for **3** and **4**.

	1		**2**	
	C		Part A	Part B
1	98.1	1a,1b	93.2	101.5
3	154.1	3a,3b	150.2	155.8
4	110.1	4a,4b	106.3	108.1
5	42.4	5a,5b	37.5	42.7
6	83.8	6a,6b	86.3	75.6
7	127.3	7a,7b	130.0	132.5
8	150.5	8a,8b	143.6	145.6
9	47.0	9a,9b	45.3	46.4
10	61.0	10a,10b	64.5	66.3
11	168.8	11a,11b	172.4	168.6
-OCH_3	52.0	-SCH_3	13.6, 13.8	
		-COS-	172.7	172.9
1'	100.4	1'a,1'b	100.0	100.8
2'	74.9	2'a,2'b	74.7	75.0
3'	78.5	3'a,3'b	77.9	77.8
4'	71.6	4'a,4'b	71.6	71.8
5'	78.0	5'a,5'b	75.9	78.6
6'	62.8	6'a,6'b	64.5	63.1
1"	126.8			
2",5"	107.1			
3",6"	149.6			
4"	138.2			
7"	147.1			
8"	116.3			
9"	169.1			
-OCH_3×2	57.0			

EXPERIMENTAL

General Procedure The following instruments were used to record physical data. Optical rotations: Union Giken PM-101 digital polarimeter; IR: Horiba FT-710 spectrophotometer, UV: JASCO V-520 UV/VIS spectrophotometer; NMR: JEOL JNM α-400 spectrometer at 400 MHz for ^{1}H and 100 MHz for ^{13}C-NMR spectra with TMS as an internal standard; HR-FAB-MS: JEOL JMS SX-102 mass spectrometer in a negative-ion

mode with PEG-400 or 600 as calibration matrices.

Plant material Aerial parts of *Paederia scandens* var *mairei* were collected in the outskirts of Hiroshima City in June (1996) and a voucher specimen was deposited in the Herbarium of Hiroshima University Faculty of Medicine (96-PSM-Hiroshima-0606).

Extraction and Isolation Air-dried material (4.30 kg) was extracted with MeOH (30 l×2). The MeOH extract was partitioned in a similar manner to that reported to give 52.0 g of a *n*-BuOH-soluble fraction.[4] The *n*-BuOH soluble fraction (50.0 g) was subjected to highly porous synthetic resin column chromatography (Diaion HP-20, Mitsubishi Chemical Co. Ltd., Φ = 50 mm, L = 40 cm) using H_2O-MeOH (4:1, 3 l), (2:3, 3 l), (3:2, 3 l) and (1:4, 3 l), and MeOH (3 l), 500 ml-fractions were collected. The residue (13.7 g in fractions 4–8) of the 20% MeOH eluent was subjected to silica gel (400 g) column chromatography, eluting with $CHCl_3$ (1 l) and $CHCl_3$-MeOH [(49:1, 3 l), (24:1, 3 l), (47:3, 3 l), (23:2, 3 l), (9:1, 3 l), (7:1, 3 l), (17:3, 3 l), (4:1, 3 l) and (3:1, 3 l)], 500 ml-fractions were collected. Combined fractions 33–41 (2.45 g) were then separated by reversed-phase open column chromatography (RPCC) [ODS: Cosmosil 75C_{18}-OPN (Nakalai Tesque, Kyoto, Japan), Φ = 50 mm, L = 25 cm, linear gradient, MeOH-H_2O (1:9, 1 l)→(1:1, 1 l), 10 g-fractions were collected]. The residue (196 mg) in fractions 36–60 was subjected to droplet counter-current chromatography (DCCC) [Chromatograph (Tokyo Rikakikai, Tokyo, Japan) was equipped with 500 glass columns (Φ = 2 mm, L = 40 cm) and the lower and upper layers of the solvent mixture of $CHCl_3$-MeOH-H_2O-*n*-PrOH (9:12:8:2) were used for the stationary and mobile phases, respectively. Five-gram fractions were collected and numbered according to their order of elution of the mobile phase.] to give 89 mg of **4** in fractions 33–38.

The residue (12.2 g in fractions 14–18), obtained on Diaion HP-20 column chromatography was similarly subjected to silica gel (400 g) column chromatography with the same solvent system. The residue (3.04 g) in fractions 35–45 was separated by RPCC to give 1.10 g of paederoside (**3**) in a crystalline state. The residue (706 mg) in fractions 46–51 was subjected to DCCC to give a fraction, enriched by compound **1**. This was finally purified by preparative HPLC [ODS (Inertsil, Φ = 6 mm, L = 25 cm, GL Science, Tokyo, Japan), MeOH-H_2O (2:3), 1.6 ml/min, detection: refractive index] to give 10 mg of **1**. The residue (1.52 g) in fractions 52–60 was subjected to RPCC (83 mg in fractions 192–200) and then DCCC to give 26 mg of **2** in fractions 79–92.

6-*O*-Sinapinoyl Scandoside Methyl Ester (1) Amorphous powder. $[\alpha]_D^{22}$ –71.2° (*c*=0.66, MeOH). UV λ_{max} (MeOH) nm (log ε): 230 (4.22), 322 (3.88). ^{1}H-NMR (CD_3OD) δ: 3.08 (1H, t, *J*=7 Hz, H-9), 3.66 (3H, s, -$COOCH_3$), 3.88 (6H, s, -OCH_3×2), 4.22 (1H, br d, *J*=6 Hz, H-10a), 4.39 (1H, d, *J*=6 Hz, H-10b), 4.70 (1H, d, *J*=8 Hz, H-1'), 5.31 (1H, d, *J*=7 Hz, H-1), 5.68 (1H, dt, *J*=5, 2 Hz, H-7), 5.86 (1H, t, *J*=2 Hz, H-6), 6.40 (1H, d, *J*=6 Hz, H-8''), 6.92 (2H, s, H-2'' and 6''), 7.51 (1H, d, *J*=1 Hz, H-3), 7.63 (1H, d, *J*=16 Hz, H-7''), other sugar protons and H-5 were buried in envelopes of strong signals. ^{13}C-NMR (CD_3OD): Table 1. HR-FAB-MS (negative-ion mode) *m/z*: 609.1844 $[M-H]^-$ (Calcd for $C_{28}H_{33}O_{15}$: 609.1819).

Paederoscandoside (2) Amorphous powder. $[\alpha]_D^{22}$ –53.9° (*c*=1.65, MeOH). IR ν_{max} (KBr) cm^{-1}: 3422, 2931, 1744, 1709, 1658, 1633, 1156, 1073. UV λ_{max} (MeOH) nm (log ε): 234 (4.13). ^{1}H-NMR (CD_3OD) δ: Part A, 2.33 (3H, s, -SCH_3), 3.24 (1H, dd, *J*=9, 8 Hz, H-2'a), 3.39 (1H, t, *J*=9 Hz, H-3'a), 3.62 (1H, ddd, *J*=10, 7, 2 Hz, H-5'a), 3.68 (1H, td, *J*=7, 2 Hz, H-5a), 4.19 (1H, dd, *J*=12, 7 Hz, H-6'aa), 4.65 (1H, dd, *J*=12, 2 Hz, H-6'ab), 4.80 (1H, d, *J*=14 Hz, H-10aa), 4.84 (1H, d, *J*=14 Hz, H-10ab), 5.57 (1H, d, *J*=6 Hz, H-6a), 5.76 (1H, s, H-7a), 5.82 (1H, d, *J*=1 Hz, H-1a), 7.30 (1H, d, *J*=2 Hz, H-3a), H-9a and H-4'a were in the solvent signal. Part B, 2.33 (3H, s, -SCH_3), 2.66 (1H, br t, *J*=9 Hz, H-9b), 3.07 (1H, ddd, *J*=8, 6, 2 Hz, H-5b), 4.73 (1H, d, *J*=8 Hz, H-1'b), 3.25 (1H, dd, *J*=9, 8 Hz, H-2'b), 3.42 (1H, t, *J*=9 Hz, H-3'b), 3.35 (1H, t, *J*=9 Hz, H-4'b), 3.64 (1H, dd, *J*=12, 6 Hz, H-6'ba), 3.87 (1H, dd, *J*=12, 2 Hz, H-6'bb), 4.86 (1H, dd, *J*=6, 2 Hz, H-6b), 4.96 (1H, br d, *J*=15 Hz, H-10ba), 5.07 (1H, d, *J*=9 Hz, H-1b), 5.08 (1H, d, *J*=15 Hz, H-10bb), 6.03 (1H, d, *J*=1 Hz, H-7b), 7.70 (1H, d, *J*=1 Hz, H-3b), H-5b was in the solvent signal. ^{13}C-NMR (CD_3OD): Table 1. HR-FAB-MS (negative-ion mode)

m/z: 891.1708 $[M-H]^-$ (Calcd for $C_{36}H_{43}O_{22}S_2$: 891.1687).

Known compounds Paederoside (**3**),[1] colorless needles, mp. 120-122℃, $[\alpha]_D^{22}$ –174° (*c*=0.72, MeOH), Scandoside methyl ester (**4**),[3] amorphous powder, $[\alpha]_D^{22}$ –46.0° (*c*=0.87, MeOH).

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