An Iridoid Gentiobioside, a Benzophenone Glucoside and Acylated Flavone *C*-Glycosides from *Tripterospermum japonicum* (Sieb. *et* Zucc.) Maxim

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From aerial parts of *Tripterospermum japonicum*, 6′-*O*-β-D-glucopyranosylmorroniside, benzophenone glucoside, named triptephenoside and 2‴- and 4‴-*O*-acetyl-2″-*O*-α-L-rhamnopyranosylisovitexins were isolated, along with known iridoid and secoiridoid glucosides, and C-glycosyl flavones.

Key words *Tripterospermum japonicum*; Gentianaceae; *seco*-iridoid glucoside; 6′-*O*-glucopyranosylmorroniside; benzophenone glucoside; triptephenoside

In a previous paper, we reported the isolation of several xanthone glucosides, named triptexanthosides A—E from aerial parts of *Tripterospermum japonicum*.[1] Further phytochemical investigation of the title plant afforded new glucosides of *seco*-iridoid (**1**), benzophenone (**2**) and C-glycosyl flavones (**3**, **4**), along with known compounds, loganic acid (**5**),[2] secologanoside (**6**),[3] morroniside (**7**),[4] sweroside (**8**),[5] 7α- and 7β-methoxyswerosides (**9**, **10**),[6] isovitexin (**11**),[7] isoorientin (**12**),[8] 2″-*O*-α-L-rhamnopyranosylisovitexin (**13**)[8] and 2″-*O*-β-D-xylopyranosylisovitexin (**14**).[8] This paper describes structural elucidation of these new compounds.

Various kinds of chromatographic techniques were employed for separation of the glycosides (see Experimental). Structures of known compounds were determined by comparison of reported spectral data.

Compound **1**, $[\alpha]_D$ −71.4°, was obtained as an amorphous powder whose elemental composition was determined to be $C_{23}H_{36}O_{16}$ by negative-ion high-resolution (HR)-FAB-MS. The UV absorption maximum at 238 nm was attributed to that of an α,β-unsaturated ketone function. The ^{1}H-NMR spectrum resembled that of morroniside (**7**), which co-occurred in this plant,[4] although signals between δ_H 3.1—3.6 showed more complexity. The ^{13}C-NMR spectrum (Table 1) showed the presence of two close siganls at δ_C 69.83 and 70.31 for the 6′-positions of the inner glucopyranose due to an epimeric mixture of the aglycone, and δ_C 62.82 for the 6″-position of the terminal glucopyranose. The gentiobiose-type linkage was confirmed by heteronulear multiple bond connectivity spectrum (HMBC) in which cross peaks were observed between δ_H 4.13 (β, H-6′) and 4.17 (α, H-6′), and δ_C 104.81 (β, C-1″) and 105.03 (α, C-1″), respectively. Anomeric protons, δ_H 4.50 (α, H-1″) and 4.54 (β, H-1″) also crossed C-6′ carbon signals, δ_C 70.31 (α) and 69.83 (β), respectively. Other signals from the aglycone and sugar moieties also appeared as sets of close peaks in the ratio of 3 (α-isomer) : 2 (β-isomer) like those of morroniside.[4] These results indicated that the structure of **1** was 6′-*O*-β-D-glucopyranosylmorroniside.

Compound **2**, named triptephenoside, $[\alpha]_D$ −54.5°, was isolated as an amorphous powder whose elemental composition was determined to be $C_{21}H_{24}O_{10}$. Six signals assignable to β-glucopyranose were observed in the ^{13}C-NMR spectrum and of the remaining 11 signals, two of which had double strength, 12 carbons out of 13 must constitute tetrasubstituted and disubstituted aromatic rings and the one remaining apparently forms a carbonyl function from its chemical shift (δ_C 197.4). These functionalities were similar to those of xanthone glucoside. However, the degree of unsaturation calculated from the results of HR-FAB-MS did not satisfy the xanthone glucoside skeleton, *i.e.* there was a shortage of one degree. In the ^{1}H-NMR spectrum, one signal (δ_H 3.63) for two methoxyl groups and one aromatic signal for two protons were observed. These constituted a symmetrically substituted aromatic ring together with a hydroxyl group (δ_C 162.4) (ring A). The highly shielded signal at δ_C 93.0 indicated that both vicial positions were occupied by methoxyl (or hydroxyl) substituents, while four coupled aromatic protons formed another with a glucosyloxyl group (δ_C 159.3) (ring B). Acetylation of 2 gave pentaacetate (**2a**) which contained four acetyl groups on aliphatic alcohols and one on a phenolic alcohol in the ^{1}H-NMR spectrum. Based on this evidence, this compound was expected to be a glucopyranoside of a benzophenone derivative with one free phenolic alcohol. The position of the glucosyloxyl group in ring B was deter-

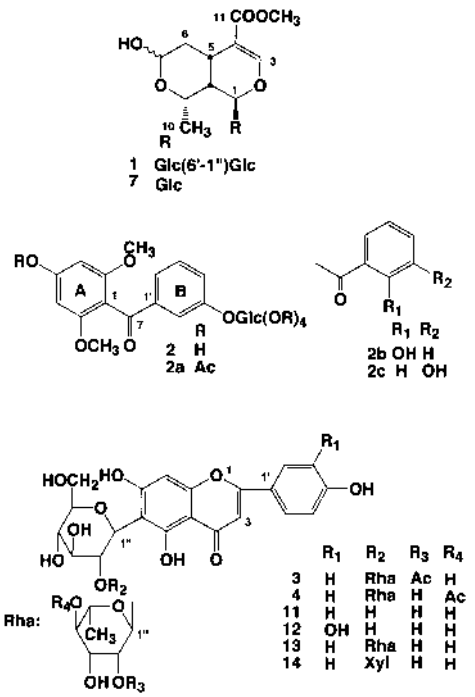

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Table 1. NMR Data for Morroniside (**7**) and 6′-*O*-*β*-D-Glucopyranosylmorroniside (**1**) (CD_3OD)[a]

Carbon No.	**7** (C)		**1** (C)		**1** (H)	
	α	*β*	*α*	*β*	*α*	*β*
1	97.12	95.67	97.25	96.21	5.90 (d, 9)	5.92 (d, 10)
3	154.51		154.64	154.61	7.52 (s)	7.51 (s)
4	110.94	111.81	110.95	111.81	—	—
5	32.07	27.55	32.04	27.46	2.80 (dt, 4, 13)	3.10 (dt, 5, 13)
6	37.32	34.66	37.41	34.41	1.24 (dt, 10, 13)	1.56 (dt, 4, 13)
					2.00 (ddd, 2, 4, 13)	1.85 (br dd, 4, 13)
7	95.96	92.48	96.49	92.26	4.77 (dd, 2, 10)	5.30 (br d, 4)
8	75.10	65.97	74.99	65.85	3.93 (dq, 2, 7)	4.51 (dq, 3, 7)
9	39.94	40.61	40.13	40.72	1.75 (ddd, 2, 4, 9)	1.78 (m)
10	19.89	19.85	20.27		1.38 (d, 7)	1.30 (d, 7)
11	168.72	168.77	168.81			
–OCH_3	51.81	51.74	51.78	51.70	3.70 (s)	3.69 (s)
1′	100.09	100.14	100.20	100.26	4.78 (d, 8)	4.79 (d, 8)
2′	74.18		74.28	74.99	[d]	[d]
3′	78.54		78.06[b]	78.35[b]	[d]	[d]
4′	71.68	71.74	72.14	72.10	[d]	[d]
5′	78.03		77.55[c]	77.70[c]	[d]	[d]
6′	62.84	62.91	70.31	69.83	3.76 (dd, 7, 12)	3.81 (dd, 7, 12)
					4.17 (dd, 2, 12)	4.13 (dd, 2, 12)
1″			105.03	104.81	4.50 (d, 8)	4.54 (d, 8)
2″			75.31	75.42	[d]	[d]
3″			78.31[b]	78.01[b]	[d]	[d]
4″			71.78	71.74	[d]	[d]
5″			77.88[c]	77.84[c]	[d]	[d]
6″			62.82		[d]	[d]
					[d]	[d]

a) Letters and figures in parentheses are multiplicities and coupling constants in Hz, respectively *b*,*c*) Assignments may be interchanged in each column. *d*) Many signals overlap each other.

Table 2. ^{13}C-NMR Data for Triptephenoside (**2**) and Reference Compounds (**2b**, **2c**) (CD_3OD)

Carbon number	**2**	**2b**	**2c**
1	110.3		
2	160.4		
3	93.0		
4	162.4		
5	93.0		
6	160.4		
7	197.4		
1′	141.3	1 120.9	139.7
2′	118.5	2 163.1	115.5
3′	159.3	3 118.8	158.9
4′	122.7	4 137.4	120.9
5′	130.5	5 120.1	130.8
6′	124.3	6 132.2	121.5
		7 206.3	200.6
		8 26.8	26.8
–OCH_3	56.2×2		
1″	102.6		
2″	74.8		
3″	78.1[a]		
4″	71.0		
5″	78.0[a]		
6″	62.2		

a) Assignments may be interchanged.

mined by the difference NOE experiment, simultaneously when location of the glycosidic linkage was being sought. On irradiation of the anomeric proton, two aromatic protons [δ_H 7.31 (H-4′) and 7.44 (H-2′)] showed significant enhancement. These results revealed that the glycosidic linkage was at the hydroxyl group on the B ring and in turn, the hydroxyl group must be at the 3′-position. To confirm this, the ^{13}C-NMR data of the B ring were compared with those of **2** and 3-hydroxyacetophenones (**2b**, **2c**) (Table 2). The chemical shift of the 1′-position in **2** at δ_C 141.3 (s) was diagnostically closer to that observed in **2c** (δ_C 120.9 and 139.7 in **2b** and **2c**, respectively). Therefore, the structure of **2** was elucidated to be 2,6-dimethoxy-4-hydroxyphenyl 3′-hydroxyphenyl ketone 3′-*O*-*β*-glucopyranoside (**2**). When both the hydroxyl groups at C-2 and 6 of the A ring of benzophenone were methylated, such substitution forced its derailment from the biosynthetic route to xanthone derivatives.

Compound **3** was obtained as an amorphous powder. The elemental composition was determined to be $C_{29}H_{32}O_{15}$. The ^{1}H- and ^{13}C-NMR spectra were similar to those of 2″-*α*-L-rhamopyranosylvitexin (**13**), except for the presence of an acetoxyl group and slight modification of C-1‴, 2‴, 3‴ and 4‴ signals.[8] Therefore, **3** was believed to be an acetyl derivative of 2″-*O*-*α*-L-rhamnopyranosylisovitexin. The position of the acetoxyl group was determined to be 2‴ by application of an acylation induced shift trend in the ^{13}C-NMR spectrum[9] (Table 3), and this was also confirmed by a significant downfield shift of H-2‴ proton at δ_H 4.97 which was crossed with the anomeric proton (H-1‴) in the H–H correlation spectroscopy.

Compound **4** was also isolated as an amorphous powder and the elemental composition was the same as that of **3**. The NMR spectra indicated that **4** was a positional isomer in its acetoxyl group. The position of the acetoxyl group was similarly determined to be at the 4‴-position using an acylation induced shift trend in the ^{13}C-NMR spectrum (Table 3) and a significant downfield shift of H-4‴ proton at δ_H 4.63 in the

Table 3. ^{13}C-NMR Data for Flavone *C*-glycosides (**13**, **3**, **4**) (DMSO-d_6, 90 °C)

Carbon number	**13**	**3**	**4**
2	163.6	163.5	163.5
3	103.0	103.0	103.0
4	181.9	181.8	181.8
4a	103.8	103.6	103.7
5	161.2	161.2	161.3
6	108.9	108.8	108.9
7	163.0	*a)*	163.1
8	93.9	94.0	93.8
8a	156.6	156.6	156.6
1′	121.4	121.4	121.3
2′	128.3	128.2	128.2
3′	116.1	116.1	116.2
4′	161.2	162.2	161.2
5′	116.1	116.1	116.2
6′	128.3	128.2	128.4
1″	71.7	71.5	71.7
2″	79.8	79.7	79.8
3″	75.8	75.6	74.6
4″	70.9	71.0	70.7
5″	81.1	81.1	81.2
6″	61.6	61.5	61.4
1‴	100.6	97.8 (−2.8)*b)*	99.6
2‴	70.7	74.6 (+3.9)	70.7
3‴	70.7	68.7 (−2.0)	68.1 (−2.6)*c)*
4‴	72.0	73.3	74.6 (+2.6)
5‴	68.2	68.2	65.5 (−2.7)
6‴	17.5	17.5	17.3
$\underline{C}H_3CO$–		20.3	20.7
$CH_3\underline{C}O$–		169.5	169.3

a) Not observed. *b*) $\Delta\delta$ **3**−**13**. *c*) $\Delta\delta$ **4**−**13**.

^{1}H-NMR spectrum.

Experimental

Optical rotations were measured on a Union Giken PM-101 digital polarimeter. IR and UV spectra were obtained on Shimadzu IR-408 and UV-160A spectrophotometers, respectively. ^{1}H- and ^{13}C-NMR were taken on a JEOL JNM α-400 spectrometer at 400 and 100 MHz, respectively, with tetramethylsilane (TMS) as an internal standard. Negative and positive-ion HR-FAB-MS and electron impact (EI)-MS (70 eV) were taken on a JEOL JMS SX-102 spectrometer.

Silica gel column chromatography (CC) and reversed-phase [octadecyl silica gel (ODS)] open column chromatography (RPCC) were performed on Silica gel-60 (Merck, 70—230 mesh) and Cosmosil 75C_{18}-OPN (Nacalai Tesque Co., Ltd.), Φ=50 mm, L=25 cm, MeOH–H_2O (1 : 9, 1.5 l)→(7 : 3, 1.5 l), and fractions of 10 g were collected. Droplet counter-current chromatography (DCCC): 500 columns (Φ=2 mm, L=40 cm). The ascending method was used with $CHCl_3$–MeOH–H_2O–*n*-PrOH (9 : 12 : 8 : 2) and 5 g fractions were collected and numbered according to elution order of the mobile phase. Preparative HPLC was carried out with ODS (Inertsil, GL Science Co., Ltd., Φ=20 mm, L=25 cm, flow rate, 6 ml/min) and detection was performed by UV at 254 nm.

Plant Material and Isolation Procedure The plant material used was the same as used in the previous report and part of isolation procedure was also described previously.[1)]

The residue (681 mg in fractions 5—6) on Diaion HP-20 CC was subjected to RPCC. The residue (67 mg in fractions 45—52 and 62 mg in fractions 83—90) were purified by two separate runs of DCCC to give 33 mg of **5** in fractions 9—16 and 47 mg of **6** in fractions 16—21, respectively.

The residue (4.23 g in fractions 7—8) on Diaion HP-20 CC was subjected to silica gel CC (200 g) with increasing amounts of MeOH in $CHCl_3$. The residue (2.40 g) of the 8% MeOH eluate was subjected to RPCC to give 1.25 g of **7** in fractions 66—68. The residue (204 mg) of the 10% MeOH eluate was purified by DCCC (22 mg in fractions 18—22) and then by HPLC to give 10 mg of **1**.

The residue (9.60 g in fractions 9—15) of Diaion HP-20 CC was subjected to silica gel CC (400 g) with increasing amounts of MeOH in $CHCl_3$. The residue (50 mg out of 142 mg) of the 6% MeOH eluate was purified by HPLC (MeOH–H_2O, 7 : 13) to give 26 mg of **9**. The residue (890 mg) of the 8% MeOH eluate on silica gel CC was separated by DCCC. The residue (232 mg) of fractions 14—22 was purified by HPLC (MeOH–H_2O, 1 : 3), Sephadex LH-20 (Φ=20 mm, L=120 cm, MeOH) and then HPLC (MeOH–H_2O, 7 : 13) to give 9 mg of triptephenoside (**2**). The residue (60 mg in fractions 23—30 on DCCC) was purified by HPLC to give 46 mg of **8**. The residue (45 mg in fractions 31—41 on DCCC) was found to be a pure compound on TLC (**10**). The residue (1.47 g) of the 10—12.5% MeOH eluate on silica gel CC was separated by RPCC. The residue (163 mg) in fractions 130—135 was purified by DCCC to give 90 mg of **11** in fractions 36—44. The residue (517 mg in fractions 155—176) was purified by DCCC (310 mg in factions 35—43) and then by HPLC to give 7.0 mg of **3**. The residue (1.56 g) of the 20% MeOH eluate on silica gel CC was subjected to RPCC. The residue (319 mg in fractions 129—134) was separated by DCCC to give 82 mg of **13**, 23 mg of **14** and 84 mg of **12** in fractions 19—22, 27—30 and 42—52, respectively.

The residue (6.02 g in fractions 16—23) of Diaion HP-20 CC was subjected to silica gel CC (400 g) with increasing amount of MeOH in $CHCl_3$. The residue (899 mg) of the 20—25% MeOH eluate was subjected to RPCC (130 mg in fractions 161—171) and then DCCC to give 65 mg of **4** in fractions 56—63.

Known Compounds Isolated Loganic acid (**5**), $[\alpha]_D^{21}$ −72.7° (*c*=0.55, MeOH)[2)]; secologanoside (**6**), $[\alpha]_D^{27}$ −115.8° (*c*=0.82, MeOH)[3)]; morroniside (**7**), $[\alpha]_D^{27}$ −89.9° (*c*=1.39, MeOH, after 24 h being dissolved in the solvent),[4)] ^{13}C- and ^{1}H-NMR (CD_3OD): Table 1; sweroside (**8**), $[\alpha]_D^{27}$ −190.6° (*c*=0.83, MeOH)[5)]; 7α-methoxysweroside (**9**), $[\alpha]_D^{27}$ −188.0° (*c*=0.79, MeOH)[6)]; 7β-methoxysweroside (**10**), $[\alpha]_D^{27}$ −113.5° (*c*=0.79, MeOH),[6)] isovitexin (**11**), $[\alpha]_D^{22}$ −9.2° (*c*=0.72, pyridine)[7)]; isoorientin (**12**), $[\alpha]_D^{22}$ *ca.* 0° (*c*=0.73, pyridine)[7)]; 2″-*O*-α-L-rhamnopyranosylisovitexin (**13**), $[\alpha]_D^{27}$ −122.4° (*c*=0.65, pyridine), ^{1}H-NMR (DMSO-d_6, 90°) δ: 0.61 (3H, d, *J*=6 Hz, H_3-6‴), 2.45 (1H, m, H-5‴), 2.95 (1H, t, *J*=10 Hz, H-2″), 3.42 (1H, t, *J*=8 Hz, H-4″), 3.49 (1H, dd, *J*=5, 12 Hz, H-6″a), 3.65 (1H, dd, *J*=2, 3 Hz, H-2‴), 3.66 (1H, dd, *J*=2, 12 Hz, H-6″b), 4.25 (1H, very br s, H-2″), 4.73 (1H, d, *J*=10 Hz, H-1″), 5.06 (1H, d, *J*=2 Hz, H-1‴), 6.49 (1H, s, H-3), 6.65 (1H, s, H-8), 6.93 (2H, d, *J*=9 Hz, H-3′, 5′), 7.85 (2H, d, *J*=9 Hz, H-2′, 6′), 13.42 (1H, s, 5-OH); ^{13}C-NMR (DMSO-d_6, 90°): Table 3.[8)] 2″-*O*-β-D-Xylopyranosylisovitexin (**14**), $[\alpha]_D^{27}$ −22.3° (*c*=1.03, pyridine), ^{1}H-NMR (DMSO-d_6, 90°) δ: 2.61 (1H, dd, *J*=9, 11 Hz, H-5‴a), 2.94 (1H, t, *J*=8 Hz, H-2‴), 3.70 (1H, dd, *J*=2, 12 Hz, H-6″b), 4.20 (1H, d, *J*=7 Hz, H-1‴), 4.32 (1H, br t, *J*=10 Hz, H-2″), 4.73 (1H, d, *J*=10 Hz, H-1″), 6.47 (1H, s, H-3), 6.66 (1H, s, H-8), 6.93 (2H, d, *J*=9 Hz, H-3′, 5′), 7.86 (2H, d, *J*=9 Hz, H-2′, 6′), 13.47 (1H, br s, 5-OH); ^{13}C-NMR (DMSO-d_6, 90°) δ: 61.6 (C-6″), 65.4 (C-5‴), 69.6 (C-4‴), 70.7 (C-4″), 71.7 (C-1″), 74.1 (C-2‴), 76.1 (C-3″), 78.5 (C-3‴), 80.6 (C-2″), 81.3 (C-5″), 93.6 (C-8), 103.0 (C-3), 103.5 (C-4a), 105.5 (C-1‴), 108.4 (C-6), 116.2 (C-3′ and 5′), 121.4 (C-1′), 128.3 (C-2′, 6′), 156.6 (C-8a), 161.2 (C-5, 4′), 163.7 (C-2, 7), 181.9 (C-4).[8)] 2″-*O*-Glycosylisovitexins (**13**, **14**) have been isolated from many sources. Rotation of *C*-glycosidic bond seemed to be restricted in ambient temperature and a highly shielded signal were observed in the ^{1}H-NMR spectra. Incomplete data, however, have been published for most cases. The ^{1}H- and ^{13}C-NMR data are, therefore, listed above.

6′-*O*-β-D-Glucopyranosylmorroniside (**1**): Amorphous powder, $[\alpha]_D^{27}$ −71.4° (*c*=0.56, MeOH, after 24 h being dissolved in the solvent), UV λ_{max} (MeOH) nm (log ε): 238 (3.99); ^{1}H- and ^{13}C-NMR (CD_3OD): Table 1; HR-FAB-MS (negative-ion mode) *m/z*: 567.1943 $[M-H]^-$ (Calcd for $C_{23}H_{35}O_{16}$: 567.1925).

Triptephenoside (**2**): Amorphous powder, $[\alpha]_D^{27}$ −54.5° (*c*=0.56, MeOH), UV λ_{max} (MeOH) nm (log ε): 216 (4.28), 252 (4.02), 304 (3.56); ^{1}H-NMR (CD_3OD) δ: 3.63 (6H, s, –OCH_3×2), 3.72 (1H, dd, *J*=5, 12 Hz, H-6″a), 3.80 (1H, dd, *J*=2, 12 Hz, H-6″b), 4.88 (1H, d, *J*=7 Hz, H-1″), 6.16 (2H, s, H-3, 5), 7.31 (1H, ddd, *J*=1, 2, 8 Hz, H-4′), 7.36 (1H, dt, *J*=0.5, 8 Hz, H-5′), 7.43 (1H, td, *J*=2, 8 Hz, H-6′), 7.44 (1H, ddd, *J*=0.5, 1, 2 Hz, H-2′); ^{13}C-NMR (CD_3OD): Table 2; HR-FAB-MS (negative-ion mode) *m/z*: 435.1297 $[M-H]^-$ (Calcd for $C_{21}H_{23}O_{10}$: 435.1291).

2‴-*O*-Acetyl-2″-*O*-α-L-rhamnopyranosylisovitexin (**3**): Amorphous powder, UV λ_{max} (MeOH) nm (log ε): 215 (4.40), 271 (4.19), 337 (4.24); ^{1}H-NMR (DMSO-d_6, 90°) δ: 0.67 (3H, d, *J*=6 Hz, H_3-6‴), 1.96 (3H, s, $C\underline{H}_3CO$–), 3.70 (1H, dd, *J*=2, 12 Hz, H-6″b), 4.75 (1H, d, *J*=10 Hz, H-1″), 4.97 (1H, dd, *J*=2, 3 Hz, H-2‴), 5.08 (1H, d, *J*=2 Hz, H-1‴), 6.47 (1H, s, H-3), 6.63 (1H, s, H-8), 6.92 (2H, d, *J*=9 Hz, H-3′, 5′), 7.85 (2H, d, *J*=9 Hz, H-2′, 6′), 13.46 (1H, s, 5-OH); ^{13}C-NMR (DMSO-d_6, 90°): Table 3; HR-FAB-MS (negative-ion mode) *m/z*: 619.1677 $[M-H]^-$ (Calcd for $C_{29}H_{31}O_{15}$: 619.1663).

4‴-*O*-Acetyl-2″-*O*-α-L-rhamnopyranosylisovitexin (**4**): Amorphous powder, $[\alpha]_D^{22}$ −34.8° (*c*=0.58, pyridine), IR ν_{max} (KBr) cm^{-1}: 3300, 1720, 1650, 1605, 1570, 1350, 1240, 1100-995, 853; UV λ_{max} (MeOH) nm (log ε): 215 (4.43), 272 (4.24), 337 (4.27); ^{1}H-NMR (DMSO-d_6, 90°) δ: 0.53 (3H, d, *J*=6 Hz, H3-6‴), 1.71 (3H, s, CH_3CO–), 2.93 (1H, m, H-5‴), 3.36 (1H, dd, *J*=3, 10 Hz, H-3‴), 3.46 (1H, t, *J*=9 Hz, H-4″), 3.52 (1H, dd, *J*=5, 12 Hz, H-6″a), 3.68 (1H, dd, *J*=2, 3 Hz, H-2‴), 3.71 (1H, dd, *J*=2, 12 Hz, H-6″b), 4.18 (1H, very br s, H-2″), 4.63 (1H, t, *J*=10 Hz, H-4‴), 4.76 (1H, d, *J*=10 Hz, H-1″), 5.21 (1H, d, *J*=2 Hz, H-1‴), 6.51 (1H, s, H-3), 6.68 (1H, s, H-8), 6.92 (2H, d, *J*=10 Hz, H-3′, 5′), 7.85 (2H, d, *J*=10 Hz, H-2′, 6′), 13.46 (1H, s, 5-OH); ^{13}C-NMR (DMSO-d_6, 90°): Table 3; HR-FAB-MS (negative-ion mode) *m*/*z*: 619.1649 $[M-H]^-$ (Calcd for $C_{29}H_{31}O_{15}$: 619.1663).

Acetylation of Triptephenoside (2) to Its Pentaacetate (2a) Triptephenoside (**2**) (*ca.* 500 μg) was treated with 100 μl each of $(Ac)_2O$ and pyridine at 25° for 18 h. After addition of MeOH (200 μl), the reaction mixture was evaporated to dryness and subjected to spectral analyses without purification. Pentaacetate (**2a**), ^{1}H-NMR ($CDCl_3$) δ: 2.04, 2.05, 2.06, 2.10 (3H each, all s, CH_3CO–×4, on the alcoholic OHs), 2.32 (3H, s, CH_3CO– on the phenolic OH), 3.69 (6H, s, CH_3O–×2), 3.87 (1H, ddd, *J*=2, 6, 10 Hz, H-5″), 4.13 (1H, dd, *J*=2, 12 Hz, H-6″a), 4.26 (1H, dd, *J*=6, 12 Hz, H-6″b), 5.12—5.19 (2H, m), 5.26—5.31 (2H, m), 6.40 (2H, s, H-3′, 5′), 6.80 (1H, ddd, *J*=1, 2, 8 Hz, H-4), 7.32 (1H, t, *J*=8 Hz, H-5), 7.44 (1H, td, *J*=1, 8 Hz, H-6), 7.58 (1H, br t, *J*=2 Hz, H-2); EI-MS (mass range 100—655) *m*/*z* (rel. int.): 646 (1.7) $[M]^+$, 604 (1.3) $[M-CH_2=C=O]^+$, 331 (99) $[Glc(OAc)_4$ oxonium ion$]^+$, 181 (80), 169 (99), 127 (56), 109 (100); HR-FAB-MS (positive-ion mode) *m*/*z*: 647.1936 $[M+H]^+$ (Calcd for $C_{31}H_{35}O_{15}$: 647.1976).

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References

1) Otsuka H., *Chem. Pharm. Bull*., **47**, 962—965 (1999).
2) Calis I., Lahloub M. F., Sticher O., *Helv. Chim. Acta*, **67**, 160—165 (1984).
3) Calis I., Sticher O., *Phytochemistry*, **23**, 2539—2540 (1984).
4) Gross G.-A., Sticher O., *Helv. Chim. Acta*, **69**, 1113—1119 (1986).
5) Inouye H., Ueda S., Nakamura Y., *Tetrahedron Lett*., **1966**, 5229—5231.
6) Kawai H., Kuroyanagi M., Ueno A., *Chem. Pharm. Bull*., **36**, 3664—3666 (1988).
7) Markham K. R., Chari V. M., Mabry T. J., "The Flavonoids: Advances in Research," ed. by Harborne J. B., Mabry T. J., Chapman & Hall, London, 1982, p. 19.
8) Chopin J., Dellamonica G., "The Flavonoids," ed. by Harborne, J. B., Chapman & Hall, London, 1988, p. 63.
9) Otsuka H., *J. Nat. Prod*., **55**, 1252—1255 (1992).