Isolation and Characterization of New Diacyl 6-*O*-α-L-Rhamnopyranosylcatalpols from the Leaves of *Premna japonica* MIQ.

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Two diacyl rhamnopyranosylcatalpols were isolated from the leaves of *Premna japonica*. The structures of the compounds were determined to be 6-*O*-α-L-(2″-*O*-isoferuloyl, 4″-*O*-acetyl)rhamnopyranosylcatalpol and 6-*O*-α-L-(3″-*O*-isoferuloyl, 4″-*O*-acetyl)rhamnopyranosylcatalpol. The locations of acyl moieties were determined by long-range proton selective decoupling experiments and by partial alkaline hydrolysis.

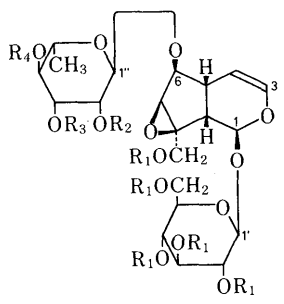
Keywords *Premna japonica*; Verbenaceae; iridoid; 6-*O*-α-L-rhamnopyranosylcatalpol

In the course of investigation of the Philippine medicinal plant, *Premna odorata* BLANCO, two monoacyl and four diacyl 6-*O*-α-L-rhamnopyranosylcatapols were isolated.[1,2] In the Philippines, this plant is used for loosening phlegm, as a cough remedy, to promote urination and vaginal irrigation, and for cardiac troubles.[3] A related plant, *P. japonica* MIQ. (*P. microphylla* TURCZ.) (Verbenaceae) (Japanese name: hamakusagi) is also used in China as an antipyretic, a hemostatic and an antidote for bites by poisonous snakes.[4] In a previous paper,[5] we reported the isolation of two monoacyl 6-*O*-α-L-rhamnopyranosylcatalpols from this plant, collected in the south-eastern part of Tokushima Prefecture, Japan. This paper describes the further isolation and structure determination of two new diacyl iridoid diglycosides.

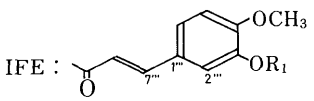
Nonacylated 6-*O*-α-L-rhamnopyranosylcatalpol was first isolated from *Scrophularia nodosa* L.[6] Saccatoside [6-*O*-α-L-(2″-*O*-*p*-coumaroyl)rhamnopyranosylcatalpol], its positional isomer [6-*O*-α-L-(3″-*O*-*p*-coumaroyl)rhamnopyranosylcatalpol] and 6-*O*-α-L-(4″-*O*-*p*-methoxycinnamoyl)rhamnopyranosylcatalpol were obtained from *Verbascum saccatum* C. KOCH,[7] *V. sinuatum* L.[8] and *V. georgicum* BENTH.,[9] respectively. A 2″,3″-diacyl-(acetyl and *p*-methoxycinnamoyl) derivative was found in the roots of *V. sinuatum* L.[10] The isolation of two triacylated derivatives from the roots of *Scrophularia scopolii* [HOPPE *ex*] PERS. var. *scopolii*[11] has been reported.

Results and Discussion

The isolation and purification procedures for two iridoid derivatives are described in detail in the experimental section.



	R_1	R_2	R_3	R_4
1	H	IFE	H	Ac
1a	Ac	IFE	Ac	Ac
2	H	H	IFE	Ac
2a	Ac	Ac	IFE	Ac
3	H	H	H	H
4	H	IFE	H	H
5	H	H	IFE	H



Compound **1**, $C_{33}H_{42}O_{18}$, was obtained as a pale yellow amorphous powder, whose molecular weight was determined by fast-atom bombardment-mass spectrometry (FAB-MS), ion peaks being observed at *m/z* 749 $[M+Na]^+$ (+NaI) and 765 $[M+K]^+$ (+KI). Its infrared (IR) spectrum indicated the presence of a conjugated ester (1710 and 1620 cm^{-1}) and an aromatic ring (1610 and 1510 cm^{-1}). The ^{1}H-nuclear magnetic resonance (^{1}H-NMR) spectrum showed an acetyl signal at δ 2.11, a *trans* double bond at δ 7.64 (d, $J=16$ Hz) and 6.43 (d, $J=16$ Hz), and three aromatic protons, coupled in an ABX system (Table I). The ^{13}C-NMR spectrum showed a total of 33 signals (Table II). Six of the latter were

TABLE I. ^{1}H-NMR Data for Compounds **1** and **2** (400 MHz, CD_3OD)[a]

Proton(s) at	1	2
C-1	5.08 (d, 10)	5.10 (d, 10)
C-3	6.39 (dd, 2, 6)	6.39 (dd, 2, 6)
C-4	5.04 (br t, 6)	5.08 (dd, 4, 6)
C-5	2.43 (m)	2.47 (m)
C-6	4.03 (dd, 1, 8)	4.05 (dd, 1, 8)
C-7	3.65 (br s)	3.63 (br s)
C-9	2.57 (dd, 7, 10)	2.58 (dd, 8, 10)
C-10	3.80 (d, 13)	3.82 (d, 13)
	4.15 (d, 13)	4.16 (d, 13)
C-1′	4.78 (d, 8)	4.78 (d, 8)
C-2′	3.26 (dd, 8, 10)	3.26 (dd, 8, 9)
C-3′	b)	b)
C-4′	3.39 (t, 9)	3.41 (t, 9)
C-5′	b)	b)
C-6′	3.62 (dd, 7, 12)	3.63 (dd, 7, 12)
	c)	3.92 (dd, 2, 12)
C-1″	5.07 (d, 2)	5.02 (d, 2)
C-2″	5.18 (dd, 2, 4)	4.11 (t, 2)
C-3″	4.10 (dd, 4, 9)	5.24 (m, 2H)
C-4″	5.00 (t, 9)	
C-5″	c)	3.97 (qd, 6, 10)
C-6″	1.19 (d, 6)	1.20 (d, 6)
C-2‴	7.11 (d, 2)	7.11 (d, 2)
C-5‴	6.95 (d, 8)	7.05—7.08 (2H)
C-6‴	7.09 (dd, 2, 8)	
C-7‴	7.64 (d, 16)	7.61 (d, 16)
C-8‴	6.43 (d, 16)	6.31 (d, 16)
C-OCH_3	3.89 (s)	3.89 (s)
CH_3CO–	2.11 (s)	2.01 (s)

a) The letters and figures in parentheses are the multiplicities and coupling constants in Hz, respectively. b) Signals buried in the solvent signal. c) Signals overlapped by the envelope of the methoxyl signal.

TABLE II. ^{13}C-NMR Data for Compounds **1**, **2** and **3** (100 MHz, CD_3OD)[a]

Carbon No.	**1**	**2**	**3**[b]
C-1	95.1 (d, 167)	95.3 (d, 167)	95.1
C-3	142.4 (d, 191)	142.4 (d, 190)	142.1
C-4	103.2 (d, 170)	103.4 (d, 168)	103.6
C-5	37.2 (d, 140)	37.3 (d, 139)	37.2
C-6	84.8 (d, 140)	84.4 (d, 140)	83.5
C-7	59.6 (d, 192)	59.5 (d, 190)	59.3
C-8	66.5 (s)	66.6 (s)	66.5
C-9	43.3 (d, 138)	43.3 (d, 138)	43.2
C-10	61.5 (t, 145)	61.5 (t, 145)	61.4
C-1′	99.7 (d, 162)	99.8 (d, 164)	99.7
C-2′	74.8 (d, 145)	74.9 (d, 140)	74.8
C-3′	78.6 (d, 142)	78.6 (d, 141)	78.5
C-4′	71.7 (d, 147)	71.8 (d, 145)	71.7
C-5′	77.6 (d, 144)	77.7 (d, 144)	77.6
C-6′	62.9 (t, 142)	63.0 (d, 144)	62.9
C-1″	97.7 (d, 170)	100.4 (d, 169)	100.3
C-2″	74.1 (d, 156)	70.3 (d, 149)	72.2
C-3″	68.4 (d, 148)	72.7 (d, 155)	72.2
C-4″	75.3 (d, 150)	73.0 (d, 151)	73.8
C-5″	68.1 (d, 147)	68.2 (d, 144)	70.1
C-6″	17.8 (q, 127)	17.8 (q, 128)	18.0
C-1‴	128.8 (s)	128.9 (s)	
C-2‴	112.5 (d, 161)	112.7 (d, 160)	
C-3‴	151.6 (s)	151.7 (s)	
C-4‴	148.0 (s)	148.0 (s)	
C-5‴	115.7 (d, 163)	115.8 (d, 163)	
C-6‴	123.0 (d, 161)	123.0 (d, 163)	
C-7‴	147.4 (d, 155)	147.4 (d, 154)	
C-8‴	114.9 (d, 156)	115.0 (d, 157)	
C-9‴	168.3 (s)	168.1 (s)	
C-OCH_3	56.4 (q, 144)	56.5 (q, 145)	
$\underline{C}H_3CO$	21.0 (q, 129)	20.9 (q, 129)	
$CH_3\underline{C}O$	172.3 (s)	172.0 (s)	

a) The letters and figures in parentheses are the multiplicities in off-resonance spectra and coupling constants (J_{CH}) in Hz, respectively. *b*) Data taken from ref. 1.

assigned to glucopyranosyl carbons and another six to rhamnopyranosyl carbons, as suggested by the signal at δ 1.19 (d, J=6 Hz) in the ^{1}H-NMR. The presence of these sugars was confirmed by gas-liquid chromatography (GLC) of a methanolysate of **1**. Nine signals at lower field and a methoxyl signals were attributed to a C_6–C_3 acyl moiety, and two signals (δ 21.0 and 172.3) to an acetyl group. The chemical shifts of ^{13}C- and ^{1}H-NMR signals of the acyl portion matched those of isoferulate better than those of ferulate.[5,12] The remaining nine signals showed good agreement with those of the aglycone portion of 6-*O*-α-L-rhamnopyranosylcatalpol (**3**) (Table II). On comparing the ^{13}C-NMR chemical shifts of compounds **1** and **3**, differences were observed only in the rhamnosyl carbon signals. Thus, the two acyl moieties are located at the hydroxyl groups of this sugar moiety. The anomeric (1″) and 5″ carbon signals (δ 97.7 and 68.1) of **1** were significantly shifted upfield from those of **3** ($\Delta\delta$ 2.6 and 2.0 ppm, respectively), and the 3″ carbon signal also showed an upfield shift, by 3.8 ppm. On the other hand, the 2″ and 4″ carbon signals (δ 74.1 and 75.3) appeared 1.9 and 1.5 ppm downfield from those of **3**, respectively. Thus, the esterified positions of the rhamnose should be the 2″ and 4″ hydroxyl groups. The location of acyl groups was determined by long-range proton selective decoupling experiments. The 2″ and 4″ rhamnosyl ring protons were well resolved, appearing at δ 5.18 (dd, J=2, 4 Hz) and 5.00 (t, J=9 Hz) (Table I). In the non-decoupled ^{13}C-NMR spectrum of **1**, one carbonyl carbon at δ 168.3 showed a doublet of doublet-like (J=2.3, 6.1 Hz) signal, and the other carbonyl carbon signal at δ 172.3 appeared as a multiplet (a quartet of doublet-like signal). This allowed unequivocal assignment of these carbonyl carbons, namely, δ 168.3 and δ 172.3 as isoferuloyl and acetyl groups, respectively. When the proton at δ 5.00 ($C_{4''}$-H) was irradiated, the carbon signal at δ 172.3 (acetyl-CO) collapsed, whereas the other signal remained intact. Irradiation of the proton at δ 5.18 ($C_{2''}$-H) removed a long-range coupling from the carbonyl carbon signal at δ 168.3 (isoferuloyl-CO). Thus the structure of compound **1** was determined to be 6-*O*-α-L-(2″-*O*-isoferuloyl, 4″-*O*-acetyl)rhamnopyranosylcatalpol. On acetylation of compound **1**, a heptaacetate (**1a**) was obtained as a colorless powder. The molecular weight of **1a** was determined by FAB-MS, and besides one original acetyl signal, six alcoholic acetyl signals and one phenolic acetyl signal were observed in the ^{1}H-NMR spectrum. All physical and spectroscopic features of this compound were identical with those of 6-*O*-α-L-(2″-*O*-isoferuloyl)-rhamnopyranosylcatalpol octaacetate.[5]

Compound **2**, $C_{33}H_{42}O_{18}$, was also obtained as a pale yellow powder. The spectroscopic data for this compound were similar to those for **1**. Since the only significant difference observed was in the ^{13}C-NMR chemical shifts of a rhamnopyranosyl moiety, compound **2** was expected to be a positional isomer as to acyl groups. Notable downfield shifts from non-substituted rhamnopyranose were observed at the 3″ and 4″ carbon signals, and upfield shifts at the 2″ and 5″ carbon signals (**2** from **3** in Table II). The anomeric carbon signal of rhamnopyranose was not affected. These data show that the esterified positions of the two acyl groups are restricted to the 3″ and 4″ hydroxyls of rhamnopyranose. To determine the locations of the acyl moieties, long-range protons selective decoupling experiments could not be conducted in this case, because the 3″ and 4″ rhamnose ring protons were not well resolved (see Table I). Therefore, compound **2** was partially hydrolyzed under mild basic conditions. Two ultraviolet (UV)-active compounds were isolated along with 6-*O*-α-L-rhamnopyranosylcatalpol (**3**), and the spectroscopic data of these compounds were identical with those of 6-*O*-α-L-(2″-*O*- and 3″-*O*-isoferuloyl)rhamnopyranosylcatalpols (**4** and **5**), which have been isolated from the same plant.[5] Since facile acyl migration has been reported between the 2- and 3-hydroxyl groups of rhamnose, but not between the 4 and other positions,[13] the original locations of the isoferuloyl and acetyl groups should be the 3″- and 4″-positions, respectively. Thus, the structure of **2** was determined to be 6-*O*-α-L-(3″-*O*-isoferuloyl, 4″-*O*-acetyl)rhamnopyranosylcatalpol. This was confirmed by an acetylation experiment. On acetylation of compound **2**, a heptaacetate (**2a**) was obtained. This compound was identical in all physical and spectroscopic aspects with the octaacetate of 6-*O*-α-L-(3″-*O*-isoferuloyl)rhamnopyranosylcatalpol.[5]

Experimental

^{1}H- and ^{13}C-NMR were measured with a JEOL GX-400 spectrometer at 400 and 100 MHz, respectively, unless otherwise stated. For 25 MHz ^{13}C-NMR and 100 MHz ^{1}H-NMR, a JEOL FX-100 spectrometer was used. IR and UV spectra were recorded with Shimadzu IR-408 and Shimadzu UV-200S spectrophotometers, respectively. FAB- and electron impact (EI) (70 eV)-MS spectra were obtained with a JEOL D-300 spectrometer. Optical rotations were measured with a Union PM-101 automatic digital polarimeter. The droplet counter-current chromatograph (DCCC) (Tokyo Rikakikai) was equipped with 500 glass columns (2 mm × 40 cm) and operated in the ascending mode. Authentic 6-*O*-α-L-rhamnopyranosylcatalpol was from our previous experiment.[1)]

Plant Material Leaves of *P. japonica* were collected in the south-eastern part of Tokushima Prefecture in May (1988). A voucher specimen (88-PJ-Tokushima-1) was deposited at the Herbarium of the Department of Pharmacognosy, Institute of Pharmaceutical Sciences, Hiroshima University School of Medicine.

Isolation Procedures The dried leaves of *P. japonica* (1.45 kg) were extracted with *n*-hexane and then MeOH. The MeOH extract (285 g) was dissolved in 95% aqueous MeOH and then extracted with *n*-hexane. The concentrated MeOH layer was suspended in H_2O and then extracted with EtOAc and 1-BuOH, successively. The 1-BuOH extract (84.4 g) was subjected to Diaion HP-20 column chromatography with stepwise increases in the MeOH content in H_2O (20, 40, 60, 80 and 100%). The 60% MeOH eluate (14.0 g) was subjected to silica gel column chromatography with the solvent system of $CHCl_3$–MeOH–H_2O (150 : 30 : 1). Further purification of an appropriate fraction on silica gel with the solvent system of $CHCl_3$–MeOH (94 : 6) gave 182 mg and 2.24 g of compound **1**- and **2**-rich fractions, respectively. Final purification of the compound **1**-rich fraction by DCCC ($CHCl_3$–MeOH–H_2O–1-PrOH, 45 : 60 : 40 : 10) afforded 102 mg of **1** as a pale yellow powder. The compound **2**-rich fraction (1.84 g) was subjected to 3 runs of DCCC ($CHCl_3$–MeOH–H_2O–1-PrOH, 45 : 60 : 40 : 10), which gave 299 mg of **2** as a pale yellow powder.

Compound 1 A pale yellow amorphous powder. $[\alpha]_D^{23}$ −82.4° (c=0.30, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3350, 2900, 1710, 1620, 1510, 1440, 1375, 1260, 1125, 1050, 920, 840, 805. UV λ_{max}^{MeOH} nm (log ε): 216 (4.26), 234 (4.10) sh, 243 (4.14), 299 (4.28), 311 (4.28), 329 (4.35). FAB-MS m/z: 749 ([M+Na]$^+$) (+NaI), 765 ([M+K]$^+$) (+KI). ^{1}H- and ^{13}C-NMR: see Tables I and II, respectively. *Anal.* Calcd for $C_{33}H_{42}O_{18}\cdot H_2O$: C, 53.22; H, 5.95. Found: C, 53.47; H, 5.95.

Compound 1 Heptaacetate (1a) Compound **1** (35 mg) was treated with a mixture of acetic anhydride and pyridine at 25 °C overnight. The usual work-up gave 42 mg of a white amorphous powder. $[\alpha]_D^{22}$ −36.1° (c=0.46, $CHCl_3$). IR ν_{max}^{KBr} cm^{-1}: 1750, 1630, 1605, 1510, 1430, 1365, 1225, 1120, 1040, 905. UV λ_{max}^{MeOH} nm (log ε): 227 (4.19), 292 (4.37) inf, 309 (4.46). EI-MS m/z: 331, 177, 169. FAB-MS m/z: 1021 ([MH]$^+$), 1043 ([M+Na]$^+$) (+NaI), 1059([M+K]$^+$) (+KI). ^{1}H-NMR ($CDCl_3$, 100 MHz) δ: 1.24 (3H, d, J=6 Hz), 1.99, 2.01, 2.03, 2.04, 2.06, 2.10, 2.12 (3H each, s, alcoholic Ac × 7), 2.32 (3H, s, phenolic Ac), 3.88 (3H, s), 6.33 (H, br d, J=6 Hz), 6.40 (H, d, J=16 Hz), 6.93 (H, d, J=8 Hz), 7.40 (H, dd, J=2, 8 Hz), 7.66 (H, d, J=16 Hz). ^{13}C-NMR ($CDCl_3$, 25 MHz) δ: 17.4, 20.6 (Ac × 8), 35.5, 41.7, 56.0, 58.0, 61.1, 62.1, 67.0, 68.3, 68.9, 70.0, 70.6, 71.2, 72.3, 72.6, 83.5, 94.3, 96.6 (× 2), 102.4, 112.4, 115.7, 122.3, 127.3, 127.9, 140.1, 141.1, 145.0, 153.3, 166.0, 168.7, 169.0, 169.2, 169.9 (× 2), 170.2, 170.6 (× 2). *Anal.* Calcd for $C_{47}H_{56}O_{25}$: C, 55.29; H, 5.52. Found: C, 55.22; H, 5.57.

Compound 2 A pale yellow amorphous powder. $[\alpha]_D^{23}$ −114.0° (c= 0.31, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3350, 2900, 1710, 1620, 1510, 1440, 1375, 1260, 1120, 1050, 920, 840, 810. UV λ_{max}^{MeOH} nm (logε): 216 (4.25), 234 (4.09) sh, 242 (4.12), 298 (4.29), 310 (4.30), 326 (4.35). FAB-MS m/z: 749 ([M+Na]$^+$) (+NaI), 765([M+K]$^+$) (+KI). ^{1}H- and ^{13}C-NMR: see Tables I and II, respectively. *Anal.* Calcd for $C_{33}H_{42}O_{18}\cdot H_2O$: C, 53.22; H, 5.95. Found: C, 53.78; H, 5.99.

Compound 2 Heptaacetate (2a) Compound **2** (38 mg) was treated with a mixture of acetic anhydride and pyridine at 25 °C overnight. The usual work-up gave 44 mg of a white amorphous powder. $[\alpha]_D^{22}$ −54.5° (c=0.44, $CHCl_3$). IR ν_{max}^{KBr} cm^{-1}: 1745, 1630, 1605, 1505, 1430, 1365, 1220, 1120, 1040, 905. UV λ_{max}^{MeOH} nm (log ε): 227 (4.23), 293 (4.41) inf, 309 (4.49). EI-MS m/z: 331, 177, 169. FAB-MS m/z: 1021 ([M]$^+$), 1043 ([M+Na]$^+$) (+NaI), 1059 ([M+K]$^+$) (+KI). ^{1}H-NMR ($CDCl_3$, 100 MHz) δ: 1.24 (3H, d, J=6 Hz), 2.03 (9H, s), 2.04 (3H, s), 2.11 (3H, s), 2.12 (3H, s), 2.17 (3H, s) (alcoholic Ac × 7), 2.32 (3H, s, phenolic Ac), 3.88 (3H, s), 6.19 (H, d, J=16 Hz), 6.23 (H, br d, J=6 Hz), 6.96 (H, d, J=8 Hz), 7.24 (H, d, J= 2 Hz), 7.32 (H, dd, J=2, 8 Hz), 7.57 (H, d, J=16 Hz). ^{13}C-NMR ($CDCl_3$, 25 MHz) δ: 17.4, 20.6 (Ac × 6), 20.8 (Ac), 20.9, 35.5, 41.7, 56.0, 58.0, 61.1, 62.1, 62.4, 67.0, 68.3, 68.9, 70.2, 70.6, 71.1, 72.3, 72.6, 83.6, 94.3, 96.5 (× 2), 102.4, 112.3, 115.6, 122.2, 127.3, 127.9, 140.0, 141.1 144.8, 153.2, 165.7, 168.7, 169.0, 169.2, 170.0 (× 2), 170.2, 170.6 (× 2), *Anal.* Calcd for $C_{47}H_{56}O_{25}$: C, 55.29; H, 5.52. Found: C, 54.84; H, 5.52.

Partial Alkaline Hydrolysis of Compound 2 Compound **2** (80 mg, 55 mg as 6-*O*-α-L-rhamnopyranosylcatalpol) was treated in a methanolic (50%) 0.05 N NaOH solution (50 ml) at 0 °C for 30 min under an N_2 atmosphere. The reaction mixture was neutralized with Dowex 50 W × 8 and then the resin was removed by filtration. The filtrate was evaporated to dryness and then subjected to DCCC ($CHCl_3$–MeOH–H_2O–1-PrOH, 45 : 60 : 40 : 10). Three compounds were isolated. The most polar compound (15 mg, 27%) was 6-*O*-α-L-rhamnopyranosylcatalpol (**3**). The remaining compounds were found to be UV-active on thin layer chromatography (TLC) (precoated silica gel GF_{254}). The more polar major compound (20 mg, 28% as 6-*O*-α-L-rhamnopyranosylcatalpol) was identified as 6-*O*-α-L-(2″-*O*-isoferuloyl)rhamnopyranosylcatalpol (**4**), which was previously isolated from the same plant[5)]: a colorless powder. $[\alpha]_D^{25}$ −100.8° (c=1.67, MeOH). FAB-MS m/z: 707 ([M+Na]$^+$) (+NaI), 723 ([M+K]$^+$) (+KI). ^{1}H-NMR (CD_3OD) δ: 1.31 (3H, d, J=6 Hz), 2.45 (H, m), 2.57 (H, dd, J=8, 10 Hz), 3.25 (H, dd, J=9, 10 Hz), 3.26 (H, dd, J=8, 9 Hz), 3.40 (H, t, J= 9 Hz), 3.62 (H, dd, J=6, 12 Hz), 3.65 (H, br s), 3.68 (H, t, J=9 Hz), 3.82 (H, d, J=13 Hz), 3.89 (3H, s), 3.92 (H, dd, J=2, 12 Hz), 4.04 (H, dd, J=1, 8 Hz), 4.08 (H, dd, J=2, 3 Hz), 4.15 (H, J=13 Hz), 4.78 (H, d, J=8 Hz), 4.97 (H, d, J=1 Hz), 5.09 (H, d, J=10 Hz), 5.11 (H, dd, J=5, 11 Hz), 6.38 (H, dd, J=2, 6 Hz), 6.40 (H, d, J= 16 Hz), 6.94 (H, d, J=8 Hz), 7.06 (H, dd, J=2, 8 Hz), 7.09 (H, d, J=2 Hz), 7.66 (H, d, J=16 Hz). ^{13}C-NMR (CD_3OD) δ: 18.1, 37.3, 43.3, 56.4, 59.5, 61.5, 63.0, 66.6, 70.3, 70.6, 71.8, 74.2, 74.3, 74.9, 77.7, 78.7, 84.4, 95.2, 97.8, 99.7, 103.5, 112.6, 114.9, 115.9, 123.0, 128.9, 142.3, 147.2, 148.1, 151.6, 168.5. The less polar minor compound (13 mg, 18% as 6-*O*-α-L-rhamnopyranosylcatalpol) was identical with 6-*O*-α-L-(3″-*O*-isoferuloyl)rhamnopyranosylcatalpol (**5**), which was also isolated from the same plant[5)]: a colorless powder. $[\alpha]_D^{25}$ −108.3° (c=1.08, MeOH). FAB-MS m/z: 707 ([M+Na]$^+$) (+NaI), 723 ([M+K]$^+$) (+KI). ^{1}H-NMR (CD_3OD) δ: 1.31 (H, d, J=6 Hz), 2.42 (H, m), 2.56 (H, dd, J=8, 10 Hz), 3.24 (H, dd, J=9, 10 Hz), 3.26 (H, dd, J=8, 9 Hz), 3.40 (H, t, J=9 Hz), 3.48 (H, t, J=10 Hz), 3.62 (H, dd, J=7, 12 Hz), 3.64 (H, br s), 3.89 (3H, s), 3.9 (H, dd-like, overlapped with a methoxyl signal), 4.02 (H, dd, J=1, 8 Hz), 4.15 (H, d, J=13 Hz), 4.77 (H, d, J=8 Hz), 5.02 (H, d, J=1 Hz), 5.07 (H, dd, J=5, 11 Hz), 5.08 (H, d, J=10 Hz), 5.14 (H, dd, J= 2, 4 Hz), 6.37 (H, dd, J=2, 6 Hz), 6.38 (H, d, J=16 Hz), 6.95 (H, d, J= 8 Hz), 7.07 (H, dd, J=2, 8 Hz), 7.10 (H, d, J=2 Hz), 7.63 (H, d, J=16 Hz). ^{13}C-NMR (CD_3OD) δ: 18.0, 37.3, 43.3, 56.4, 59.3, 61.5, 63.0, 66.6, 70.3, 70.4, 71.4, 71.8, 74.9, 75.4, 77.7, 78.7, 83.8, 95.2, 99.8, 100.3, 103.6, 112.6, 114.8, 116.4, 122.8, 129.1, 142.3, 146.8, 151.5, 168.7.

Analysis of the Sugar Portion Methanolysis of about 2 mg of compounds **1** and **2** was carried out in 1.5 ml of 5% hydrogen chloride in dry MeOH at 95 °C for 3 h. The reaction mixture was neutralized by the addition of Ag_2CO_3 and then filtered. The filtrate was evaporated to dryness and then several drops of trimethylsilyl (TMS)-imidazole were added. After 30 min at 60 °C, 1 ml of H_2O was added, followed by extraction with 2 ml of *n*-hexane. The *n*-hexane layer was dried and then subjected to GLC analysis. Column, 1.5% OV-1; temperature, 180 °C (isothermal); N_2, 40 ml/min. t_R: authentic rhamnose, 2.81 min; authentic glucose, 9.08 and 9.98 min; Compound **1**: rhamnose, 2.80 min; glucose, 9.09 (overlapped by methyl isoferulate-3-*O*-TMS) and 9.99 min. Compound **2**: rhamnose, 2.81 min; glucose, 9.09 (overlapped by methyl isoferulate-3-*O*-TMS) and 9.99 min.

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References

1) H. Otsuka, N. Kubo, K. Yamasaki, and W. G. Padolina, *Phytochemistry*, **28**, 513 (1989).
2) H. Otsuka, N. Kubo, K. Yamasaki, and W. G. Padolona, *Phytochemistry*, **28**, 3063 (1989).
3) A. Quisumbing, "Medicinal Plants of the Philippines," Katha Pub. Co., Manila, 1978, pp. 800—801.
4) "Inconographia Cormophytorum Sinicorum, Tomus III," Science

Press, Beijing, 1980, p. 589.
5) H. Otsuka, Y. Sasaki, K. Ymasaki, Y. Takeda, and T. Seki, *Phytochemistry*, **28**, 3069 (1989).
6) K. Weinges and H. von der Eltz, *Justus Liebigs Ann. Chem.*, **1978**, 1968.
7) V. A. Mnatsukanyan, L. S. Arutyunyan, and M. I. Eribekyan, *Khim. Prir. Soedin.*, **1983**, 38.
8) M. I. Eribekyan, L. S. Arutyunyan, and V. A. Mnatsukanyan, *Khim. Prir. Soedin.*, **1987**, 146.
9) E. Yu. Agabayan, L. S. Arutyunyan, V. A. Mnatsukanyan, E. Gach-Baitts, and L. Radich, *Khim. Prir. Soedin.*, **1982**, 446.
10) G. Falsone, M. D. Laryea, A. E. G. Crea, and E. Finner, *Planta Med.*, **44**, 150 (1982).
11) I. Calis, G.-A. Gross, T. Winkler, and O. Sticher, *Planta Med.*, **1988**, 168.
12) Y. Takeda, *J. Nat. Prod.*, **51**, 180 (1988).
13) S. A. Abbas and A. H. Haines, *Carbohydr. Res.*, **39**, 358 (1975).