Megastigmane Glycosides and an Acylated Triterpenoid from *Eriobotrya japonica*

Hideyuki Ito,[†] Eri Kobayashi,[†] Shu-Hua Li,[†] Tsutomu Hatano,[†] Daigo Sugita,[‡] Naoki Kubo,[‡] Susumu Shimura,[‡] Yoshio Itoh,[‡] and Takashi Yoshida*[,†]

Faculty of Pharmaceutical Sciences, Okayama University, Tsushima, Okayama 700-8530, Japan, and Central Research Institute of Lotte Co., Ltd., Urawa, Saitama 336-0027, Japan

Received January 12, 2001

Two new megastigmane glycosides, eriojaposides A (**1**) and B (**2**), and a new acylated triterpenoid (**3**) were isolated along with nine known compounds from a leaf extract of *Eriobotrya japonica*. The structures of **1**−**3** were characterized as (6*R*,9*R*)-3-oxo-α-ionyl-9-*O*-*β*-xylopyranosyl-(1″→6′)-*β*-glucopyranoside, (6*R*,9*R*)-3-oxo-α-ionyl-9-*O*-α-rhamnopyranosyl-(1″→6′)-*β*-glucopyranoside, and 3α-*trans*-feruloyloxy-2α-hydroxyurs-12-en-28-oic acid, respectively, on the basis of spectral and chemical evidence.

Eriobotrya japonica Lindl. (Rosaceae) has been used traditionally as a folk medicine for the treatment of chronic bronchitis, cough, phlegm, high fever, ulcers, and cancer in Japan and other Asian countries.[1] The chemical constituents of this popular folk medicine include several polyphenolic compounds,[2] triterpenoids,[3,4] and megastigmane glycosides[5] found in the leaves. We recently reported the isolation and characterization of 17 polyphenols including three new flavonoid glycosides from the leaves of *E. japonica* and the potent cytotoxic activity of a procyanidin oligomer, a major polyphenol of the plant, against human oral tumor cell lines.[6] Further investigation of this plant part has led to the isolation of two new megastigmane glycosides, named eriojaposides A (**1**) and B (**2**), and a new acylated triterpenoid (**3**) together with nine known terpenoid constituents. This paper deals with the structure elucidation of these new compounds.

Results and Discussion

The aqueous acetone extract of the leaves of *E. japonica* was extracted sequentially with ether, EtOAc, and *n*-BuOH. Repeated column chromatography of the *n*-BuOH-soluble portion yielded eight megastigmane glycosides, among which six were identified by spectroscopic data and by comparison with literature values as (6*S*,9*R*)-roseoside,[5,7,8] (6*S*,9*R*)-vomifoliol-9-*O*-*β*-apiofuranosyl-(1″→6′)-*β*-glucopyranoside (**4**),[5] (6*S*,9*R*)-vomifoliol-9-*O*-*β*-xylopyranosyl-(1″→6′)-*O*-*β*-glucopyranoside (**5**),[9] (6*R*,9*R*)-3-oxo-α-ionyl-9-*O*-*β*-glucopyranoside,[5,10] (6*R*,9*R*)-3-oxo-α-ionyl-9-*O*-*β*-apiofuranosyl-(1″→6′)-*O*-*β*-glucopyranoside,[5,7] and citroside A.[11] From the ether extract, a new triterpenoid (**3**) was obtained together with *β*-sitosterol, ursolic acid,[3] and 3-*O*-*trans*-feruloyleuscaphic acid.[12] The presence of **5** and citroside A in this plant is reported for the first time.

Eriojaposide A (**1**) was isolated as an amorphous powder. Its molecular formula was derived as $C_{24}H_{38}O_{11}$ by HRESIMS (*m*/*z* 503.2507 $[M + H]^+$). The ^{1}H NMR spectrum of **1** showed signals assignable to a vinyl proton at *δ* 5.93 (br s) and two mutually coupled vinyl protons at *δ* 5.81 (dd, *J* = 7, 15 Hz) and *δ* 5.69 (dd, *J* = 9, 15 Hz). Proton signals due to two methines (*δ* 4.43, 2.73), an isolated methylene (*δ* 2.48, 2.09), a vinyl methyl (*δ* 1.98, d, *J* = 1.5 Hz), a secondary methyl (*δ* 1.33, d, *J* = 6 Hz), and two tertiary methyl groups (*δ* 1.08, 1.05, each s) were observed in the aliphatic region. The presence of two sugar residues was suggested by two doublet signals [*δ* 4.39, 4.32 (*J* = 7 Hz each)] due to anomeric protons. The ^{13}C NMR spectrum of **1** showed 24 carbon signals, among which 13 resonances were similar to those corresponding to a 3-oxo-α-ionyl moiety of (6*R*,9*R*)-3-oxo-α-ionyl-9-*O*-*β*-glucopyranoside and (6*R*,9*R*)-3-oxo-α-ionyl-9-*O*-*β*-apiofuranosyl-(1″→6′)-*O*-*β*-glucopyranoside. The remaining 11 carbon resonances could be superimposed on signals due to *β*-glucopyranosyl and *β*-xylopyranosyl units in **5** (Table 1). Upon acid hydrolysis of **1**, glucose and xylose were liberated as judged by GLC. The locations of the sugar residues in **1** were established by the HMBC experiment. Hence, the anomeric proton signal (*δ* 4.39) of glucose, which was assigned from the ^{1}H−^{1}H COSY NMR spectrum, was correlated through a three-bond coupling with C-9 (*δ* 77.2) of the aglycon. The other anomeric proton (xylose) at *δ* 4.32 was also correlated with C-6′ of glucose at *δ* 69.6. A xylopyranosyl-(1″→6′)-glucopy-

Table 1. ^{13}C NMR Spectral Data of **1**, **2**, and **5** (126 MHz, methanol-d_4)

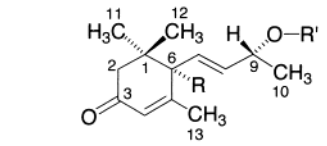
carbon	**1**	**2**	**5**
1	37.1	37.2	42.5
2	48.4	48.4	50.8
3	202.1	202.1	201.3
4	126.1	126.2	127.2
5	165.9	165.8	167.2
6	56.7	56.8	78.0
7	129.0	129.1	131.7
8	138.1	138.1	135.0
9	77.2	77.0	76.9
10	21.1	21.1	21.1
11	27.6	27.7	23.5
12	28.1	28.1	24.7
13	23.8	23.8	19.7
1′	102.6	102.5	102.6
2′	74.9	75.2	74.8
3′	77.9	78.1	77.9
4′	71.3	71.6	71.3
5′	76.9	77.2	76.8
6′	69.6	68.1	69.8
1″	105.5	102.3	105.6
2″	75.2	72.4[a]	75.2
3″	77.7	72.2[a]	77.7
4″	71.2	74.1	71.2
5″	66.9	69.8	66.9
6″		18.1	

[a] These values may be interchanged.

* To whom correspondence should be addressed. Tel and Fax: +81-86-251-7936. E-mail: yoshida@pheasant.pharm.okayama-u.ac.jp.
[†] Okayama University.
[‡] Lotte Co., Ltd.

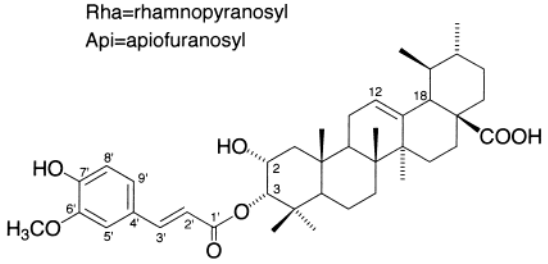
10.1021/np010004x CCC: $20.00
Published on Web 05/23/2001

Chart 1



	R	R'
1	H	Glc (6'→1") Xyl
2	H	Glc (6'→1") Rha
4	OH	Glc (6'→1") Api
5	OH	Glc (6'→1") Xyl

Glc=glucopyranosyl
Xyl=xylopyranosyl
Rha=rhamnopyranosyl
Api=apiofuranosyl



3

ranosyl moiety as the disaccharide sugar chain was thus allocated to C-9 of a 3-oxo-α-ionyl moiety. The CD spectrum of **1** showed a positive Cotton effect at 245 nm ($\Delta\epsilon$ + 22.3), which is similar to that of **5**, indicating the 6*R*-configuration. The absolute configuration at C-9 of the aglycon was assigned as *R* on the basis of a diagnostic chemical shift of the C-9 signal (δ 77.2) in the ^{13}C NMR spectrum.[10] Consequently, the structure of **1** was determined to be (6*R*,9*R*)-3-oxo-α-ionyl-9-*O*-*β*-xylopyranosyl-(1″→6′)-*β*-glucopyranoside.

Eriojaposide B (**2**) was obtained as an amorphous powder, and its ESIMS showed a $[M + H]^+$ ion peak at *m*/*z* 517, which was 14 mass units larger than that of **1**. The molecular formula $C_{25}H_{40}O_{11}$ was confirmed by HRESIMS. The ^{1}H and ^{13}C NMR spectra of **2** were very similar to those of **1** and showed signals of a 3-oxo-α-ionyl moiety and a glucose unit (Table 1). The presence of a rhamnose residue instead of a xylose unit in **1** was indicated by the difference of 14 mass units in the ESIMS and an anomeric proton signal [δ 4.78 (br d, J = 1.5 Hz)] and a methyl signal [δ_H 1.31 (d, J = 6 Hz), δ_C 18.1] in the NMR spectra of **2**. Acid hydrolysis of **2** gave rhamnose and glucose, which were identified by HPLC. The HMBC spectrum of **2** showed long-range correlations through a three-bond coupling between the glucose H-1 (δ 4.37) and C-9 (δ 77.0) of the aglycon and between the rhamnose H-1 (δ 4.78) and C-6 (δ 68.1) of the glucose core. A positive Cotton effect at 245 nm ($\Delta\epsilon$ +30.9) in the CD spectrum of **2** and a chemical shift of C-9 (δ 77.0) in the ^{13}C NMR spectrum were indicative of the 6*R*- and 9*R*-configurations, respectively.[10] On the basis of these data, the structure of **2** was assigned as (6*R*,9*R*)-3-oxo-α-inonyl-9-*O*-α-rhamnopyranosyl-(1″→6′)-*β*-glucopyranoside.

Among the known megastigmane glycosides isolated, **4** and **5** were formulated as shown without the assignment of absolute configurations at C-6 and C-9.[5,9] The 6*S*-configuration of both **4** and **5** was confirmed from positive Cotton effects at ca. 240 nm in their CD spectra. On the other hand, the ^{13}C NMR spectra of **4** and **5** showed resonances for C-9 at δ 76.7 and 76.9, respectively, indicating a 9*R*-configuration for both compounds. Therefore, the gross stereostructures of **4** and **5** were established as (6*S*,9*R*)-vomifoliol-9-*O*-*β*-apiofuranosyl-(1″→6′)-*O*-*β*-glucopyranoside and (6*S*,9*R*)-vomifoliol-9-*O*-*β*-xylopyranosyl-(1″→6′)-*O*-*β*-glucopyranoside, respectively.

Compound **3**, obtained as an amorphous powder, gave a positive result in the Liebermann-Burchard test for an unsaturated triterpenoid. The ^{1}H NMR spectrum of **3** in pyridine-d_5 indicated the presence of five tertiary methyl groups [δ 0.94, 0.97, 1.05, 1.11, 1.17 (each 3H, s)], two secondary methyl groups [δ 0.93, 0.98 (each 3H, d, J = 6.5−7.0 Hz)], an olefinic proton characteristic of H-12 [δ 5.48 (br t, J = 3.5 Hz)] of an ursene skeleton, and a methoxyl group (3H, δ 3.69, s). The presence of a *trans*-feruloyl unit, which was obscured in pyridine-d_5 owing to overlapping of the signals concerned with solvent signals, was revealed in the spectrum measured in methanol-d_4 [δ 7.66 (d, J = 16 Hz), 7.24 (d, J = 2 Hz), 7.11 (dd, J = 2, 8.5 Hz), 6.85 (d, J = 8.5 Hz), 6.47 (d, J = 16 Hz)]. These spectral characteristics together with two oxymethine signals [δ 4.48 (br dt, J = 2.5, 11.5 Hz), 5.61 (d, J = 2.5 Hz)] were similar to those of 3-*O*-*trans*-feruloyleuscaphic acid. A distinguishable feature from the latter compound was a doublet (J = 11.5 Hz) at δ 2.63 assignable to H-18, suggesting the absence of a tertiary hydroxyl group at C-19 of the urs-12-ene skeleton in **3**. This skeleton with α-oxy functions at C-2 and C-3 was substantiated by the chemical shifts and coupling patterns of H-2 and H-3 and the ^{13}C NMR data assigned by HMQC and HMBC. The ester function was confirmed by the ^{13}C NMR spectrum, which showed an ester carbonyl carbon signal at δ 167.9 besides the C-28 signal at δ 179.9. Acid hydrolysis of **3** gave ferulic acid. The position of the ester linkage at O-3 was evident from a large downfield shift of the H-3 signal (δ 5.61) as found in 3-*O*-*trans*-feruloyleuscaphic acid, although a HMBC correlation between H-3 and C-1′ was not observed. Compound **3** was thus assigned the structure 3α-*trans*-feruloyloxy-2α-hydroxyurs-12-en-28-oic acid, which was consistent with the molecular formula $C_{40}H_{56}O_7$ established by a $[M + H]^+$ peak at *m*/*z* 649.4094 in the HRESIMS. This compound was found to be susceptible to partial isomerization to the *cis*-feruloyl isomer, so as to form a mixture.

Experimental Section

General Experimental Procedures. Optical rotations were recorded on a JASCO DIP-1000 polarimeter. CD spectra were measured on a JASCO J-720W spectrometer. ^{1}H and ^{13}C NMR spectra were measured in methanol-d_4 and pyridine-d_5 on a Varian VXR-500 (500 MHz for ^{1}H NMR and 126 MHz for ^{13}C NMR) instrument. ESIMS were obtained using a Micromass Auto Spec OA-TOF mass spectrometer (solvent, 50% MeOH + 0.1% $AcONH_4$; flow rate, 20 μL/min). YMC-Pack A 324 (YMC Co., Ltd.) (10 i.d. × 300 mm) and Luna 5U C_{18} (4.6 i.d. × 250 mm) columns were used for preparative reversed-phase HPLC. Detection was conducted with a Shimadzu SPD-6A spectrophotometric detector at 280 nm and/or a Hitachi L-7455 photodiode array detector. Column chromatography was performed with Diaion HP-20, MCI gel CHP-20P (75−150 μm) (Mitsubishi), Toyopearl HW-40 (coarse grade) (Tosoh), Sephadex LH-20 (Pharmacia), and Si gel (Merck). Solvents were evaporated under reduced pressure below 40 °C.

Plant Material. Fresh leaves of *E. japonica* cultivated in the herbal garden of Okayama University were collected in April 1996. A voucher specimen (OPH-ROS 011) has been deposited in the Herbarium, Faculty of Pharmaceutical Sciences, Okayama University.

Extraction and Isolation. The fresh leaves (1.0 kg) of *E. japonica* were homogenized in 70% acetone (10 L), and

the concentrated solution (1 L) was extracted with ether (1 L × 5), EtOAc (1 L × 6), and *n*-BuOH saturated with water (1 L × 4), successively. The Et_2O extract (10 g) was applied to a Si gel column (5.5 cm i.d. × 55 cm) ($CHCl_3$ and increasing amounts of MeOH) to afford ursolic acid (16 mg) and β-sistosterol (18 mg) from the $CHCl_3$ eluate. Fractions containing UV-sensitive compounds as judged by HPLC with a photodiode array detector were combined and purified by preparative HPLC (Luna 5U C_{18}, MeOH−H_2O, 86:14; flow rate, 1 mL/min) to give three fractions (t_R 6.4, 11.1, 15.7 min). The fractions with t_R 6.4 and 11.1 min gave 3-*O*-*trans*-feruloyleuscaphic acid (5 mg) and 3α-*trans*-feruloyloxy-2α-hydroxyurs-12-en-28-oic acid (**3**) (2 mg), respectively. Although the last fraction (t_R 15.7 min) should have contained the *cis*-feruloyl isomer of **3** (2 mg), its isolation in the pure state was unsuccessful due to the facile formation of a *cis*−*trans* mixture. The *n*-BuOH extract (16 g) was subjected to column chromatography over Diaion HP-20 (2.2 cm i.d. × 60 cm) with H_2O containing increasing amounts of MeOH in a stepwise gradient mode. The 50% MeOH eluate (4.4 g) was separated by a combination of column chromatography over Toyopearl HW-40 (coarse grade), MCI gel CHP-20P (75−150 μm), and/or Sephadex LH-20 and/or purified by preparative reversed-phase HPLC (YMC-Pack A324, MeOH−H_2O, 1:1 or 2:3; flow rate, 1.4 mL/min) to afford eriojaposides A (**1**) (23 mg) and B (**2**) (4 mg), (6*S*,9*R*)-roseoside (77 mg), (6*S*,9*R*)-vomifoliol-9-*O*-β-apiofuranosyl-(1″→6′)-*O*-β-glucopyranoside (29 mg), (6*S*,9*R*)-vomifoliol-9-*O*-β-xylopyranosyl-(1″→6′)-*O*-β-glucopyranoside (**4**) (7 mg), (6*R*,9*R*)-3-oxo-α-ionyl-9-*O*-β-glucopyranoside (**5**) (33 mg), (6*R*,9*R*)-3-oxo-α-ionyl-9-*O*-β-apiofuranosyl-(1″→6′)-*O*-β-glucopyranoside (20 mg), and citroside A (10 mg).

Eriojaposide A (1): amorphous powder; $[\alpha]^{23}_D$ +26.7° (*c* 1.0, MeOH); UV (MeOH) λ_{max} (log ε) 238 (4.08) nm; CD (MeOH) Δε (nm) +22.3 (245), −1.0 (320); ^{1}H NMR (methanol-d_4) δ 5.93 (1H, br s, H-4), 5.81 (1H, dd, *J* = 7.0, 15.0 Hz, H-8), 5.69 (1H, dd, *J* = 9.0, 15.0 Hz, H-7), 4.43 (1H, br t, *J* = 7.0 Hz, H-9), 4.39 (1H, d, *J* = 7.0 Hz, H-1′), 4.32 (1H, d, *J* = 7.0 Hz, H-1″), 4.09 (1H, br d, *J* = 10.0, 5.0 Hz, H-6′), 3.90 (1H, dd, *J* = 5.0, 11.0 Hz, H-6″), 3.75 (1H, dd, *J* = 5.0, 10.0 Hz, H-6′), 3.52 (1H, ddd, *J* = 5.5, 9.0, 12.0 Hz, H-4″), 3.41 (1H, m, H-5′), 3.35 (1H, m, H-4′), 3.25 (1H, dd, *J* = 7.0, 9.0 Hz H-2″), 3.20−3.26 (4H, m, H-2′, 3′, 3″, 5″), 2.73 (1H, d, *J* = 9.0 Hz, H-6), 2.48, 2.09 (each 1H, d, *J* = 17.0 Hz, H-2), 1.98 (3H, d, *J* = 1.5 Hz, H-13), 1.33 (3H, d, *J* = 6.0 Hz, H-10), 1.08 (3H, s, H-12), 1.05 (3H, s, H-11); ^{13}C NMR spectral data, see Table 1; ESIMS *m*/*z* 503 $[M + H]^+$, 371 $[M\text{-xyl} + H]^+$, 209 $[M\text{-xyl-glc} + H]^+$; HRESIMS *m*/*z* 503.2507 $[M + H]^+$ ($C_{24}H_{38}O_{11}$ + H, 503.2492).

Acid Hydrolysis of 1. A solution of **1** (1 mg) in 2.5% H_2SO_4 (0.5 mL) was heated in a boiling waterbath for 2 h. The aqueous layer was neutralized with Amberlite IR-120 (OH form) and evaporated to dryness. The residue after trimethylsilylation was analyzed by GLC (2% OV-17, column temperature 170 °C), which showed the retention times of the sugars to be identical with those of authentic D-glucose and D-xylose.

Eriojaposide B (2): amorphous powder; $[\alpha]^{23}_D$ +33.5° (*c* 1.0, MeOH); UV (MeOH) λ_{max} (log ε) 234 (4.01) nm; CD (MeOH) Δε (nm) +30.9 (245), −1.7 (320); ^{1}H NMR (methanol-d_4) δ 5.93 (1H, br s, H-4), 5.79 (1H, dd, *J* = 7.0, 16.0 Hz, H-8), 5.70 (1H, dd, *J* = 10.0, 16.0 Hz, H-7), 4.78 (1H, d, *J* = 1.5 Hz, H-1″), 4.40 (1H, br t, *J* = 7.0 Hz, H-9), 4.37 (1H, d, *J* = 8.0 Hz, H-1′), 3.98 (1H, dd, *J* = 1.5, 11.0 Hz, H-6′), 3.87 (1H, dd, *J* = 1.5, 3.5 Hz, H-2″), 3.2−3.7 (H-2′−5′, 3″−5″), 2.72 (1H, d, *J* = 9.0 Hz, H-6), 2.48, 2.10 (each 1H, d, *J* = 17.0 Hz, H-2), 1.98 (3H, d, *J* = 1.5 Hz, H-13), 1.33 (3H, d, *J* = 6.5 Hz, H-10), 1.31 (3H, d, *J* = 6.0 Hz, H-6″), 1.08 (3H, s, H-12), 1.05 (3H, s, H-11); ^{13}C NMR spectral data, see Table 1; ESIMS *m*/*z* 517 $[M + H]^+$, 371 $[M\text{-xyl} + H]^+$, 209 $[M\text{-xyl-glc} + H]^+$; HRESIMS *m*/*z* 517.2626 $[M + H]^+$ ($C_{25}H_{40}O_{11}$ + H, 517.2649).

Acid Hydrolysis of 2. A solution of **2** (0.5 mg) in 2.5% H_2SO_4 (0.5 mL) was heated at 90 °C for 2 h. The reaction mixture was neutralized with Amberlite IR-120 (OH form) and analyzed by HPLC {TSK-gel Amide-80, 4.6 i.d. × 250 mm (Tosoh), CH_3CN−H_2O (75:25), RI detection}, which showed the retention times of the sugars to be identical with those of authentic D-glucose and L-rhamnose.

3α-*trans*-Feruloyloxy-2α-hydroxyurs-12-en-28-oic acid (3): white amorphous powder; $[\alpha]^{22}_D$ +7.2° (*c* 0.12, MeOH); UV (MeOH) λ_{max} (log ε) 217 (4.14), 237 (4.04), 297 sh (4.04), 324 (4.18) nm; ^{1}H NMR (pyridine-d_5) δ 8.03 (1H, d, *J* = 16 Hz, H-3′), 7.15−7.23 (3H, H-5′, 8′, 9′), 6.84 (1H, d, *J* = 16 Hz, H-2′), 5.61 (1H, d, *J* = 2.5 Hz, H-3), 5.48 (1H br *t*, *J* = 3.5 Hz, H-12), 4.48 (1H, br dt, *J* = 2.5, 11.5 Hz, H-2), 3.69 (3H, s, OCH_3), 2.63 (1H, d, *J* = 11.5 Hz, H-18), 1.17 (3H, s, H-27), 1.11 (3H, s, H-23), 1.05 (3H, s, H-26), 0.98 (3H, d, *J* = 6.5 Hz, H-29), 0.97 (3H, s, H-25), 0.94 (3H, s, H-24), 0.93 (3H, d, *J* = 7 Hz, H-30); ^{1}H NMR (methanol-d_4) δ 7.66 (1H, d, *J* = 16 Hz, H-3′), 7.24 (1H, d, *J* = 2 Hz, H-5′), 7.11 (1H, dd, *J* = 2, 8.5 Hz, H-8′) 6.85 (1H, d, *J* = 8.5 Hz, H-9′), 6.47 (1H, d, *J* = 16 Hz, H-2′), 5.31 (1H br *t*, *J* = 3.5 Hz, H-12), 5.06 (1H, d, *J* = 2.5 Hz, H-3), 4.14 (1H, m, H-2), 3.94 (3H, s, OCH_3), 2.28 (1H, d, *J* = 10.5 Hz, H-18), 1.33, 1.26, 1.12, 0.95, 0.93 (each 3H, s), 1.04, 0.96 (each 3H, d, *J* = 7 Hz); ^{13}C NMR (pyridine-d_5) δ 179.9 (C-28), 167.9 (C-1′), 151.0 (C-7′), 149.4 (C-6′), 145.5 (C-3′), 139.3 (C-13), 126.6 (C-4′), 125.5 (C-12), 123.1 (C-9′), 116.8 (C-8′), 116.2 (C-2′), 111.5 (C-5′), 80.9 (C-3), 64.7 (C-2), 55.8 (OCH_3), 53.5 (C-18), 50.4 (C-5), 48.1 (C-9), 48.0 (C-17), 43.6. (C-1), 42.5 (C-14), 40.2 (C-8), 39.4, 39.5 (C-19, C-20), 38.7 (2C, C-4, C-10), 37.4 (C-22), 33.3 (C-7), 31.0 (C-21), 28.6 (C-23), 28.4 (C-15), 24.9 (C-16), 24.0 (C-27), 23.7 (C-11), 22.0 (C-24), 21.4 (C-29), 18.3 (C-6), 17.53, 17.49, 16.6 (C-25, C-26, C-30); ESIMS *m*/*z* 649 $[M + H]^+$, 455, 203, 195, 177, 133; HRESIMS *m*/*z* 649.4094 $[M + H]^+$ ($C_{40}H_{56}O_7$ + H, 649.4104).

Acid Hydrolysis of 3. A solution of **3** (0.5 mg) in 1 M HCl (0.5 mL) was heated in a boiling waterbath for 2 h. The reaction mixture was extracted with EtOAc, and the EtOAc layer after evaporation was analyzed by reversed-phase HPLC (Luna 5U C_{18}, MeCN−H_2O, 4:1; flow rate, 1 mL/min; detection, UV 280 nm) to detect ferulic acid (t_R 10.3 min), which was clearly distinguished from isoferulic acid (t_R 11.2 min).

(6*S*,9*R*)-Vomifoliol-9-*O*-β-apiofuranosyl-(1″→6′)-*O*-β-glucopyranoside (4): amorphous powder; $[\alpha]^{23}_D$ +37.2° (*c* 1.2, MeOH); UV (MeOH) λ_{max} (log ε) 236 (3.80) nm; CD (MeOH) Δε (nm) +10.1 (240), −0.6 (318); ^{1}H NMR (methanol-d_4) δ 5.93 (1H, m, H-4), 5.89 (1H, m, H-7), 5.89 (1H, br s, H-8), 5.03 (1H, d, *J* = 2.5 Hz, H-1″), 4.46 (1H, q, *J* = 6.0 Hz, H-9), 4.38 (1H, d, *J* = 7.5 Hz, H-1′), 4.10 (1H, dd, *J* = 2.0, 11.5 Hz, H-6′), 4.01 (1H, d, *J* = 10.0 Hz, H-4″), 3.95 (1H, d, *J* = 2.5 Hz, H-2″), 3.80 (1H, d, *J* = 10.0 Hz, H-4″), 3.73 (1H, dd, *J* = 5.0, 11.5 Hz, H-6″), 3.61 (2H, brs, H-5″), 3.19−3.41 (4H, m, H-2′−5′), 2.56, 2.19 (each 1H, d, *J* = 17.0 Hz, H-2), 1.97 (3H, d, *J* = 1.0 Hz, H-13), 1.33 (3H, d, *J* = 6.0 Hz, H-10), 1.08 (3H, s, H-12), 1.07 (3H, s, H-11); ^{13}C NMR (methanol-d_4) δ 201.3 (C-3), 167.9 (C-5), 134.9 (C-8), 131.6 (C-7), 127.2 (C-4), 111.0 (C-1″), 102.6 (C-1′), 80.5 (C-3″), 78.1 (C-6), 78.0 (C-3′), 77.9 (C-5′), 77.0 (C-2″), 76.7 (C-9), 76.2 (C-2′), 75.0 (C-4″), 71.6 (C-4′), 68.6 (C-6′), 66.6 (C-5″), 50.7 (C-2), 42.5 (C-1), 24.7 (C-12), 23.5 (C-11), 21.1 (C-10), 19.7 (C-13); ESIMS *m*/*z* 519 $[M + H]^+$, 387 $[M\text{-api} + H]^+$, 225 $[M\text{-api-glc} + H]^+$.

(6*S*,9*R*)-Vomifoliol-9-*O*-β-xylopyranosyl-(1″→6′)-*O*-β-glucopyranoside (5): amorphous powder; $[\alpha]^{23}_D$ +22.8° (*c* 1.0, MeOH); UV (MeOH) λ_{max} (log ε) 233 (3.96) nm; CD (MeOH) Δε (nm) +14.1 (242), −1.0 (325); ^{1}H NMR (methanol-d_4) δ 5.91 (2H, m, H-7, 8), 5.90 (1H, m, H-4), 4.48 (1H, m, H-9), 4.39 (1H, d, *J* = 7.5 Hz, H-1′), 4.32 (1H, d, *J* = 7.0 Hz, H-1″), 4.10 (1H, dd, *J* = 2.0, 11.0 Hz, H-6′), 3.90 (1H, dd, *J* = 4.5, 11.0 Hz, H-5″), 3.73 (1H, dd, *J* = 4.5, 11.0 Hz, H-6′), 3.52 (1H, ddd, *J* = 4.5, 8.5, 9.5 Hz. H-4″), 3.2−3.4 (H-2′−5′, 2″, 3″, 5″), 2.56, 2.20 (each 1H, d, *J* = 17.0 Hz, H-2), 1.96 (3H, d, *J* = 1.5 Hz, H-13), 1.33 (3H, d, *J* = 6.5 Hz, H-10), 1.09 (3H, s, H-12), 1.07 (3H, s, H-11); ^{13}C NMR spectral data, see Table 1; ESIMS *m*/*z* 519 $[M + H]^+$, 373 $[M\text{-xyl} + H]^+$, 212 $[M\text{-xyl-glc} + H]^+$; HRESIMS *m*/*z* 519.2416 $[M + H]^+$ ($C_{24}H_{38}O_{12}$ + H, 519.2442).

Acknowledgment. The authors are grateful to the SC-NMR Laboratory of Okayama University for NMR experiments. This work was supported in part by a Grant-in-Aid for Scientific Research (No. 10557207) from the Ministry of Education, Science, Sports and Culture of Japan.

References and Notes

(1) Perry, L. M. *Medicinal Plants of East and Southeast Asia*; MIT Press: Cambridge, 1980; pp 342−343.
(2) Jung, H. A.; Park, J. C.; Chung, H. Y.; Kim, J.; Choi, J. S. *Arch. Pharmacol. Res.* **1999**, *22*, 213−218.
(3) Shimizu, M.; Fukumura, H.; Tsuji, H.; Tanaami, S.; Hayashi, T.; Morita, N. *Chem. Pharm. Bull.* **1986**, *34*, 2614−2617.
(4) De Tommasi, N.; De Simone, F.; Pizza, C.; Mahmood, N.; Orsi, N.; Stein, M. L. *J. Nat. Prod.* **1992**, *55*, 1067−1073.
(5) De Tommasi, N.; Aquino, R.; De Simone, F.; Pizza, C. *J. Nat. Prod.* **1992**, *55*, 1025−1032.
(6) Ito, H.; Kobayashi, E.; Takamatsu, Y.; Li, S.-H.; Hatano, T.; Sakagami, H.; Kusama, K.; Satoh, K.; Sugita, D.; Shimura, S.; Yoshida, T. *Chem. Pharm. Bull.* **2000**, *48*, 687−693.
(7) Otsuka, H.; Yao, M.; Kamada, K.; Takeda, Y. *Chem. Pharm. Bull.* **1995**, *43*, 754−759.
(8) Yoshikawa, M.; Shimada, H.; Saka, M.; Yoshizumi, S.; Yamahara, J.; Matsuda, H. *Chem. Pharm. Bull.* **1997**, *45*, 464−469.
(9) Schwab, W.; Schreier, P. *Phytochemistry* **1990**, *29*, 161−164.
(10) Pabst, A.; Barron, D.; Semon, E.; Schreier, P. *Phytochemistry* **1992**, *31*, 1649−1652.
(11) Umehara, K.; Hattori, I.; Miyase, T.; Ueno, A.; Hara, S.; Kageyama, C. *Chem. Pharm. Bull.* **1988**, *36*, 5004−5008.
(12) Shimizu, M.; Uemitsu, N.; Shirota, M.; Matsumoto, K.; Tezuka, Y. *Chem. Pharm. Bull.* **1996**, *44*, 2181−2182.

NP010004X