# Iridoids from the Green Leaves of *Eucommia ulmoides*

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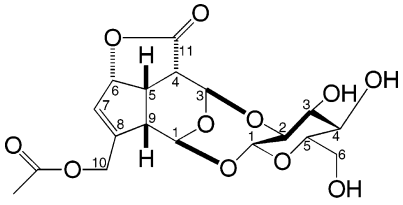
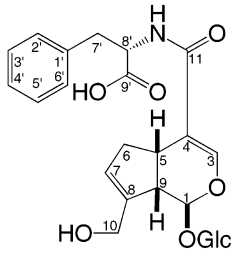
The bark of *Eucommia ulmoides* is a well-known crude drug in oriental medicine, and its leaves have been consumed as a beverage. From the green leaves of this plant, three new iridoids (**1**−**3**) were isolated, together with 12 known compounds. Compound **1** is the first iridoid possessing a saturated bond between C-3 and C-4 and having an ether linkage between C-3 and C-2 of the glucose unit. Furthermore, **2** and **3** may be regarded as the first naturally occurring conjugates of an iridoid and an amino acid.

*Eucommia ulmoides* Oliv. belongs to a single species genus in the plant family Eucommiaceae. The bark of this plant has been used as a tonic to strengthen the liver and kidneys, and its leaves have been used as a food. In the western region of the People's Republic of China, *E. ulmoides* leaves have long been used in tea and in a gruel. Recent pharmacological research on the leaves have shown their hypotensive effects in humans and in spontaneous hypertensive rats (SHR).[1] In a preceding paper, 22 known compounds, consisting of eight iridoids, seven flavonoids, five phenolic derivatives, and two triterpenoids, were identified from the roasted leaves of *E. ulmoides*.[2−5]

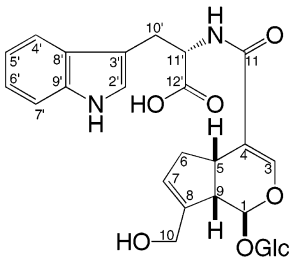
In the present investigation, the constituents have been examined in the green leaves of *E. ulmoides*, which were momentarily treated with steam, then dried at low temperature, to identify 12 known compounds, namely, six iridoids {asperuloside, asperulosidic acid, deacetyl asperulosidic acid, scandoside 10-*O*-acetate, geniposidic acid (**4**), and aucubin}, five flavonoids (quercetin 3-*O*-glucopyranoside, quercetin 3-*O*-sambubioside, rutin, astragalin, and kaempferol 3-*O*-rutinoside), and one phenolic derivative, chlorogenic acid.[6] Three new compounds, named eucomosides A−C (**1**−**3**) were also isolated, and the structural characterization of these three compounds is described herein.



**1**



**2**



**3**

## Results and Discussion

Eucomoside A (**1**) was obtained as a white powder with $[\alpha]_D$ +99.1 (*c* 0.1 MeOH). In the positive FABMS, **1** indicated a $[M + H]^+$ ion peak at *m*/*z* 415. The molecular formula, $C_{18}H_{22}O_{11}$, was determined from the positive HRESIMS data. Acidic hydrolysis of **1** gave D-glucose, and the $^1$H NMR spectroscopic coupling constant of the anomeric proton indicated the linkage mode of the D-glucopyranosyl unit to be $\beta$. The $^{13}$C NMR spectrum (in $C_5D_5N$) displayed a total of 18 carbon signals, which were composed of a monoterpene moiety including two acetal carbon signals at $\delta$ 91.9 and 95.8, one carbonyl carbon signal at $\delta$ 178.5, two olefinic carbon signals at $\delta$ 129.3 and 143.7, two oxygen-bearing carbon signals at $\delta$ 61.0 and 86.9, three methine carbon signals at $\delta$ 35.3, 38.3, and 45.5, together with signals for a $\beta$-D-glucopyranosyl moiety at $\delta$ 99.0, 80.3, 75.8, 71.2, 79.9, and 62.6 (C-1−6), and an acetyl group at $\delta$ 20.5 and 170.3. The respective $^1$H and $^{13}$C NMR signals (Table 1) were assigned by the $^1$H−$^1$H COSY, HMQC, and HMBC spectra as shown in Figure 1.

In the difference NOE spectrum of **1**, NOEs were observed between H-4 and H-5, H-5 and H-6, H-5 and H-9, H-1 and H-9, H-1 and glc H-1, and H-4 and glc H-2, respectively (Figure 2). These correlations were indicated as $\beta$ at C-4 and C-6. Morever, the above data and the $^1$H NMR coupling constants of H-1 ($\delta$ 5.67, d, $J$ = 1.2 Hz) and H-3 ($\delta$ 5.87, d, $J$ = 2.4 Hz) were indicated to be $\alpha$ at both C-1 and C-3. Therefore, the absolute configuration is represented as shown in Figure 1.

Eucomoside B (**2**) was obtained as a white powder showing $[\alpha]_D$ −10.6 (*c* 0.1 $CH_3CN$). In the positive FABMS, **2** indicated a $[M + Na]^+$ ion peak at *m*/*z* 544, and the molecular formula of **2** was determined to be $C_{25}H_{31}NO_{11}$ by positive HRFABMS. The $^{13}$C NMR spectrum (in $CD_3OD$) showed a total of 25 carbon signals, which constituted three moieties: a monoterpene moiety with signals at $\delta$ 97.8, 148.6, 115.8, 36.2, 39.2, 127.9, 144.8, 47.4, 61.4, 168.9 (C-1, 3−11), a $\beta$-D-glucopyranosyl moiety with signals at $\delta$ 100.3, 74.9, 77.9, 71.6, 78.4, 62.7 (C-1−6), and one phenylalanine moiety with signals at $\delta$ 139.8, 130.7, 129.2, 127.4, 129.2, 130.7, 38.7, 56.7, 177.0 (C-1′−9′). The assignments of the respective $^1$H

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**Table 1.** NMR Spectroscopic Data (in $C_5D_5N$) for Eucomoside A (**1**)

| position | $\delta_C$ | $\delta_H$ (*J* in Hz) | HMBC | NOE |
|---|---|---|---|---|
| 1 | 91.9 | 5.67 d (1.2) | H-3, H-9, Glc H-1 | H-9, Glc H-1 |
| 3 | 95.8 | 5.87 d (2.4) | H-1, H-4, Glc H-2 | |
| 4 | 38.3 | 3.54 dd (2.5, 12.8) | | H-5, Glc H-2 |
| 5 | 35.3 | 3.72 m | H-1, H-7 | H-4, H-6, H-9 |
| 6 | 86.9 | | H-7 | H-5, |
| 7 | 129.3 | 5.90 s | H-5, H-9, H-10*a*, H-10*b* | |
| 8 | 143.7 | | H-5, H-6, H-9, H-10*a*, H-10*b* | |
| 9 | 45.5 | 3.30 d | H-1, H-7 | H-1, H-5 |
| 10a | 61.0 | 4.78 d (14.7) | H-7 | |
| 10b | | 4.85 d (14.7) | | |
| 11 | 178.5 | | H-3, H-4, H-6 | |
| OCOCH3 | 170.3 | 2.07 s | $OCOCH_3$ | |
| OCOCH3 | 20.5 | | | |
| Glc 1 | 99.0 | 5.32 d (7.3) | Glc H-2, | H-1 |
| Glc 2 | 80.3 | 4.03 m | H-3, Glc H-1, Glc H-3 | H-4 |
| Glc 3 | 75.8 | 4.29 dd (8.6, 9.2) | Glc H-2, Glc H-4 | |
| Glc 4 | 71.2 | 4.23 dd (9.2, 9.2) | Glc H-3 | |
| Glc 5 | 79.9 | 4.03 m | Glc H-4 | |
| Glc 6a | 62.6 | 4.39 dd (5.5, 11.6) | Glc H-4 | |
| Glc 6b | | 4.60 dd (2.1, 12.2) | Glc H-4 | |

and $^{13}C$ NMR signals (Table 2) in the monoterpene moiety were made by the aid of $^1H-^1H$ COSY, HMQC, and HMBC spectra, as illustrated as Figure 3, suggesting the monoterpene moiety to be **4**.

The linkage location of the phenylalanine moiety was deduced to be at C-11 of **4**; however, no HMBC correlation from H-8′ in the phenylalanine moiety to the iridoid moiety was observed. Moreover, we could not determine the absolute configuration at C-8′. Hence, a synthetic study for the production of **2** was required. Geniposidic acid (**4**) and L-phenylalanine methyl ester (**5a**)[7] were condensed with 1-hydroxybenzotriazole (HOBt) and 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (WSC) to afford a conjugated methyl ester (**5a**), which was then deprotected with 0.5 M NaOH to provide a final product (**7a**), which was identical with **2** with respect to the $^1H$ and $^{13}C$ NMR spectra and HPLC analysis. Compounds **6b** and **7b** were prepared in a similar way (Scheme 1). Compound **7b** was not coincident with **2** with respect to their $^1H$ NMR spectra and HPLC analysis. In particular, the $^1H$ NMR signals due to H-6a and H-6b were shifted by +0.1 and +0.6 ppm, respectively. Hence, **2** was determined to be a condensed compound between the C-11 of the carboxylic acid of **4** and an

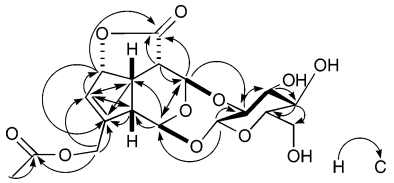


**Figure 1.** HMBC spectrum of eucomoside A (**1**) (in $C_5D_5N$).

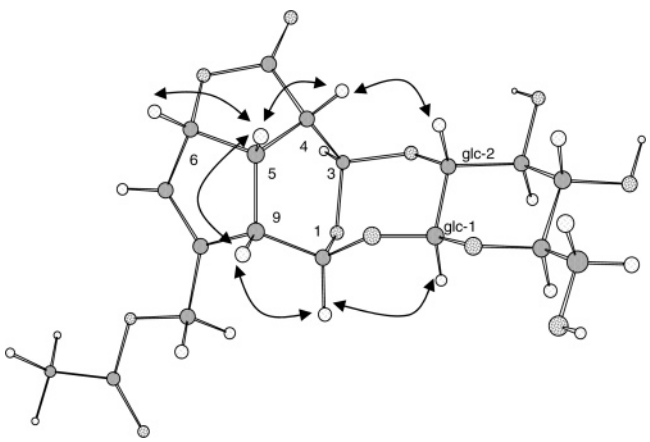



**Figure 2.** NOE spectrum of eucomoside A (**1**) (in $C_5D_5N$).

**Table 2.** NMR Spectroscopic Data (in $CD_3OD$) for Eucomoside B (**2**)

| position | $\delta_C$ | $\delta_H$ (*J* in Hz) | HMBC |
|---|---|---|---|
| 1 | 97.8 | 5.10 d (7.3) | H-5, Glc H-1 |
| 3 | 148.6 | 7.14 d (1.2) | H-1, H-4, H-11 |
| 4 | 115.8 | | H-1, H-6 |
| 5 | 36.2 | 3.20 m | H-1, H-6 |
| 6a | 39.2 | 1.88 dd (7.9, 15.9) | |
| 6b | | 2.50 dd (7.9, 15.3) | |
| 7 | 127.9 | 5.69 s | H-5, H-6, H-9 |
| 8 | 144.8 | | |
| 9 | 47.4 | 2.72 dd (9.2, 9.2) | H-1, H-6, H-7, H-8 |
| 10a | 61.4 | 4.16 d (14.0) | H-7, H-8, H-9 |
| 10b | | 4.29 d (14.0) | |
| 11 | 168.9 | | |
| Glc 1 | 100.3 | 4.69 d (7.3) | H-1 |
| Glc 2 | 74.9 | 3.27 m | H-1, H-3 |
| Glc 3 | 77.8 | 3.39 dd (9.2, 9.2) | H-4 |
| Glc 4 | 71.6 | 3.27 m | |
| Glc 5 | 78.3 | 3.27 m | H-4, H-5 |
| Glc 6a | 62.7 | 3.65 dd (1.8, 12.2) | |
| Glc 6b | | 3.85 dd (1.8, 12.2) | |
| 1′ | 139.8 | | |
| 2′ | 130.7 | 7.20 s | |
| 3′ | 129.2 | 7.21 | |
| 4′ | 127.4 | 7.17 s | |
| 5′ | 129.2 | 7.21 | |
| 6′ | 130.7 | 7.20 s | |
| 7′a | 38.7 | 3.09 dd (7.3, 14.0) | H-1, H-2. H-8 |
| 7′b | | 3.27 m | |
| 8′ | 56.7 | 4.60 m | H-1, H-7, H-9 |
| 9′ | 177.0 | | |

amine function of L-phenylalanine. As far as we know, this is the first example of an iridoid conjugated with an amino acid.

Eucomoside C (**3**) was obtained as a white powder, showing $[\alpha]_D$ −13.1 (*c* 0.5 $H_2O$). The molecular formula of **3** was determined to be $C_{27}H_{32}N_2O_{11}$ by positive HRESIMS. The $^{13}C$ NMR spectrum (in $CD_3OD$) of **3** showed the occurrence of the **4** moiety and a tryptophan moiety, with signals at $\delta$ 124.4, 111.8, 119.6, 119.8, 122.4, 112.2, 129.0, 138.1, 28.3, 57.5, and 180.0 (C-2′−12′). The assignment of the respective $^1H$ and $^{13}C$ NMR signals was confirmed with the $^1H-^1H$ COSY, HMQC, and HMBC spectra, as shown in Figure 4.

Therefore, the structure of **3** was deduced to be a conjugated compound of tryptophan and **4**, to form an amide bond at C-11 of **4**; however, no HMBC correlation between H-11 in the tryptophan moiety and the iridoid moiety was observed. Moreover, the absolute configuration at C-11′ was not readily apparent, so a synthetic study for the production of **3** was required. Compounds **6c**−**7c** and **6d**−

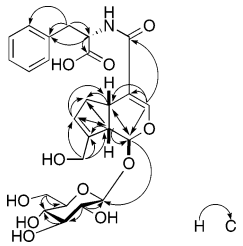


**Figure 3.** HMBC spectrum of eucomoside B (**2**) (in $CD_3OD$).

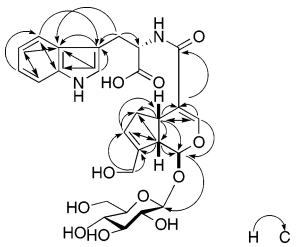
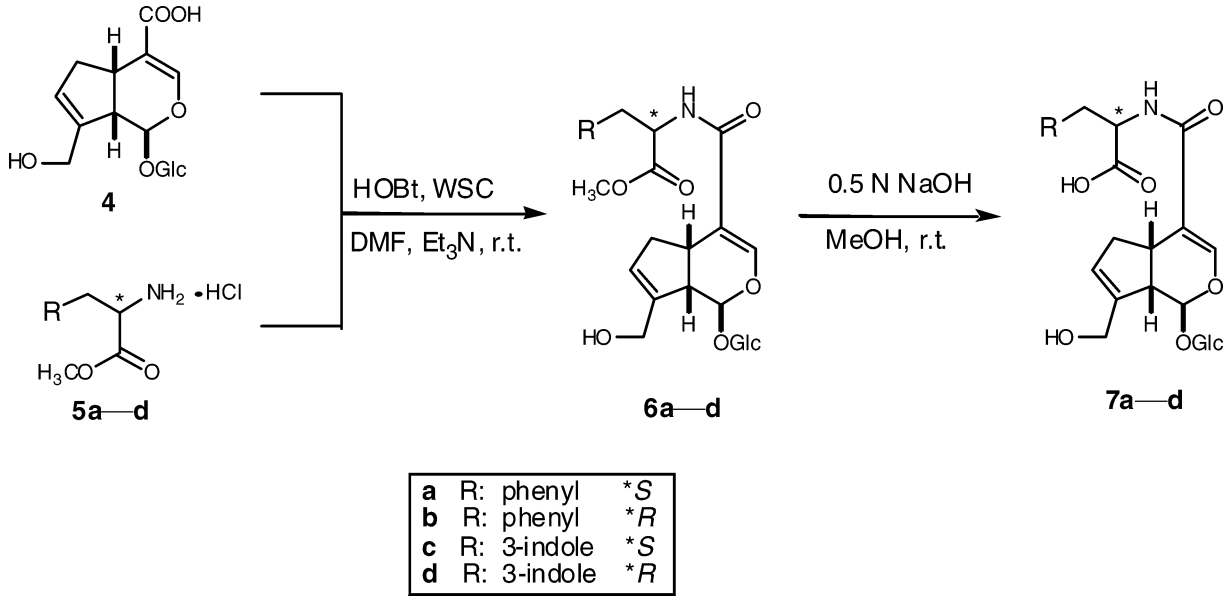


**Figure 4.** HMBC spectrum of eucomoside C (**3**) (in $CD_3OD$).

**Scheme 1.** Synthesis of Iridoids Possessing Aromatic Amino Acids




| | | |
|---|---|---|
| **a** | R: phenyl | *S |
| **b** | R: phenyl | *R |
| **c** | R: 3-indole | *S |
| **d** | R: 3-indole | *R |

**7d** were prepared in a similar way (Scheme 1). Compound **7c** corresponded to **3** with respect to its $^1$H and $^{13}$C NMR data. On the other hand, **7d** was not coincident with **3** with respect to its $^1$H NMR spectrum. In particular, H-6a, H-6b, H-5, and H-7 were shifted by +0.3, +0.4, −0.2, and +0.1 ppm, respectively. Hence, **3** was determined to be a condensed compound between the C-11 of the carboxylic acid of **4** and an amine function of L-tryptophan.

Compound **1** is regarded as the first iridoid possessing a saturated bond between C-3 and C-4 and having an ether bond between C-3 and C-2 of the glucose unit. Furthermore, **2** and **3** are the first natural compounds conjugated between iridoid and an amino acid.

## Experimental Section

**General Experimental Procedures.** Optical rotations were measured using a JASCO DIP-1000 KUY digital polarimeter ($l$ = 0.5). $^1$H and $^{13}$C NMR spectra were recorded on a JEOL α-500 spectrometer, and chemical shifts are given on the $\delta$ (ppm) scale using tetramethylsilane (TMS) as internal standard. FABMS were recorded on a JEOL JMS-700 spectrometer. The HRESIMS were recorded on a JEOL JMS-T100LC "AccuTOF" spectrometer. Column chromatography was carried out with Diaion HP-20P, MCI gel CHP 20P (Mitsubishi Chemical Industries Co., Ltd.), silica gel 60 (Merck, Art. 9385), Sephadex LH-20 (Pharmacia Fine Chemicals), Chromatorex ODS (Fuji Silysia Chemical Co., Ltd.), and Amberlite MB-3 (Organo Co., Ltd.). TLC was performed on precoated silica gel 60 $F_{254}$ (Merck) and RP-18 $F_{254}S$ (Merck), and the compounds were detected by spraying with 10% $H_2SO_4$ in MeOH, followed by heating on a hot plate.

**Plant Material.** The leaves of *E. ulmoides* collected in August 2005 at Chengdu, Sichuan Province, People's Republic of China, were momentarily treated with steam. A voucher specimen (No. 200) was identified by T.N. and deposited in the Herbarium of the Department of Natural Medicines, Kumamoto University, Kumamoto, Japan.

**Extraction and Isolation.** Dried leaves (577.8 g) of *E. ulmoides* were extracted with hot water for 10 h at 60 °C. The extract was subjected to Diaion HP-20 column chromatography with a gradient of $H_2O$−MeOH (1:0 → 0:1) to afford fractions 1−6, in the order of elution. A part (2.0 g) of fraction 1 (135.5 g) was chromatographed over MCI gel CHP 20P with $H_2O$ and MeOH to give geniposidic acid (**4**, 216.1 mg), aucubin (255.2 mg), and fractions 1a and 1b. Fraction 1a (760.4 mg) was subjected to Sephadex LH-20 column chromatography eluted with $H_2O$, MeOH, and $H_2O$−acetone and then purified by Chromatorex ODS with a gradient of $H_2O$−MeOH (1:0 → 0:1) to furnish asperulosidic acid (9.3 mg). Fraction 1b (37.2 mg) was subjected to silica gel 60 column chromatography with a gradient of $CHCl_3$−MeOH−$H_2O$ (8:2:0.2 → 0:1:0) to give chlorogenic acid (3.7 mg). A part (1.0 g) of fraction 2 (17.28 g) was chromatographed over silica gel 60 with a gradient of $CHCl_3$−MeOH−$H_2O$ (20:1:0 → 0:1:0) to give asperuloside (275.0 mg) and fractions 2a and 2b. Fraction 2a (107.7 mg) was subjected to silica gel 60 column chromatography with a gradient of $CHCl_3$−MeOH−$H_2O$ (8:2:0.2 → 0:1:0) and then purified by HPLC with 20% MeOH to give scandoside 10-*O*-glucopyranoside (3.8 mg) and asperulosidic acid (6.0 mg). Fraction 3 (3.8 g) was chromatographed over Chromatorex ODS with a gradient of $H_2O$−MeOH (1:0 → 0:1) to give eucomosides A (**1**, 12.0 mg) and B (**2**, 4.0 mg). A part (2.0 g) of fraction 4 (9.0 g) was subjected to Sephadex LH-20 column chromatography eluted with $H_2O$−MeOH (5:5 → 0:1) to give astragalin (55.3 mg), quercetin 3-*O*-glucopyranoside (133.9 mg), and fractions 4a and 4b. Fraction 4a (327.1 mg) was purified by silica gel 60 column chromatography with a gradient of $CHCl_3$−MeOH−$H_2O$ (15:1:0 → 0:1:0) to furnish **3** (3.0 mg). Fraction 4b (441.2 mg) was chromatographed over Chromatorex ODS with a gradient of $H_2O$−MeOH (3:7 → 0:1) to give quercetin 3-*O*-sambubioside (155.2 mg), fraction 4b-1, and fraction 4b-2. A part (10.0 mg) of fraction 4b-1 (94.3 mg) was subjected to HPLC with 35% MeOH to give rutin (1.7 mg). Fraction 4b-2 (94.3 mg) was subjected to HPLC with 50% MeOH to give kaempferol 3-*O*-rutinoside (6.0 mg).

**EucomosideA (1):** white powder; $[\alpha]_D$ +99.1 (*c* 0.1, MeOH); $^1$H NMR (in $C_5D_5N$, 500 MHz) $\delta$ 2.07 (3H, s, acetyl), 3.30 (2H, d, $J$ = 8.5 Hz, H-9), 3.54 (1H, dd, $J$ = 2.5, 12.8 Hz, H-4), 3.72 (1H, m, H-5), 4.03 (2H, m, glc H-2, 5), 4.23 (1H, dd, $J$ = 9.2, 9.2 Hz, glc H-4), 4.29 (1H, dd, $J$ = 8.6, 9.2 Hz, glc H-3), 4.39 (1H, dd, $J$ = 5.5, 11.6 Hz, glc H-6a), 4.60 (1H, dd, $J$ = 2.1, 12.2 Hz, glc H-6b), 4.78 (1H, d, $J$ = 14.7 Hz, H-10*a*), 4.85 (1H, d, $J$ = 14.7 Hz, H-10*b*), 5.32 (1H, d, $J$ = 7.3 Hz, glc H-1), 5.55 (1H, d, $J$ = 7.3 Hz, H-6), 5.67 (1H, d, $J$ = 1.2 Hz, H-1), 5.87 (1H, d, $J$ = 2.4 Hz, H-3), 5.90 (1H, s, H-7); $^{13}$C NMR (in $C_5D_5N$, 125 MHz), see Table 1; positive FABMS *m*/*z* 415 $[M + H]^+$; HRESIMS *m*/*z* 437.1076 $[M + Na]^+$ (calcd for $C_{18}H_{22}NaO_{11}$, 437.1059). Compound **1** (1.0 mg) in 2 N HCl/$H_2O$ (0.2 mL) was heated at 90 °C for 0.5 h. The reaction mixture was neutralized with Amberlite MB-3, filtered, and evaporated under reduced pressure to give a residue. The residue in MeOH was analyzed by HPLC under the following condition: column, YMC pack Polyamine II (YMC Co., Ltd., 4.6 mm i.d. × 250 mm); solvent, 80% $CH_3CN$; flow rate, 0.8 mL/min; column temperature, 45 °C; detector, JASCO OR-2090 plus; pump, JASCO PU-2080; column oven, JASCO CO-2060. The retention time and optical activity of the sample were identical with those [$t_R$ (min): 6.7; optical activity: positive] of D-glucose. The aglycon could not be detected by TLC.

**Eucomoside B (2):** white powder; $[\alpha]_D$ −10.6 (*c* 0.1, $CH_3CN$); $^1$H NMR (in $CD_3OD$, 500 MHz) $\delta$ 1.88 (1H, dd, $J$ = 7.9, 15.9 Hz, H-6*a*), 2.50 (1H, dd, $J$ = 7.9, 15.3 Hz, H-6*b*), 2.72 (1H, dd, $J$ = 9.2, 9.2 Hz, H-9), 3.09 (1H, dd, $J$ = 7.3, 14.0 Hz, H-7′a), 3.20 (1H, m, H-5), 3.22−3.31 (4H, m, glc H-2, 4, 5, H-7′*b*), 3.39 (1H, dd, $J$ = 9.2, 9.2 Hz, glc H-3), 3.65 (1H, dd, $J$ = 5.5, 12.2 Hz, glc H-6*a*), 3.85 (1H, dd, $J$ = 1.8,

12.2 Hz, glc H-6b), 4.16 (1H, d, $J$ = 14.0 Hz, H-10a), 4.29 (1H, d, $J$ = 14.0 Hz, H-10b), 4.60 (1H, m, H-8′), 4.69 (1H, d, $J$ = 7.3 Hz, glc H-1), 5.10 (1H, d, $J$ = 7.3 Hz, H-1), 5.69 (1H, s, H-7), 7.14 (1H, dd, $J$ = 1.2 Hz, H-3), 7.17 (1H, s, H′-4), 7.20 (2H, s, H-2′, 6′), 7.21 (2H, s, H-3′, 5′); $^{13}$C NMR (in $CD_3OD$, 125 MHz), see Table 2; positive FABMS $m/z$ 544 $[M + Na]^+$; HRFABMS $m/z$ 544.1790 $[M + Na]^+$ (calcd for $C_{25}H_{31}NNaO_{11}$, 544.1794).

**Conjugated Compound (6a) of Geniposidic acid and L-Phenylalanine Methyl Ester.** To a solution of geniposidic acid (**4**) (30.0 mg, 0.080 mmol) in DMF (2.0 mL) were added L-phenylalanine methyl ester (27.7 mg, 0.128 mmol), $Et_3N$ (0.03 mL, 0.215 mmol), HOBt (28.5 mg, 0.174 mmol), and 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (WSC, 40.0 mg, 0.209 mmol). After being stirred for 17 h at room temperature, the solvent was removed in vacuo. It was purified by column chromatography (silica gel) using a 14:2:0.2 mixture of $CHCl_3$−MeOH−$H_2O$ as eluent to obtain **6a** (42.0 mg, 98% yield) as a white powder: $[\alpha]_D$ −39.2 ($c$ 0.1, MeOH); $^1$H NMR (in $CD_3OD$, 500 MHz) $\delta$ 1.91 (1H, m, H-6a), 2.56 (1H, dd, $J$ = 8.6, 15.9 Hz, H-6b), 2.71 (1H, m, H-9), 3.01 (1H, dd, $J$ = 9.8, 13.4 Hz, H′-7*a*), 3.19−3.39 (6H, m, H-5, glc H-2, 3, 4, 5, H′-7*b*), 3.64 (1H, m, glc H-6a), 3.71 (3H, s, OMe), 3.85 (1H, dd, $J$ = 1.8, 11.6 Hz, glc H-6b), 4.16 (1H, d, $J$ = 14.0 Hz, H-10a), 4.29 (1H, d, $J$ = 14.0 Hz, H-10b), 4.68 (1H, d, $J$ = 7.9 Hz, glc H-1), 4.71 (1H, dd, $J$ = 5.5, 9.2 Hz, H-8′), 5.10 (1H, d, $J$ = 7.3 Hz, H-1), 5.72 (1H, s, H-7), 7.10 (1H, s, H-3), 7.21−7.30 (5H, m, H-2′, 3′, 4′, 5′, 6′); $^{13}$C NMR (in $CD_3OD$, 125 MHz) $\delta$ 36.1, 38.0, 39.0, 47.3, 52.7, 55.3, 61.4, 62.7, 71.6, 74.9, 77.9, 78.4, 97.8, 100.3, 115.8, 127.9 × 2, 129.5 × 2, 130.2 × 2, 138.4, 144.8, 148.7, 170.2, 173.9; positive FABMS $m/z$ 558 $[M + Na]^+$.

**Deprotection of 6a.** To a solution of **6a** (42.0 mg, 0.079 mmol) in MeOH (1.0 mL) was added 0.5 N NaOH (1.0 mL). After being stirred for 5 h at room temperature, the mixture was purified by column chromatography (Shephadex LH-20) using MeOH as eluent. Compound **7a** was obtained as white powder (18.8 mg, 46% yield): $[\alpha]_D$ −52.0 ($c$ 0.1, $CH_3CN$); $^1$H NMR (in $CD_3OD$, 500 MHz) $\delta$ 1.86 (1H, m, H-6a), 2.48 (1H, dd, $J$ = 7.9, 16.5 Hz, H-6b), 2.70 (1H, dd, $J$ = 7.3, 7.3 Hz, H-9), 3.08 (1H, dd, $J$ = 7.3, 13.4 Hz, H-7′a), 3.21 (1H, m, H-5), 3.28−3.32 (4H, m, glc H-2, 4, 5, H-7′b), 3.38 (1H, dd, $J$ = 8.3, 9.2 Hz, glc H-3), 3.64 (1H, m, glc H-6a), 3.85 (1H, dd, $J$ = 1.8, 12.2 Hz, glc H-6b), 4.15 (1H, d, $J$ = 14.0 Hz, H-10a), 4.28 (1H, d, $J$ = 14.0 Hz, H-10b), 4.60 (1H, dd, $J$ = 5.8, 7.3 Hz, H′-8), 4.68 (1H, dd, $J$ = 2.4, 7.9 Hz, glc H-1), 5.10 (1H, dd, $J$ = 2.4, 7.3 Hz, H-1), 5.68 (1H, s, H-7), 7.13 (1H, d, $J$ = 1.2 Hz, H-3), 7.15−7.25 (5H, m, H-2′, 3′, 4′, 5′, 6′); $^{13}$C NMR (in $CD_3OD$, 125 MHz) $\delta$ 36.2, 38.7, 39.2, 47.4, 56.8, 61.4, 62.7, 71.6, 75.0, 77.9, 78.4, 97.9, 100.4, 115.8, 127.5, 127.9, 129.3 × 2, 130.7 × 2, 139.3, 144.9, 148.8, 169.2, 175.0; positive FABMS $m/z$ 566 [M (COONa) + Na]$^+$.

**Conjugated Compound (6b) of Geniposidic Acid and D-Phenylalanine Methyl Ester.** Compound **6b** was prepared in a similar way to **6a**, using **4** (28.9 mg, 0.077 mmol), D-phenylalanine methyl ester (28.0 mg, 0.129 mmol), $Et_3N$ (0.03 mL, 0.215 mmol), HOBt (23.5 mg, 0.174 mmol), and 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (WSC, 40.0 mg, 0.209 mmol) in DMF (2.0 mL). Compound **6b** was obtained as a white powder (35.0 mg, 85% yield): $[\alpha]_D$ −21.2 ($c$ 0.1, MeOH); $^1$H NMR (in $CD_3OD$, 500 MHz) $\delta$ 1.89 (1H, m, H-6*a*), 2.59 (1H, dd, $J$ = 8.5, 15.9 Hz, H-6b), 2.73 (1H, m, H-9), 3.04 (1H, m, H′-7a), 3.18−3.38 (6H, m, H-5, glc H-2, 3, 4, 5, H′-7b), 3.64 (1H, m, glc H-6a), 3.70 (3H, s, OMe), 3.85 (1H, dd, $J$ = 1.8, 11.6 Hz, glc H-6*b*), 4.16 (1H, d, $J$ = 14.7 Hz, H-10a), 4.29 (1H, d, $J$ = 15.3 Hz, H-10b), 4.68 (1H, dd, $J$ = 2.4, 7.9 Hz, glc H-1), 4.73 (1H, dd, $J$ = 5.5, 9.2 Hz, H′-8), 5.12 (1H, dd, $J$ = 2.4, 7.3 Hz, H-1), 5.72 (1H, s, H-7), 7.17−7.27 (6H, m, H-2, 3, 4, 5, 6, H-3); $^{13}$C NMR (in $CD_3OD$, 125 MHz) $\delta$ 36.1, 38.2, 39.1, 47.4, 52.7, 55.1, 61.3, 62.7, 71.6, 74.9, 77.9, 78.4, 97.7, 100.3, 115.9, 127.8, 128.2, 129.5 × 2, 130.2 × 2, 138.4, 144.8, 148.3, 169.8, 173.7; positive FABMS $m/z$ 558 $[M + Na]^+$.

**Deprotection of 6b.** Compound **7b** was prepared in a similar way to **7a**, using **6b** (35.0 mg, 0.065 mmol), and **6b** was obtained as a white powder (25.0 mg, 73% yield): $[\alpha]_D$ −40.2 ($c$ 0.5, $CH_3CN$); $^1$H NMR (in $CD_3OD$, 500 MHz) $\delta$ 1.94 (1H, m, H-6a), 2.68 (1H, dd, $J$ = 7.9, 7.9 Hz, H-9), 3.08 (1H, m, H-7′a), 3.11 (1H, m, H-6b), 3.21 (1H, m, H-5), 3.26−3.31 (4H, m, glc H-2, 4, 5, H-7b), 3.38 (1H, dd, $J$ = 7.9, 8.6 Hz, glc H-3), 3.63 (1H, dd, $J$ = 5.2, 11.6 Hz, glc H-6a), 3.84 (1H, dd, $J$ = 1.2, 11.6 Hz, glc H-6b), 4.16 (1H, d, $J$ = 14.7 Hz, H-10a), 4.28 (1H, d, $J$ = 14.7 Hz, H-10b), 4.55 (1H, dd, $J$ = 6.1, 6.7 Hz, H′-8), 4.68 (1H, dd, $J$ = 3.1, 7.9 Hz, glc H-1), 5.10 (1H, d, $J$ = 2.4, 7.3 Hz, H-1), 5.72 (1H, s, H-7), 7.12−7.25 (6H, m, H-3, H-2′, 3′, 4′, 5′, 6′); $^{13}$C NMR (in $CD_3OD$, 125 MHz) $\delta$ 36.5, 39.0, 39.3, 47.4, 57.2, 61.5, 62.7, 71.6, 75.0, 77.9, 78.4, 98.0, 100.3, 116.2, 127.4, 128.0, 129.2 × 2, 130.7 × 2, 139.5, 145.0, 148.1, 169.2, 178.5; positive FABMS $m/z$ 544 $[M + Na]^+$.

**Table 3.** NMR Spectroscopic Data (in $CD_3OD$) for Eucomoside C (**3**)

| position | $\delta_C$ | $\delta_H$ ($J$ in Hz) | HMBC |
|---|---|---|---|
| 1 | 97.7 | 5.06 d (7.3) | Glc H-1 |
| 3 | 150.2 | 7.11 s | H-1, H-4, H-5, |
| 4 | 115.5 | | H-11 |
| 5 | 36.0 | 3.13 m | H-1, H-3, H-4, H-6, H-9 |
| 6a | 38.8 | 1.63 d (7.9, 15.9) | |
| 6b | | 2.24 dd (7.9, 15.3) | |
| 7 | 127.9 | 5.58 s | H-5, H-6, H-9, H-10 |
| 8 | 144.7 | | |
| 9 | 47.4 | 2.67 dd (9.2, 9.2) | H-1, H-6, H-7, H-8 |
| 10a | 61.3 | 4.12 d (14.7) | H-7, H-8 |
| 10b | | 4.29 d (14.7) | |
| 11 | 169.9 | | |
| Glc 1 | 100.3 | 4.65 d (7.9) | |
| Glc 2 | 74.9 | 3.20 m | |
| Glc 3 | 77.9 | 3.37 m | |
| Glc 4 | 71.6 | 3.27 m | |
| Glc 5 | 78.4 | 3.27 m | |
| Glc 6a | 62.7 | 3.63 dd (5.5, 11.6) | |
| Glc 6b | | 3.84 dd (1.8, 12.2) | |
| 2′ | 124.4 | 7.09 s | H-8, H-9 |
| 3′ | 111.8 | | H-4, H-8 |
| 4′ | 119.6 | 7.57 d (7.9) | H-3, H-6, H-9 |
| 5′ | 119.8 | 6.98 dd (7.3, 7.9) | H-7, H-8 |
| 6′ | 122.4 | | H-4 |
| 7′ | 112.2 | 7.30 d (8.6) | |
| 8′ | 129.0 | | |
| 9′ | 138.1 | | |
| 10a | 28.3 | 3.20 m | H-3, H-8, H-11 |
| 10b | | 3.43 dd (4.3, 14.7) | |
| 11′ | 57.5 | 4.61 d (6.7) | |
| 12′ | 180.0 | | |

**Eucomoside C (3):** white powder; $[\alpha]_D$ −13.1 ($c$ 0.5, $H_2O$); $^1$H NMR (in $CD_3OD$, 500 MHz) $\delta$ 1.63 (1H, dd, $J$ = 7.9, 15.9 Hz, H-6a), 2.24 (1H, dd, $J$ = 7.9, 15.3 Hz, H-6b), 2.67 (1H, dd, $J$ = 9.2, 9.2 Hz, H-9), 3.13 (1H, m, H-5), 3.18−3.21 (2H, m, glc H-2, H′-10a), 3.26−3.27 (2H, m, glc H-4, 5), 3.37 (1H, m, glc H-3), 3.43 (1H, dd, $J$ = 4.3, 14.7 Hz, H-10′b), 3.63 (1H, dd, $J$ = 5.5, 11.6 Hz, glc H-6a), 3.84 (1H, dd, $J$ = 1.8, 12.2 Hz, glc H-6b), 4.12 (1H, d, $J$ = 14.7 Hz, H-10*a*), 4.29 (1H, d, $J$ = 14.7 Hz, H-10*b*), 4.61 (1H, d, $J$ = 6.7 Hz, H-11′), 4.65 (1H, d, $J$ = 7.9 Hz, glc H-1), 5.06 (1H, d, $J$ = 7.3 Hz, H-1), 5.58 (1H, s, H-7), 6.98 (1H, dd, $J$ = 7.3, 7.9 Hz, H-5′), 7.06 (1H, dd, $J$ = 7 0.3, 7.9 Hz, H′-6), 7.09 (1H, s, H-2′), 7.11 (1H, s, H-3), 7.30 (1H, d, $J$ = 8.6 Hz, H-7′), 7.57 (1H, d, $J$ = 7.9 Hz, H′-4); $^{13}$C NMR (in $CD_3OD$, 125 MHz), see Table 3; HRESIMS $m/z$ 559.1980 $[M - H]^+$ (calcd for $C_{27}H_{31}N_2O_{11}$, 559.1927).

**Conjugated Compound (6c) of Geniposidic Acid and L-Tryptophan Methyl Ester.** Compound **6c** was prepared in a similar way to **6a**, using **4** (27.0 mg, 0.072 mmol), L-tryptophan methyl ester (18.0 mg, 0.071 mmol), $Et_3N$ (0.03 mL, 0.215 mmol), HOBt (18.5 mg, 0.137 mmol), and WSC (35.0 mg, 0.183 mmol) in DMF (2.0 mL). Compound **6c** was obtained as a white powder (27.8 mg, 67% yield): $[\alpha]_D$ −60.9 ($c$ 0.5 DMSO); $^1$H NMR (in $CD_3OD$, 500 MHz) $\delta$ 1.85 (1H, m, H-6a), 2.45 (1H, dd, $J$ = 8.5, 15.9 Hz, H-6b), 2.71 (1H, dd, $J$ = 7.3, 7.9 Hz, H-9), 3.18−3.37 (7H, m, glc H-2, 3, 4, 5, H-5, H-10′a, 10′b), 3.63 (1H, m, glc H-6a), 3.71 (1H, s, OMe), 3.85 (1H, dd, $J$ = 1.8, 11.0 Hz, glc H-6b), 4.15 (1H, d, $J$ = 14.0 Hz, H-10a), 4.27 (1H, d, $J$ = 14.0 Hz, H-10b), 4.67 (1H, d, $J$ = 4.3, 8.6 Hz, H-11′), 4.77 (1H, dd, $J$ = 5.5, 8.6 Hz, glc H-1), 5.10 (1H, dd, $J$ = 4.3, 7.3 Hz, H-1), 5.67 (1H, s, H-7), 7.01 (1H, dd, $J$ = 7.2, 7.9 Hz, H-5′), 7.08−7.10 (3H, m, H-3, H-2′, 6′), 7.34 (1H, dd, $J$ = 4.3, 7.9 Hz, H-7′), 7.53 (1H, dd, $J$ = 4.3, 7.9 Hz, H-4′); $^{13}$C NMR (in $CD_3OD$, 125 MHz) $\delta$ 28.1, 36.1, 39.0, 47.5, 52.6, 55.0, 61.4, 62.8, 71.6, 75.0, 77.9, 78.4, 97.7, 100.3, 110.9, 112.5, 115.8, 119.2, 120.0, 122.6, 124.5, 128.0, 128.8, 138.2, 144.7, 148.8, 170.2, 174.4; positive FABMS $m/z$ 575 $[M + H]^+$.

**Deprotection of 6c.** Compound **7c** was prepared in a similar way to **7a**, using **6c** (27.8 mg, 0.048 mmol). Compound **7c** was obtained as

a white powder (11.0 mg, 41% yield): $[\alpha]_D$ −0.1 (*c* 0.5, MeOH); $^1$H NMR (in $CD_3OD$, 500 MHz) $\delta$ 1.61 (1H, m, H-6a), 2.21 (1H, dd, $J$ = 8.5, 15.9 Hz, H-6b), 2.67 (1H, dd, $J$ = 6.7, 7.9 Hz, H-9), 3.13 (1H, dd, $J$ = 8.3, 15.3 Hz, H-5), 3.20 (1H, dd, $J$ = 7.9, 8.5 Hz, glc H-2), 3.24−3.40 (3H, m, glc H-4, 5, H-10′a), 3.37 (1H, m, glc H-3), 3.45 (1H, dd, $J$ = 4.9, 14.9 Hz, H-10′b), 3.64 (1H, dd, $J$ = 5.2, 12.0 Hz, glc H-6*a*), 3.84 (1H, dd, $J$ = 1.8, 11.6 Hz, glc H-6*b*), 4.12 (1H, d, $J$ = 14.7 Hz, H-10a), 4.24 (1H, d, $J$ = 14.7 Hz, H-10b), 4.63 (1H, d, $J$ = 5.2, 6.7 Hz, H′-11), 4.66 (1H, d, $J$ = 4.3, 7.3 Hz, glc H-1), 5.01 (1H, dd, $J$ = 4.3, 7.3 Hz, H-1), 5.56 (1H, s, H-7), 6.95 (1H, dd, $J$ = 7.3, 7.3 Hz, H-5′), 7.03 (1H, dd, $J$ = 7.3, 7.7 Hz, H-6′), 7.06 (1H, d, $J$ = 3.7 Hz, H-2′), 7.08 (1H, d, $J$ = 4.3 Hz, H-3), 7.29 (1H, dd, $J$ = 4.9, 7.9 Hz, H-7′), 7.57 (1H, d, $J$ = 3.7, 8.5 Hz, H-4′); $^{13}$C NMR (in $CD_3OD$, 125 MHz) $\delta$ 28.5, 36.2, 38.8, 47.5, 57.0, 61.4, 62.7, 71.6, 75.0, 77.9, 78.4, 97.8, 100.3, 112.0 × 2, 115.9, 119.6, 119.8, 122.2, 124.5, 128.0, 129.7, 137.9, 144.6, 148.7, 168.9, 179.0; positive FABMS *m*/*z* 583 $[M + Na]^+$.

**Conjugated Compound (6d) of Geniposidic Acid and D-Tryptophan Methyl Ester.** Compound **6d** was prepared in a similar way to **6a**, using **4** (27.0 mg, 0.072 mmol), D-tryptophan methyl ester (25.4 mg, 0.100 mmol), $Et_3N$ (0.03 mL, 0.215 mmol), HOBt (36.5 mg, 0.270 mmol), and WSC (69.0 mg, 0.360 mmol) in DMF (2.0 mL). Compound **6d** was obtained as a white powder (34.3 mg, 83% yield): $[\alpha]_D$ −20.3 (*c* 0.5, MeOH); $^1$H NMR (in $CD_3OD$, 500 MHz) $\delta$ 1.89 (1H, m, H-6a), 2.47 (1H, dd, $J$ = 8.6, 15.9 Hz, H-6b), 2.70 (1H, dd, $J$ = 7.3, 14.7 Hz, H-9), 3.12 (1H, dd, $J$ = 7.9, 16.5 Hz, H-5), 3.20−3.40 (6H, m, glc H-2, 3, 4, 5, H-10′a, 10′b), 3.64 (1H, dd, $J$ = 5.5, 11.0 Hz, glc H-6a), 3.71 (1H, s, OMe), 3.85 (1H, dd, $J$ = 1.8, 11.6 Hz, glc H-6b), 4.16 (1H, d, $J$ = 13.4 Hz, H-10a), 4.28 (1H, d, $J$ = 14.0 Hz, H-10b), 4.68 (1H, d, $J$ = 3.7, 7.9 Hz, H-11′), 4.80 (1H, dd, $J$ = 5.5, 7.9 Hz, glc H-1), 5.11 (1H, dd, $J$ = 3.7, 6.7 Hz, H-1), 5.68 (1H, s, H-7), 7.00 (1H, m, H-5′), 7.06−7.10 (2H, m, H-2′, 6′), 7.20 (1H, m, H-3), 7.34 (1H, d, $J$ = 3.7, 7.9 Hz, H-7′), 7.52 (1H, dd, $J$ = 3.7, 7.9 Hz, H-4′); $^{13}$C NMR spectral data (in $CD_3OD$, 125 MHz) $\delta$ 28.2, 36.2, 39.1, 47.4, 52.7, 55.0, 61.4, 62.8, 71.7, 75.0, 78.0, 78.4, 97.8, 100.3, 110.8, 112.4, 115.7, 119.2, 120.0, 122.6, 124.5, 128.0, 128.9, 138.1, 144.8, 148.8, 169.8, 174.1; positive FABMS *m*/*z* 597 $[M + Na]^+$.

**Deprotection of 6d.** Compound **7d** was prepared in a similar way to **6a** using **6d** (34.3 mg, 0.059 mmol). Compound **7d** was obtained as a white powder (24.3 mg, 73% yield): $[\alpha]_D$ −62.2 (*c* 0.5, MeOH); $^1$H NMR (in $CD_3OD$, 500 MHz) $\delta$ 1.93 (1H, m, H-6a), 2.61 (2H, m, H-6b, H-9), 2.95 (1H, dd, $J$ = 7.9, 17.1 Hz, H-5), 3.19−3.38 (5H, m, glc H-2, 3, 4, 5, H-10′*a*), 3.42 (1H, dd, $J$ = 5.5, 13.4 Hz, H-10′b), 3.63 (1H, dd, $J$ = 5.5, 12.8 Hz, glc H-6a), 3.83 (1H, dd, $J$ = 2.4, 12.2 Hz, glc H-6b), 4.14 (1H, d, $J$ = 14.0 Hz, H-10a), 4.27 (1H, d, $J$ = 14.6 Hz, H-10b), 4.62 (1H, dd, $J$ = 5.5, 9.1 Hz, H′-11), 4.69 (1H, dd, $J$ = 2.4, 7.9 Hz, glc H-1), 5.03 (1H, dd, $J$ = 3.1, 7.9 Hz, H-1), 5.68 (1H, s, H-7), 7.01 (1H, dd, $J$ = 7.6, 7.9 Hz, H-6′), 7.06 (2H, s, H-2′, H-3′), 7.27 (1H, dd, $J$ = 3.1, 11.6 Hz, H-7′), 7.53 (1H, dd, $J$ = 3.7, 7.9 Hz, H-4′); $^{13}$C NMR (in $CD_3OD$, 125 MHz) $\delta$ 28.9, 36.6, 39.3, 47.3, 57.2, 61.5, 62.7, 71.6, 75.0, 77.9, 78.4, 98.1, 100.4, 112.0, 112.1, 116.0, 119.5, 119.8, 122.1, 124.6, 128.0, 129.7, 137.9, 144.9, 148.5, 168.9, 178.8; positive FABMS *m*/*z* 583 $[M + Na]^+$.

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