Pyronane Monoterpenoids from the Fruit of *Gardenia jasminoides*

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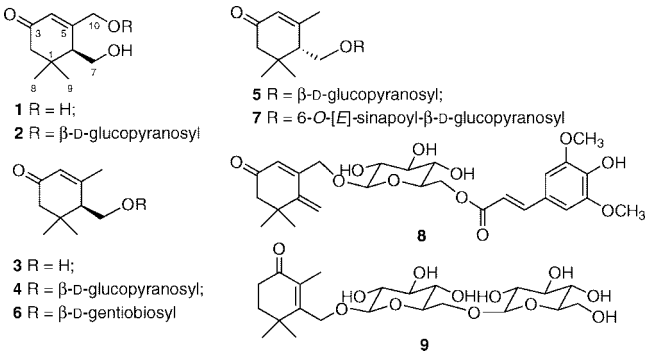
Five new pyronane-type monocyclic monoterpenoids, jasminodiol (**1**), jasminoside H (**6**), 6′-*O*-sinapoyljasminoside A (**7**), 6′-*O*-sinapoyljasminoside C (**8**), and jasminoside I (**9**), together with four known analogues, were isolated from the fruit of *Gardenia jasminoides*. The structures of the new metabolites were characterized using spectroscopic data, and the absolute configurations of **1**, **6**, and **7** were established using circular dichroism (CD) analysis. Compound **1** showed tyrosinase inhibitory activity (IC_{50} 2.2 mM).

The fruit of *Gardenia jasminoides* Ellis (Rubiaceae) is widely used as traditional medicine in many Asian countries for its cholagogue, sedative, diuretic, antiphlogistic, homeostatic, and antipyretic effects.[1] Previous investigations of *G. jasminoides* revealed the presence of a variety of terpenoids, including monocyclic monoterpenoids and their glycosides,[2,3] iridoids and their glycosides,[4–6] triterpenoids, crocetin and its glycosides,[7–9] vanillic acid glycosides, and quinic acid.[10] The present study describes the structural elucidation of five new pyronane monoterpenoids from the EtOAc- and *n*-BuOH-soluble fractions, partitioned from the MeOH extract of the fruit of *G. jasminoides*, as well as their relative tyrosinase inhibitory activities.

Results and Discussion

The MeOH extract of the fruit of *G. jasminoides* was partitioned into *n*-hexane-, EtOAc-, and *n*-BuOH-soluble fractions. From the EtOAc- and *n*-BuOH-soluble fractions, chromatography produced nine compounds (**1**−**9**). Four of these compounds were identified as jasminoside B (**2**), crocusatin-C (**3**), epijasminoside A (**4**), and jasminoside A (**5**).[2,11]



Compound **1** was obtained as an optically active, colorless oil, $[\alpha]^{22}_D$ −54.5 (*c* 0.22, MeOH). Its HREIMS exhibited a molecular ion at *m/z* 184.1098, corresponding to a molecular formula of $C_{10}H_{16}O_3$, with three degrees of unsaturation. The IR spectrum indicated the presence of hydroxy (3390 cm^{-1}) and ketone (1650 cm^{-1}) groups. The ^1H and ^{13}C NMR data of **1** were similar to those of jaminoside B (**2**),[2] except for the absence of the sugar moiety (Table 1). Combined analysis of ^1H and ^{13}C NMR spectra of **1** revealed the presence of an α,β-unsaturated ketone [δ 6.17 (1H, s, H-4), δ 123.9 (C-4), 168.6 (C-5), and 202.9 (C-3)], a methylene group adjacent to the α,β-unsaturated ketone, two oxygenated methylene groups, and a *gem*-dimethyl attached to a quaternary carbon. This combined data indicated that **1** was a monocyclic monoterpenoid. The locations of two oxygenated methylene groups were determined by the oxygenated methylene protons at δ 3.84 (2H, br d, J = 4.2 Hz, H-7), which showed HMBC correlations with C-1 (δ 36.5), C-5 (δ 168.6), and C-6 (δ 50.7), and the oxygenated methylene protons at δ 4.40 (1H, dd, J = 17.4, 1.2 Hz, H-10a)/4.20 (1H, dd, J = 17.4, 1.2 Hz, H-10b), correlating with C-4, C-5, and C-6. The absolute configuration at C-6 of **1** was established as *S*, based on the negative Cotton effects at 328.0 nm (Δε −0.35), 249.0 nm (Δε −1.17), and 211.0 nm (Δε −1.39) in the CD spectrum, compared with those of jasminoside B (**2**).[2] Therefore, the structure of **1** was established as a monocyclic monoterpenoid and was named jasminodiol.

Compound **6** was isolated as an optically active, white, amorphous powder, $[\alpha]^{22}_D$ −70.0 (*c* 0.22, MeOH). Its HRFABMS exhibited a molecular ion at *m/z* 493.2253 $[M + H]^+$, in agreement with a molecular formula of $C_{22}H_{36}O_{12}$. The spectroscopic data of **6** were similar to those of epijasminoside A (**4**), except for the obvious differences due to the presence of additional glucosyl signals in the ^1H and ^{13}C NMR spectra (Table 2). After enzymatic hydrolysis of **6**, the sugar was identified as D-glucose by TLC and GC analysis.[12] Two anomeric proton resonances at δ 4.22 (1H, d, J = 7.8 Hz, H-1′) and 4.38 (1H, d, J = 7.8 Hz, H-1″) in the ^1H NMR spectrum of **6** indicated β-glycosidic linkages. The deshielded methylene carbon signal at δ 70.3 (C-6′) in the ^{13}C NMR spectrum of **6** suggested that two sugar moieties formed a β-gentiobiosyl moiety by a 1″→6′ connection, which was further confirmed by observation of the correlation between H-1″ and C-6′ in the HMBC spectrum. In addition, H-1′ showed long-range HMBC correlations with the oxygenated methylene carbon C-7 (δ 69.4), indicating that the β-gentiobiose was linked to the 7-OH of the aglycone (Figure 1). The CD spectrum of **6** showed negative Cotton effects at 329.0 nm (Δε −0.25), 240.0 nm (Δε −1.58), and 205.0 nm (Δε −2.21), suggesting the *S* absolute configuration at C-6.[2] Accordingly, the structure of **6** was elucidated to be a monoterpenoid diglucoside, named jasminoside H.

Compound **7** was obtained as an optically active, pale yellow, amorphous powder, $[\alpha]^{22}_D$ +50.0 (*c* 0.40, MeOH). Its molecular formula, $C_{27}H_{36}O_{11}$, was established by HRFABMS, which showed a quasi-molecular ion at *m/z* 537.2275 $[M + H]^+$. The presence of hydroxy groups (3360 cm^{-1}), an aromatic ring (1608, 1520 cm^{-1}), and conjugated unsaturated carbonyl groups (1710, 1680, 1615 cm^{-1}) was indicated by the IR spectrum. Two overlapping proton singlets at δ 6.92 (2H, s, H-5″, 9″) and 3.88 (6H, s, 6″, 8″-OCH_3) observed in the ^1H NMR spectrum of **7**, together with an overlapping methine carbon signal at δ 107.1 (C-5″, 9″), an overlapping quaternary carbon signal at δ 148.7 (C-6″, 8″), and

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Table 1. NMR Spectroscopic Data of Compounds **1** and **2**

	1			**2**	
position	δ_C, mult.	δ_H (J in Hz)	HMBC	δ_C, mult.	δ_H (J in Hz)
1	36.5, qC			36.5, qC	
2	50.1, CH_2	2.74, d (17.1)	1, 3, 4, 6, 8, 9	50.1, CH_2	2.75, d (17.1)
		2.03, d (17.1)	1, 3, 4, 6, 8, 9		2.03, d (17.1)
3	202.9, qC			202.9, qC	
4	123.9, CH	6.17, s	2, 6, 10	125.4, CH	6.28 br s
5	168.6, qC			164.4, qC	
6	50.7, CH	2.08, t (4.2)	1, 2, 4, 5, 7, 9, 10	50.8, CH	2.14, t (3.9)
7	62.5, CH_2	3.84 br d (4.2)	1, 5, 6	62.3, CH_2	3.87, m
8	27.5, CH_3	1.15, s	1, 2, 6, 9	27.5, CH_3	1.15, s
9	29.4, CH_3	1.03, s	1, 2, 6, 8	29.4, CH_3	1.04, s
10	65.1, CH_2	4.40, dd (17.4, 1.2)	4, 5, 6	71.7, CH_2	4.70, dd (16.5, 1.5)
		4.20, dd (17.4, 1.2)	4, 5, 6		4.32, dd (16.5, 1.5)
1′				104.5, CH	4.35, d (7.5)
2′				75.2, CH	3.25, m
3′				78.2, CH	3.34, m
4′				71.8, CH	3.28, m
5′				78.3, CH	3.37, m
6′				62.9, CH_2	3.87, m
					3.65, dd (12.0, 4.8)

an overlapping *O*-methyl carbon signal at δ 57.0 (6″, 8″-OCH_3), observed in the ^{13}C NMR and DEPT spectra, suggested the presence of a symmetrical 1,3,4,5-tetrasubstituted benzene ring (Table 2). Additionally, the ^1H and ^{13}C NMR spectra of **7** showed *trans* double-bond signals [δ 6.42 (1H, d, J = 15.9 Hz, H-2″), 115.8 (C-2″), 7.62 (1H, d, J = 15.9 Hz, H-3″), 147.4 (C-3″)], of which H-3″ showed HMBC correlations with C-4″ (δ 126.7), C-5″ (δ 107.1), and C-6″ (δ 148.7). Additionally, H-3″ showed HMBC correlation with the C-1″ carbonyl carbon (δ 169.1) (Figure 1). This data revealed the presence of a sinapoyl group. In addition, the ^1H and ^{13}C NMR spectra of **7** showed typical glucosyl signals. The configuration of the glycosidic linkage unit was determined as β, on the basis of the coupling constant observed for the anomeric proton H-1′ (δ 4.27, d, J = 7.8 Hz). The remaining ^1H and ^{13}C NMR signals were very similar to those of crocusatin-C (**3**), which was initially isolated from the pollen of *Crocus sativus* L.[11] The chemical shifts of the six carbons at δ 104.3 (C-1′), 75.1 (C-2′), 78.3 (C-3′), 71.8 (C-4′), 75.6 (C-5′), and 64.6 (C-6′) in the ^{13}C NMR spectrum of **7** indicated that the sugar was a 1′,6′-disubstituted glucose. In the HMBC spectrum of **7**, the anomeric proton (H-1′) showed long-range correlations with the oxygenated methylene carbon at δ 69.2 (C-7), suggesting that the glucosyl was linked to the 7-OH of crocusatin-C with the same pattern as jasminoside A. Furthermore, the oxygenated methylene protons at δ 4.48 (1H, dd, J = 12.0, 2.4 Hz, H-6′a)/4.38 (1H, dd, J = 12.0, 6.0 Hz, H-6′b) showed long-range HMBC correlations with the carbonyl carbon at δ 169.1 (C-1″) of the sinapoyl group, indicating that the sinapoyl group was connected to the 6′-OH of the glucosyl moiety (Figure 1). The CD spectrum of **7** showed positive Cotton effects at 253.0 nm ($\Delta\epsilon$ +0.65) and 200.0 nm ($\Delta\epsilon$ +1.51), suggesting the 6*R* absolute configuration.[2] Thus, the structure of **7** was assigned as 6′-*O*-sinapoyljasminoside A.

Compound **8** was purified as a pale yellow, amorphous powder. Its molecular formula, $C_{27}H_{34}O_{11}$, was established by HRFABMS, with a mass of $[M + H]^+$ (*m*/*z* 535.2184, calcd 535.2179). The ^1H and ^{13}C NMR spectral data of **8** were similar to those of jasminoside C together with those of sinapoyl moiety in **7**.[2] The β-linkage of the glucosidic unit was consistent with the coupling constant of the anomeric proton at δ 4.39 (1H, d, J = 7.5 Hz, H-1′). The chemical shifts of the six carbons at δ 104.2 (C-1′), 75.2 (C-2′), 78.1 (C-3′), 71.8 (C-4′), 75.7 (C-5′), and 64.7 (C-6′) in the ^{13}C NMR indicated that the sugar was a 1′,6′-disubstituted glucose. The above data suggested that the structure of **8** was jasminoside C with a sinapoyl moiety connected to 6′-OH of the glucosyl moiety. The linkage among glucose and aglycones was deduced by the HMBC correlations between the oxygenated methylene protons at δ 4.67 (1H, dd, J = 15.3, 1.5 Hz, H-10a)/4.53 (1H, dd, J = 15.3, 1.5 Hz, H-10b) and the anomeric carbon (C-1′) of glucose, as well as correlations between the sugar protons at δ 4.49 (1H, dd, J = 12.0, 2.4 Hz, H-6′a)/4.36 (1H, dd, J = 12.0, 6.0 Hz, H-6′b) and the C-1″ carbonyl carbon (δ 169.1) of the sinapoyl group. Therefore, compound **8** was determined to be 6′-*O*-sinapoyljasminoside C.

Compound **9** was obtained as a colorless, amorphous powder. The molecular formula of **9**, $C_{22}H_{36}O_{12}$, was deduced from its HRFABMS, which showed a quasi-molecular ion at *m*/*z* 493.2214 $[M + H]^+$. The NMR data of **9** were similar to those of jasminoside E,[2] except for the presence of additional glucosyl signals in the ^1H and ^{13}C NMR spectra (Table 2). The β-linkages of the glucosidic units were consistent with the coupling constants observed for the anomeric protons at δ 4.32 (1H, d, J = 7.5 Hz, H-1′) and 4.42 (1H, d, J = 7.8 Hz, H-1″). The downfield shift of the methylene carbon at δ 70.3 (C-6′) in the ^{13}C NMR spectrum of **9** suggested that two sugar moieties formed a β-gentiobiosyl moiety via a 1″→6′ connection, which was confirmed by the HMBC correlation between H-1″ and C-6′. Additionally, the anomeric proton (H-1′) showed a long-range HMBC correlation with the oxygenated methylene carbon at δ 67.0 (C-7), indicating that the β-gentiobiose was linked to the 7-OH of aglycone. Thus, the structure of **9** was characterized as a monoterpenoid diglucoside, named jasminoside I.

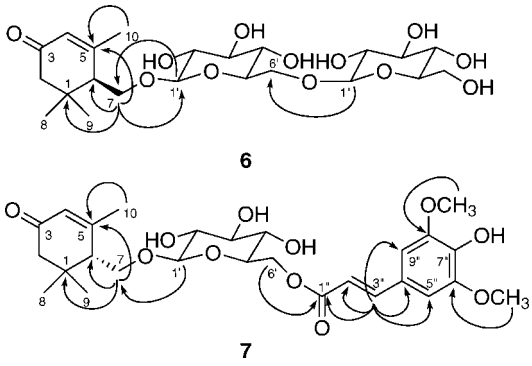
All isolated compounds are pyronane monoterpenoids. Aside from the compounds isolated previously from *G. jasminoides*, many pyronane monoterpenoids are found in *Crocus sativus* L.[2,6,11,13–18] Some of these pyronane monoterpenoids showed significant tyrosinase inhibitory activity.[11,15] Thus, our isolated compounds were evaluated for their antityrosinase activity. The antityrosinase activity of **1** (IC_{50} 2.2 mM) was lower than that of kojic acid (IC_{50} 0.2 mM); however, **1** showed more significant activity than that of hydroquinone (IC_{50} 4.5 mM), a known commercial whitening agent in cosmetics.[19] The other compounds showed no or very weak activities (data not shown). The inhibition kinetics of **1**, with three concentrations (1.0, 2.0, and 3.0 mM), were measured every minute for a total of 50 min, following a preincubation of tyrosinase for 10 min (Figure 2). The results showed a dose-dependent correlation for the tyrosinase inhibitory activity of **1**.

Experimental Section

General Experimental Procedures. Optical rotations were measured on a JASCO DIP-370 polarimeter. UV spectra were obtained using a Beckman Du-650 recording spectrophotometer. CD spectra were measured on a JASCO J-715 CD/ORD spectropolarimeter. FT-IR spectra were obtained using a JASCO Report-100 infrared spectrometer. FT-NMR spectra were recorded on a Bruker DRX-300 spectrometer

Table 2. NMR Spectroscopic Data of Compounds **6**−**9**

	6			**7**			**8**			**9**		
position	δ_C, mult.	δ_H (J in Hz)	HMBC	δ_C, mult.	δ_H (J in Hz)	HMBC	δ_C, mult.	δ_H (J in Hz)	HMBC	δ_C, mult.	δ_H (J in Hz)	HMBC
1	36.5, qC			36.3, qC			39.8, qC			36.7, qC		
2	50.5, CH_2	2.65, d (16.8)	1, 3, 6, 8, 9	49.6, CH_2	2.73, d (17.1)	1, 3, 8, 9	53.0, CH_2	2.35, s	1, 3, 4, 6, 8	38.5, CH_2	1.85, m	1, 3, 4, 6, 8, 9
		2.00, d (16.8)	1, 3, 4, 6, 8, 9		1.96, d (17.1)	1, 3, 6, 9						
3	203.0, qC			203.1, qC			201.8, qC			35.4, CH_2	2.49 br, t (6.6)	1, 2, 4, 5
4	127.6, CH	5.90 br, s	2, 6, 10	127.7, CH	5.86 br, s	2, 6, 10	125.8, CH	6.30 br, s	2, 6, 10	202.2, qC		
5	165.8, qC			165.4, qC			154.7, qC			135.5, qC		
6	52.8, CH	2.25, t (4.2)	1, 2, 4, 5, 7, 9, 10	52.8, CH	2.20, t (3.6)	1, 2, 4, 5, 7, 9, 10	150.1, qC			160.2, qC		
7	69.4, CH_2	4.19, dd (10.8,4.2)	1, 5, 6, 1′	69.2, CH_2	4.13, dd (10.5, 3.6)	1, 5	114.2, CH_2	5.48 br, s	1, 5, 6	67.0, CH_2	4.67, d (10.8)	1, 5, 6, 1′
		3.82, dd (10.8, 4.2)	1, 5, 6, 1′		3.81, dd (10.5, 3.6)	1, 5		5.36, d (1.5)	1, 5, 6		4.23, d (10.8)	1, 5, 6, 1′
8	27.3, CH_3	1.15, s	1, 2, 6, 9	27.3, CH_3	1.12, s	1, 2, 6, 9	28.6, CH_3	1.17, s	1, 2, 6, 9	27.0, CH_3	1.24, s	1, 2, 6, 9
9	29.4, CH_3	1.05, s	1, 2, 6, 8	29.4, CH_3	1.01, s	1, 2, 6, 8	28.7, CH_3	1.16, s	1, 2, 6, 8	27.1, CH_3	1.22, s	1, 2, 6, 8
10	24.5, CH_3	2.10, d (1.2)	4, 5, 6	24.3, CH_3	2.04, d (0.9)	4, 5, 6	69.1, CH_2	4.67, dd (15.3, 1.5)	4, 5, 6, 1′	12.1, CH_3	1.84, s	4, 5, 6
								4.53, dd (15.3, 1.5)	4, 5, 6, 1′			
1′	104.6, CH	4.22, d (7.8)	7	104.3, CH	4.27, d (7.8)	7	104.2, CH	4.39, d (7.5)	10	104.3, CH	4.32, d (7.5)	7, 2′
2′	75.3, CH	3.13, m	1′, 3′	75.1, CH	3.13, m	1′, 3′	75.2, CH	3.30, m	1′, 3′	75.3, CH	3.20, m	1′, 3′, 4′
3′	78.3, CH	3.33, m	2′, 4′	78.3, CH	3.36, m	4′	78.1, CH	3.40, m	4′	78.2, CH	3.38, m	2′, 4′, 5′
4′	71.8, CH	3.31, m	3′, 5′	71.8, CH	3.35, m	3′	71.8, CH	3.37, m	3′	71.9, CH	3.32, m	2′, 3′, 5′
5′	77.4, CH	3.46, m	3′, 4′	75.6, CH	3.51, m	3′, 4′	75.7, CH	3.54, m	3′, 4′	77.4, CH	3.50, m	3′, 4′, 6′
6′	70.3, CH_2	4.15, dd (12.0, 2.1)	4′, 1″	64.6, CH_2	4.48, dd, (12.0, 2.4)	1″	64.7, CH_2	4.49, dd, (12.0, 2.4)	1″	70.3, CH_2	4.18, dd (12.0, 2.1)	4″, 5′, 1″
		3.77, dd (12.0, 5.4)	5′, 1″		4.38, dd, (12.0, 6.0)	5′, 1″		4.36, dd, (12.0, 6.0)	5′, 1″		3.82, dd (12.0, 5.4)	4′, 5′, 1″
1″	105.2, CH	4.38, d (7.8)	6′, 2″, 3″	169.1, qC			169.1, qC			105.2, CH	4.42, d (7.8)	6′, 2″, 3″
2″	75.2, CH	3.21, m	1″, 3″	115.9, CH	6.42, d (15.9)	1″, 4″	115.8, CH	6.66, d (15.9)	1″, 4″	75.2, CH	3.23, m	1″, 3″, 4″
3″	78.1, CH	3.50, m	2″, 4″	147.4, CH	7.62, d (15.9)	1″, 2″, 4″, 5″, 9″	147.4, CH	7.62, d (15.9)	1″, 2″, 4″, 5″, 9″	78.2, CH	3.39, m	2″, 4″
4″	71.8, CH	3.28, m	3″, 5″	126.7, qC			126.7, qC			71.8, CH	3.28, m	2″, 5″, 6″
5″	78.2, CH	3.26, m	3″, 4″	107.1, CH	6.92, s	3″, 4″, 6″, 7″, 9″	107.2, CH	6.94, s	3″, 4″, 6″, 7″, 9″	78.2, CH	3.36, m	4″, 6″
6″	62.9, CH_2	3.86, dd (12.0, 2.1)	4″	148.7, qC			149.6, qC			62.9, CH_2	3.88, dd (12.0, 2.1)	4″
		3.67, dd (12.0, 5.4)	4″, 5″								3.67, dd (12.0, 5.4)	4″, 5″
7″				139.9, qC			139.9, qC					
8″				148.7, qC			149.6, qC					
9″				107.1, CH	6.92, s	3″, 4″, 5″, 7″, 8″	107.2, CH	6.94, s	3″, 4″, 5″, 7″, 8″			
6″-OCH_3				57.0, CH_3	3.88, s	6″	57.0, CH_3	3.88, s	6″			
8″-OCH_3				57.0, CH_3	3.88, s	8″	57.0, CH_3	3.88, s	8″			



6

7

Figure 1. Selected HMBC (→) correlations of compounds **6** and **7**.

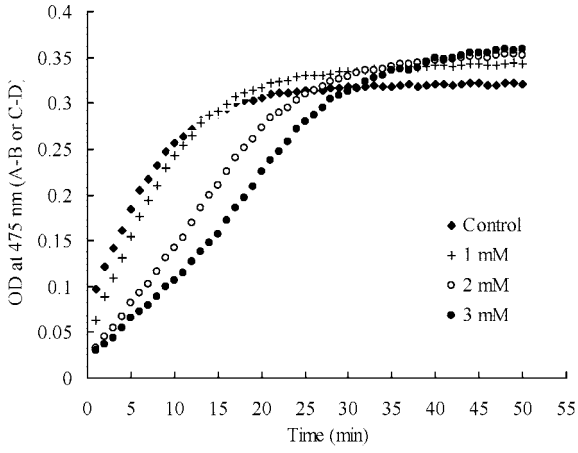


Figure 2. Inhibitory effect of jasminodiol (**1**) on tyrosinase in a dose-dependent manner. Mushroom tyrosinase and L-Dopa were incubated in the absence (control) and presence of **1** (1, 2, and 3 mM).

(^{1}H NMR, 300 MHz; ^{13}C NMR, 75 MHz) using CD_3OD as the solvent and tetramethylsilane (TMS) as an internal standard. Chemical shifts (δ) were expressed in ppm with reference to the TMS signals. Two-dimensional (2D) NMR (HMQC, HMBC) experiments were performed on a Bruker Avance 500 spectrometer. HRMS was measured on a JMS-700 Mstation mass spectrometer. Semipreparative HPLC were conducted on a TRILUTION LC with a UV/vis-151 detector, a 321 pump, a 402 syringe pump, and a GX-271 liquid handler (Gilson, Inc.), using a YMC-pack Pro C_{18} (250 × 20 mm, i.d.) column. Column chromatography was performed using Si (Kieselgel 60, 70−230 mesh and 230−400 mesh, Merck). TLC was performed on Merck precoated silica gel 60 F_{254} and/or RP-18 F_{254s} plates (0.25 mm), and compounds were observed under UV 254 and 365 nm, or visualized by spraying the dried plates with 10% H_2SO_4, followed by heating at 180 °C. Optical density (OD) values in the tyrosinase inhibitory activity assays were read on an Emax Precision microplate reader.

Plant Material. The dried, ripe fruit of *G. jasminoides* was purchased from a pharmacy store in Daejeon, Korea, in July 2006, and the fruit was identified by one of the authors (K.B.). A voucher specimen (CNU 1516-3) was deposited at the herbarium in the College of Pharmacy, Chungnam National University.

Extraction and Isolation. A dried fruit slice of *G. jasminoides* (4.5 kg) was extracted with hot MeOH (5 L × 3 times) for 2 days. The MeOH extracts were filtered, combined, and concentrated *in vacuo*, resulting in a residue (1.0 kg). The residue was suspended into H_2O and then fractionated successively with *n*-hexane, EtOAc, and *n*-BuOH, producing an *n*-hexane-soluble fraction (120.0 g), EtOAc-soluble fraction (80.0 g), and *n*-BuOH-soluble fraction (700.0 g), respectively. The *n*-BuOH-soluble fraction was dissolved in a minimum amount of 50% MeOH and then applied to a Diaion HP 20 column (100 × 20 cm, i.d.) with H_2O and MeOH gradient eluent to give fractions GjB1−5. GjB1 was subjected to an ODS column (50 × 2.5 cm, i.d.), using MeOH−H_2O (1:2, v/v) as eluent. The third subfraction from this column was applied to an RP HPLC [Gilson TRILUTION LC system; YMC-pack Pro C18 (250 × 20 mm, i.d.) column; MeOH−H_2O (2:8, v/v); UV detection, 254 nm; flow rate 6 mL/min], producing **1** (25.9 mg), **2** (145.0 mg), **3** (22.0 mg), **4** (21.0 mg), **5** (16.5 mg), **6** (182.0 mg), and **9** (6.0 mg). The EtOAc-soluble fraction (70.0 g) was chromatographed on a Si gel column (200−300 mesh, 3.6 × 50 cm) and eluted with a gradient of acetone in $CHCl_3$, yielding a crude fraction containing **7** and **8**. This was further purified by chromatography on a Sephadex LH-20 column (2.5 × 50 cm) with CH_2Cl_2−MeOH (1:1, v/v) and then by HPLC over a YMC-pack Pro C18 (250 × 10 mm, i.d.) column using MeOH−H_2O (6:4, v/v) as the mobile phase, followed by UV 240 nm for detection, to give **7** (7.5 mg) and **8** (7.0 mg).

Jasminodiol (1): colorless oil; $[\alpha]^{22}_D$ −54.5 (*c* 0.22, MeOH); UV (MeOH) λ_{max} (log ε) 248 (3.91) nm; CD (MeOH) 211.0 ($\Delta\varepsilon$ −1.39), 249.0 ($\Delta\varepsilon$ −1.17), 328.0 ($\Delta\varepsilon$ −0.35) nm; IR (KBr) ν_{max} 3390, 2900, 1650, 1045 cm^{-1}; ^{1}H NMR (300 MHz, CD_3OD) and ^{13}C NMR (75 MHz, CD_3OD), see Table 1; HREIMS *m/z* 184.1098 $[M]^+$ (calcd for $C_{10}H_{16}O_3$, 184.1099).

Jasminoside H (6): white, amorphous powder; $[\alpha]^{22}_D$ −70.0 (*c* 0.22, MeOH); UV (MeOH) λ_{max} (log ε) 246 (4.06) nm; CD (MeOH) 205.0 ($\Delta\varepsilon$ −2.21), 240.0 ($\Delta\varepsilon$ −1.58), 329.0 ($\Delta\varepsilon$ −0.25) nm; IR (KBr) ν_{max} 3400, 2925, 1680, 1070 cm^{-1}; ^{1}H NMR (300 MHz, CD_3OD) and ^{13}C NMR (75 MHz, CD_3OD), see Table 2; HRFABMS *m/z* 493.2253 $[M + H]^+$ (calcd for $C_{22}H_{37}O_{12}$, 493.2285).

6′-*O*-Sinapoyljasminoside A (7): white, amorphous powder; $[\alpha]^{22}_D$ +50.0 (*c* 0.40, MeOH); UV (MeOH) λ_{max} (log ε) 216 (4.45), 244 (4.42), 337 (4.14) nm; CD (MeOH) 200.0 ($\Delta\varepsilon$ +1.51), 253.0 ($\Delta\varepsilon$ +0.65), 318.0 ($\Delta\varepsilon$ +0.40), 339.0 ($\Delta\varepsilon$ +0.38) nm; IR (KBr) ν_{max} 3360, 2920, 1710, 1680, 1615, 1608, 1520, 1440, 1110 cm^{-1}; ^{1}H NMR (300 MHz, CD_3OD) and ^{13}C NMR (75 MHz, CD_3OD), see Table 2; HRFABMS *m/z* 537.2275 $[M + H]^+$ (calcd for $C_{27}H_{37}O_{11}$, 537.2336).

6′-*O*-Sinapoyljasminoside C (8): white, amorphous powder; $[\alpha]^{22}_D$ −12.5 (*c* 0.24, MeOH); UV (MeOH) λ_{max} (log ε) 213 (4.50), 235 (4.37), 284 (4.15), 339 (4.18) nm; IR (KBr) ν_{max} 3360, 2920, 1700, 1640, 1440, 1300, 1080 cm^{-1}; ^{1}H NMR (300 MHz, CD_3OD) and ^{13}C NMR (75 MHz, CD_3OD), see Table 2; HRFABMS *m/z* 535.2184 $[M + H]^+$ (calcd for $C_{27}H_{35}O_{11}$, 535.2179).

Jasminoside I (9): white, amorphous powder; $[\alpha]^{22}_D$ −7.5 (*c* 0.40, MeOH); UV (MeOH) λ_{max} (log ε) 254 (4.07) nm; IR (KBr) ν_{max} 3400, 2900, 1670, 1440, 1070 cm^{-1}; ^{1}H NMR (300 MHz, CD_3OD) and ^{13}C NMR (75 MHz, CD_3OD), see Table 2; HRESIMS *m/z* 493.2214 $[M + H]^+$ (calcd for $C_{22}H_{37}O_{12}$, 493.2285).

Enzymatic Hydrolysis of 6. Naringinase (100.0 mg) was added to a suspension of **6** (50.0 mg) in 50 mM acetate buffer (pH, 5.5), and the mixture was stirred at 37 °C for 3 h. The reaction mixture was extracted with EtOAc (20 mL × 3 times) and evaporated to dryness. The residue was dissolved in MeOH and applied to RP HPLC [Gilson trilution system; YMC-pack Pro C18 (250 × 20 mm, i.d.) column; MeOH−H_2O (2:8, v/v); UV detection, 240 nm], yielding **3** (5.0 mg) and **4** (4.5 mg), which were identified by comparison of the spectroscopic data with literature values. The H_2O layer was concentrated and passed through a Sep-Pak C_{18} cartridge (Waters, Milford, MA). The sugar was identified as D-glucose by TLC in EtOAc−MeOH−AcOH (13:4:3:3) with an authentic D-(+)-glucose (Sigma) (R_f 0.35), in conjunction with the following GC analysis. The remaining eluate was concentrated to dryness, and the residue was stirred with D-cysteine methyl ester hydrochloride, hexamethyldisilazane, and trimethylsilyl chloride in pyridine using the same procedures as in previous reports.[12] After the reactions, the supernatant was analyzed by GC [column: GL capillary column TC-1 (GL Science, Inc.) 0.25 mm × 30 m, detector, FID; detector temp, 270 °C; injector temp, 270 °C; carrier gas, N_2; column temperature, 230 °C]. A peak corresponding to D-glucose appeared at a t_R of 21.6 min.

Tyrosinase Inhibitory Activity Assay. The mushroom tyrosinase and L-Dopa used for the bioassay were purchased from Sigma Chemical Co. Antityrosinase activity was measured by spectrophotometry, according to the method of Mason and Peterson with minor modifications.[20] The test substance was dissolved in 0.1 mL of 10% DMSO in aqueous solution and incubated with 0.1 mL of 135 U/mL mushroom tyrosinase in phosphate buffer solution (PBS, pH 6.8) at 25 °C for 10 min, and then 0.1 mL of L-Dopa (0.5 mM, PBS pH 6.8) was added. The reaction mixture was incubated for 5 min. The amount of dopachrome in the mixture was determined by the optical density (OD) at 475 nm using an Emax Precision microplate reader. Kojic acid and hydroquinone (Sigma Chemical Co.) were used as positive control

agents. The inhibitory percentage of tyrosinase was calculated as follows: % inhibition = $\{[(A - B) - (C - D)]/(A - B)\} \times 100$ (A, OD at 475 nm without test substance; B, OD at 475 nm without test substance and tyrosinase; C, OD at 475 nm with test substance; D, OD at 475 nm with test substance, but without tyrosinase).

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