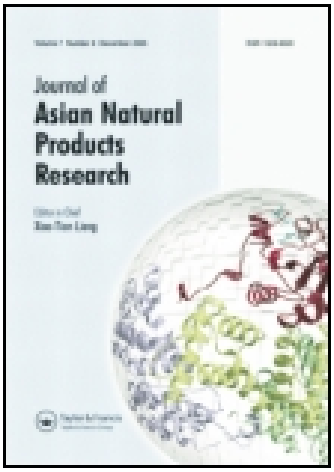

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Aromatic glycosides from the flower buds of Lonicera japonica

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Aromatic glycosides from the flower buds of *Lonicera japonica*

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Six new glycosides (**1**–**6**) have been isolated from the flower buds of *Lonicera japonica*. Their structures including the absolute configurations were determined by spectroscopic and chemical methods as (−)-2-hydroxy-5-methoxybenzoic acid 2-*O*-β-D-(6-*O*-benzoyl)-glucopyranoside (**1**), (−)-4-hydroxy-3,5-dimethoxybenzoic acid 4-*O*-β-D-(6-*O*-benzoyl)-glucopyranoside (**2**), (−)-(*E*)-3,5-dimethoxyphenylpropenoic acid 4-*O*-β-D-(6-*O*-benzoyl)-glucopyranoside (**3**), (−)-(7*S*,8*R*)-(4-hydroxyphenylglycerol 9-*O*-β-D-[6-*O*-(*E*)-4-hydroxy-3,5-dimethoxyphenylpropenoyl]-glucopyranoside (**4**), (−)-(7*S*,8*R*)-(4-hydroxy-3-methoxyphenylglycerol 9-*O*-β-D-[6-*O*-(*E*)-4-hydroxy-3,5-dimethoxyphenylpropenoyl]-glucopyranoside (**5**), and (−)-4-hydroxy-3-methoxyphenol β-D-{6-*O*-[4-*O*-(7*S*,8*R*)-(4-hydroxy-3-methoxyphenylglycerol-8-yl)-3-methoxybenzoyl]}-glucopyranoside (**6**), respectively.

Keywords: flower buds; *Lonicera japonica*; Caprifoliaceae; aromatic glycosides

1. Introduction

Lonicera japonica Thunb. (Caprifoliaceae) is widely cultivated in the Shandong and Henan provinces of China. Its flower buds and stems are used in traditional Chinese medicine for the treatment of various diseases, and are among the most common ingredients of formulations used for treating influenza, cold, fever, and infections [1–5]. Chemical and pharmacological studies have resulted in the characterization of constituents with different structural features and biological activities from the flower buds of *L. japonica*, including caffeoyl quinic acids, secoiridoids, flavonoids, saponins, cerebrosides, polyphenols, and nitrogen-containing iridoids [6–13]. As part of a program to assess the chemical and biological diversity of traditional Chinese medicines, 14 pyridinium alkaloid-coupled (lonijaposides A–N) and 4 phenylpyruvic acid-derived moieties-coupled (loniphenyruviridosides A–D) homosecoiridoids, together with 7 known iridoid derivatives, were reported from the water extract of the flower buds of *L. japonica* [14,15]. Focusing our investigation on the components of this plant material, after the flower buds were extracted by water, the residue was further extracted with EtOH (95%). From the EtOH extract, six new glycosides (**1**–**6**) have been characterized (Figure 1). The isolation and structure determination of the new isolates have been reported in this study.

2. Results and discussion

Compound **1** was obtained as a white amorphous solid with $[\alpha]_D^{20}$ −51.5 (*c* 0.18, MeOH). The IR spectrum of **1** showed absorption bands for hydroxyl (3307 cm^{-1}), carbonyl (1708 cm^{-1}), and

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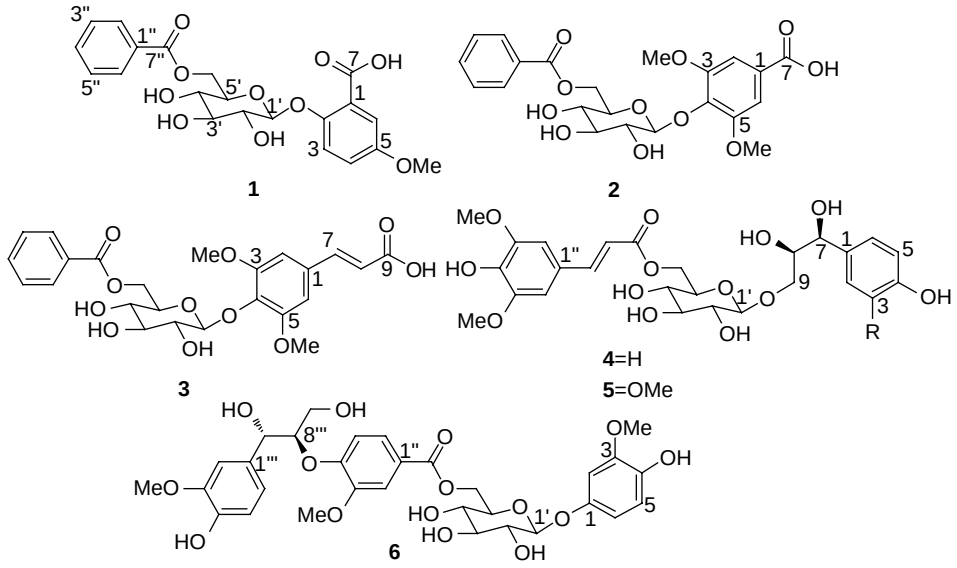

Figure 1. The structures of compounds **1**–**6**.

aromatic (1603 and 1516 cm^{-1}) functional groups. The positive mode electrospray ionization mass spectrometry (ESIMS) of **1** gave a quasi-molecular ion peak at *m/z* 457 $[M + Na]^+$. The molecular formula $C_{21}H_{22}O_{10}$ was indicated by high resolution electrospray ionization mass spectrometry (HRESIMS) at *m/z* 457.1106 $[M + Na]^+$. The ^1H NMR spectrum of **1** in CD_3OD showed signals attributed to a *meta-ortho*-trisubstituted phenyl group at δ_H 7.07 (1H, d, $J = 8.4$ Hz, H-3), 7.40 (1H, d, $J = 8.4$ Hz, H-4), and 7.55 (1H, s, H-6); a benzoyl unit at δ_H 7.94 (2H, d, $J = 7.2$ Hz, H-2″/6″), 7.43 (2H, t, $J = 7.2$ Hz, H-3″/5″), and 7.55 (1H, t, $J = 7.2$ Hz, H-4"); and an aromatic methoxy group at δ 3.82 (s). In addition, it showed signals assignable to a β-glucopyranosyl unit, of which the anomeric proton was resonated at δ_H 4.98 (d, $J = 7.2$ Hz, H-1′), whereas the oxymethylene protons were at δ_H 4.66 (dd, $J = 12.0$ and 1.8 Hz, H-6′a) and 4.32 (dd, $J = 12.0$ and 7.8 Hz, H-6′b). The presence of a *β*-glucopyranosyl unit was confirmed by acid hydrolysis of **1** that produced glucose identified by a thin-layer liquid chromatography (TLC) comparison with an authentic sugar sample. The glucose isolated from the hydrolysate gave a positive optical rotation, $[\alpha]_D^{20} + 40.7$ (*c* 0.10, H_2O), indicating that it was the β-D-glucose [15]. The ^{13}C NMR and DEPT spectra of **1** showed carbon signals (Table 1) corresponding to the above units, including two oxygen-bearing aromatic carbons [δ_C 150.2 (C-5) and 151.0 (C-2)] and a carbonyl carbon [δ_C 167.8 (C-7″)]. These spectroscopic data suggested that **1** was a *meta-ortho*-substituted benzolic acid β-glucopyranoside with the benzoyl unit esterified at C-6′. The suggestion was supported by comparing of the NMR data of **1** with those of the co-occurring vanillic acid 4-*O*-β-D-(6-*O*-benzoyl)-glucopyranoside [16], and further confirmed by 2D NMR data analysis. The HMQC and $^1H-^1H$ correlation spectroscopy (COSY) spectra of **1** provided unambiguous assignments of proton and carbon signals in the NMR spectra. In the HMBC spectrum, correlations from H_2-6′ and H-2″/H-6″ to C-7″ confirmed that the benzoyl unit was located at C-6′ of the β-glucopyranosyl moiety. In addition, correlations of C-2 with H-4 and H-6 and C-5 with H-3 and

Table 1. ^{1}H and ^{13}C NMR spectral data (δ) for compounds **1**–**6**.[a]

No.	**1**		**2**		**3**		**4**		**5**		**6**[b]	
	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C
1		ND		132.5		132.7		133.6		134.8		152.5
2		151.0	7.23 s	108.6	6.74 s	104.9	7.10 d (8.0)	129.2	6.92 d (1.5)	112.1	6.63 s	104.1
3	7.07 d (8.4)	116.5		154.3		152.9	6.71 d (8.0)	116.0		149.0		149.2
4	7.40 d (8.4)	124.4		138.6		134.0		158.1		147.3		143.1
5		150.2		154.3		152.9	6.71 d (8.0)	116.0	6.65 d (8.5)	116.0	6.47 d (8.0)	115.9
6	7.55 s	114.6	7.23 s	108.6	6.74 s	104.9	7.10 d (8.0)	129.2	6.73 dd (8.5, 1.5)	121.3	6.48 d (8.0)	110.2
7		ND		173.5	7.01 d (16.0)	135.4	4.53 d (6.6)	75.3	4.50 d (6.0)	75.7		
8					6.32 d (16.0)	129.8	3.85 m	76.2	3.85 m	75.8		
9a						170.4	3.78 dd (10.8, 3.0)	72.7	3.94 dd (10.5, 2.5)	73.1		
9b							3.35 d (10.8)		3.58 dd (10.5, 7.5)			
OCH_3-3			3.72 s	57.1	3.68 s	56.2			3.79 s	57.1	3.61 s	56.3
OCH_3-5	3.82 s	56.6	3.72 s	57.1	3.68 s	56.2						
1′	4.98 d (7.2)	101.9	4.96 d (7.5)	104.7	4.92 d (7.0)	102.5	4.23 d (7.8)	105.0	4.27 d (8.0)	105.4	4.70 d (7.5)	103.7
2′	3.52 dd (9.0, 7.2)	74.8	3.50 dd (8.5,7.5)	76.0	3.23 dd (7.5,9.0)	74.0	3.24 dd (8.0, 7.8)	75.1	3.21 dd (8.5, 8.0)	75.5	3.38 dd (8.5, 7.5)	75.0
3′	3.48 t (9.0)	77.8	3.43 t (8.5)	78.2	3.32 t (9.0)	76.5	3.37 t (8.0)	77.7	3.35 t (8.5)	78.0	3.42 t (8.5)	78.0
4′	3.39 dd (9.6, 9.0)	72.0	3.39 t (9.0,8.5)	72.5	3.32 t (9.0)	70.4	3.37 t (8.0)	71.6	3.33 t (8.5)	72.0	3.34 t (8.5)	72.1
5′	3.77 m	75.6	3.45 m	76.0	3.38 m	74.2	3.49 m	75.5	3.48 m	75.7	3.66 m	75.6
6′a	4.66 dd (12.0, 1.8)	65.4	4.55 d (12.0)	65.5		64.2	4.47 dd (12.0, 2.4)	64.5	4.46 dd (12.0, 2.0)	65.0	4.64 d (11.0)	65.3
6′b	4.32 dd (12.0, 7.8)		4.35 dd (12.0,7.0)				4.30 dd (12.0, 6.0)		4.25 dd (12.0, 6.0)		4.34 dd (11.0, 6.0)	
1″		131.2		131.6		132.7		126.5		126.9		124.2
2″	7.94 d (7.2)	130.6	7.75 d (7.5)	130.8	7.75 d (7.5)	129.1	6.92 s	107.0	6.84 s	107.2	7.49 s	114.2
3″	7.43 t (7.2)	129.6	7.37 t (7.5)	129.9	7.46 t (7.5)	128.7		149.5		149.8		151.0
4″	7.55 t (7.2)	134.4	7.50 t (7.5)	134.6	7.61 t (7.5)	133.3		140.4		140.0		154.4

5″	7.43 t (7.2)	129.6	7.37 t (7.5)	129.9	7.46 t (7.5)	128.7		149.5		149.8	7.04 d (8.5)	116.1
6″	7.94 d (7.2)	130.6	7.75 d (7.5)	130.8	7.75 d (7.5)	129.1	6.92 s	107.0	6.84 s	107.2	7.50 d (8.5)	124.8
7″		167.8		168.1		165.6	7.62 d (16.2)	147.4	7.56 d (16.0)	147.6		167.6
8″							6.41 d (16.2)	115.5	6.35 d (16.0)	115.9		
9″								169.0		169.3		
OCH_3-3″							3.87 s	56.9	3.80 s	56.6	3.81 s	56.6
OCH_3-5″							3.87 s	56.9	3.80 s	56.6		

[a] ^{1}H NMR data (δ) were measured in MeOH-d_4 for **1**, **2**, and **4–6** and in DMSO-d_6 for **3**, at 600 MHz for ^{1}H and 150 MHz for ^{13}C (**1**, **4**, and **6**) and at 500 MHz for ^{1}H and 125 MHz for ^{13}C (**2**, **3**, and **5**). Proton-coupling constants (J) in Hz are given in parentheses. ND means that the signal was not detected. The assignments were based on ^{1}H–^{1}H COSY, HSQC, and HMBC experiments.

[b] Data for the 4‴-hydroxy-3‴-methoxyphenylglycerol moiety of **6**: δ_H 7.00 (1H, s, H-2‴), 6.82 (1H, brd, J = 8.0 Hz, H-6‴), 6.70 (1H, d, J = 8.0 Hz, H-5‴), 4.85 (1H, d, J = 5.0 Hz, H-7‴), 4.52 (1H, m, H-8‴), 3.73 (1H, dd, J = 12.0 and 4.0 Hz, H-9‴a), 3.50 (1H, dd, J = 12.0 and 6.0 Hz, H-9‴b), and 3.77 (3H, s, OCH_3-3‴); δ_C 133.7 (C-1‴), 111.7 (C-2‴), 148.9 (C-3‴), 147.2 (C-4‴), 115.9 (C-5‴), 120.7 (C-6‴), 74.0 (C-7‴), 85.8 (C-8‴), 62.1 (C-9‴), and 56.4 (OCH$_3$-3‴).

OCH_3, together with their shifts and the molecular composition, demonstrated that the *meta-ortho*-substituted benzolic acid moiety was 2-oxy-5-methoxybenzolic acid though the signals of C-1 and C-7 were not displayed in the ^{13}C NMR spectrum of **1**. Meanwhile, a HMBC correlation of C-2 with H-1′ revealed that the β-glucopyranosyl moiety was located at C-2 in **1**. Therefore, the structure of compound **1** was determined as (−)-2-hydroxy-5-methoxybenzoic acid 2-*O*-β-D-(6-*O*-benzoyl)-glucopyranoside.

Compound **2** was obtained as a white amorphous powder with $[\alpha]_D^{20}$ −31.8 (*c* 0.09, MeOH). The molecular formula $C_{22}H_{24}O_{11}$ was indicated from HRESIMS at *m/z* 487.1212 $[M + Na]^+$, combined with the NMR spectral data. The UV, IR, and NMR spectroscopic features of **2** (see Section 3 and Table 1) were similar to those of **1**. Comparison of the NMR spectral data of **1** and **2** indicated that the 2-oxy-5-methoxybenzolic acid moiety in **1** was replaced by a 4-oxy-3,5-dimethoxybenzoic acid unit in **2**. This demonstrated that **2** was 4-hydroxy-3,5-dimethoxybenzoic acid 4-*O*-β-D-(6-*O*-benzoyl)-glucopyranoside, which was confirmed by 2D NMR data analysis and acid hydrolysis. Particularly, the HMBC spectrum showed correlations from H-1′ and H-2/H-6 to C-4, indicating the β-D-glucopyranosyl moiety at C-4 in **2**. Therefore, the structure of compound **2** was assigned as (−)-4-hydroxy-3,5-dimethoxybenzoic acid 4-*O*-β-D-(6-*O*-benzoyl)-glucopyranoside.

Compound **3**, a white amorphous powder with $[\alpha]_D^{20}$ −10.1 (*c* 0.37, MeOH), has the molecular formula $C_{24}H_{26}O_{11}$ as indicated by HRESIMS at *m/z* 513.1374 $[M + Na]^+$ and the NMR spectral data (Table 1). Comparison of the NMR spectral data of **3** and **2** indicated the substitution of the 4-oxy-3,5-dimethoxybenzoic acid unit in **2** by a *trans*-4-oxy-3,5-dimethoxyphenylpropenoic acid moiety in **3**. This was confirmed by correlations from H-2/H-6 to C-1, C-3/C-5, C-4, and C-7; from H-7 to C-1, C-2/C-6, and C-9; and from H-8 to C-1 and C-9, in the HMBC spectrum of **3**, in combination with the shifts of the proton and carbon resonances. In addition, the HMBC correlations from H-1′ to C-4 and from H_2-6′ to C-7″ proved the linkage of the three moieties in **3**. The D-configuration of the β-glucopyranosyl moiety was also confirmed by the acid hydrolysis of **3** using the same protocol as described earlier. Thus, the structure of compound **3** was designated as (*E*)-3,5-dimethoxyphenylpropenoic acid (−)-4-*O*-β-D-(6-*O*-benzoyl)-glucopyranoside.

Compound **4** was obtained as a yellowish amorphous powder with $[\alpha]_D^{20}$ −10.5 (*c* 0.20, CH_3CN), and has the molecular formula $C_{26}H_{32}O_{13}$ as indicated by HRESIMS at *m/z* 575.1745 $[M + Na]^+$ and the NMR spectral data (Table 1). The NMR spectra of **4** showed the presence of (*E*)-4″-hydroxy-3″,5″-dimethoxyphenylpropenoyl, 4-hydroxyphenyl, and β-glucopyranosyl moieties in the molecule. In addition, it demonstrated the presence of two oxygen-bearing methines (CH-7 and CH-8) and an oxygen-bearing methylene (CH_2-9) in **4**. The ^{1}H−^{1}H gCOSY spectrum of **4** displayed vicinal coupling correlations of H-7/H-8/H_2-9, whereas the HMBC spectrum showed correlations from H-7 to C-1 and C-2/C-6 of the 4-hydroxyphenyl moiety. These 2D NMR data revealed that there was a 4-hydroxyphenylglycerol moiety. In addition, the HMBC spectrum exhibited correlations from H_2-9 to C-1′ and from H-1′ to C-9, indicating that the β-glucopyranosyl was located at C-9. Moreover, HMBC correlations of C-9″ with H_2-6′, H-7″, and H-8″ and of C-3″/C-5″ with OCH_3-3″/OCH_3-5″, together with their shifts, demonstrated that the (*E*)-4″-hydroxy-3″,5″-dimethoxyphenylpropenoyloxy moiety was substituted at C-6′ in **4**. The $\Delta\delta_{C8-C7}$ value in DMSO was smaller than 0.5 ppm, suggesting a 7,8-*erythro* configuration for **4** [17–19]. In the CD spectrum of **4**, a negative Cotton

effect at 227 nm indicated the 8*R* configuration [19]. The D-configuration of the β-glucopyranosyl moiety in **4** was proved by acid hydrolysis using the aforementioned method. Therefore, the structure of compound **4** was determined as (−)-(7*S*,8*R*)-(4-hydroxyphenylglycerol 9-*O*-β-D-[6-*O*-(*E*)-4-hydroxy-3,5-dimethoxyphenylpropenoyl]-glucopyranoside.

Compound **5**, a yellowish amorphous powder with $[\alpha]_D^{20}$ −25.1 (*c* 0.12, MeOH), has the molecular formula $C_{27}H_{34}O_{14}$ as indicated by HRESIMS at *m/z* 605.1854 $[M + Na]^+$ and the NMR spectral data (Table 1). Comparison of the NMR spectral data of **4** and **5** indicated the replacement of the 4-hydroxyphenylglycerol moiety in **4** by a 4-hydroxy-3-methoxyphenylglycerol moiety in **5**. This was confirmed by 2D NMR data analysis of **5**. Particularly, $^1H-^1H$ gCOSY correlations of H-7/H-8/H_2-9 and HMBC correlations from H-7 to C-1, C-2, and C-6 and from O*C*H_3-3 to C-3, together with their shifts, proved the presence of 4-hydroxy-3-methoxyphenylglycerol moiety. HMBC correlations from H_2-9 to C-1′, from H-1′ to C-9, and from H_2-6′ to C-9″ confirmed the linkage of the moieties in **5**. The $\Delta\delta_{C8-C7}$ value in DMSO was similar to that of **4**, indicating the 7,8-*erythro* configuration for **5** [17–19]. The 8*R* configuration was supported by the negative Cotton effect at 230 nm in the CD spectrum [19], and the D-configuration of the β-glucopyranosyl moiety in **5** was confirmed by acid hydrolysis using the method as described earlier. Thus, the structure of compound **5** was assigned as (−)-(7*S*,8*R*)-(4-hydroxy-3-methoxyphenylglycerol 9-*O*-β-D-[6-*O*-(*E*)-4-hydroxy-3,5-dimethoxyphenylpropenoyl]-glucopyranoside.

Compound **6** was obtained as a white amorphous powder. The molecular formula $C_{31}H_{36}O_{15}$ was indicated by HRESIMS at *m/z* 671.1950 and the NMR spectral data. The NMR spectra of **6** (Table 1) showed the presence of three *meta-ortho*-trisubstituted phenyl groups and a β-glucopyranosyl moiety, as well as two oxygen-bearing methines (CH-7‴ and CH-8‴), an oxygen-bearing methylene (CH_2-9‴), and an ester carbonyl carbon (δ_C 167.6, C-7″). In the HMBC spectrum of **6**, correlations of H-2/C-1, C-3, C-4, and C-6; H-5 and/or H-6/C-1, C-3, and C-4; and O*C*H_3-3/C-3; together with their shifts, suggested the presence of a 4-hydroxy-3-methoxyphenoxy unit, which was supported by a correlation between H-2 and O*C*H_3-3 in the NOESY spectrum. The HMBC spectrum also showed correlations of H-2″/C-1″, C-3″, C-4″, C-6″, and C-7″; H-5″/C-1″, C-3″, C-4″, and C-6″; H-6″/C-1″, C-2″, C-4″, C-5″, and C-7″; and O*C*H_3-3″/C-3″. The correlations in combination with shifts of these proton and carbon resonances indicated that there was a 4″-hydroxy-3″-methoxybenzoyloxy moiety. In addition, correlations of H-7‴/C-1‴, C-2‴, C-6‴, C-8‴ and C-9‴; H-2‴/C-1‴, C-3‴, C-4‴, C-6‴, and C-7‴; H-5‴/C-1‴, C-3‴, C-4‴, and C-6‴; H-6‴/C-1‴, C-2‴, C-4‴, C-5‴, and C-7‴; and O*C*H_3-3‴/C-3‴, together with their shifts, demonstrated the presence of a 4‴-hydroxy-3‴-methoxyphenylglycerol moiety. This was supported by $^1H-^1H$ gCOSY correlations of H-7‴/H-8‴/H_2-9‴. Furthermore, HMBC correlations of H-1′/C-1 and H_2-6′/C-7″ indicated that the 4-hydroxy-3-methoxyphenoxy and 4″-hydroxy-3″-methoxybenzoyloxy units were connected at C-1′ and C-6′ of the β-glucopyranosyl moiety, respectively, whereas a linkage between C-8‴ and C-4″ was established by an HMBC correlation of H-8‴/C-4″. The $\Delta\delta_{C8'''-C7'''}$ value (11.8 ppm) suggested an 7‴,8‴-*erythro* configuration for **6** [17–19]. In the CD spectrum of **6**, a negative Cotton effect at 228 nm indicated the 8*R* configuration [19]. Acid hydrolysis of **6** produced glucose identified by TLC comparison with an authentic sugar sample. The glucose isolated from the hydrolysate gave a positive optical rotation, $[\alpha]_D^{20}$ +42.3 (*c* 0.08, H_2O), indicating that it was the β-D-glucose [7]. Therefore, compound **6** was

determined as (−)-4-hydroxy-3-methoxyphenol β-D-{6-*O*-[4-*O*-(7*S*,8*R*)-(4-hydroxy-3-methoxyphenylglycerol-8-yl)-3-methoxybenzoyl]}-glucopyranoside.

3. Experimental

3.1 General experimental procedures

Optical rotations were measured on P-2000 polarimeter (JASCO, Tokyo, Japan). UV spectra were measured on a JASCOP-650 spectrometer (JASCO). CD spectra were recorded on a JASCO J-815 CD spectrometer (JASCO). IR spectra were recorded on a Nicolet 5700 FT-IR microscope instrument (FT-IR microscope transmission; Thermo Electron Corporation, Madison, WI, USA). NMR spectra were obtained at 500 MHz for ^{1}H and 125 MHz for ^{13}C, respectively, on an Inova 500 spectrometer (Varian Associates Inc., Palo Alto, CA, USA) in MeOH-d_4 with solvent peaks used as references. ESIMS and HRESIMS data were measured using an AccuToFCS JMS-T100CS spectrometer (Agilent Technologies Ltd. Santa Clara, CA, USA). Column chromatography (CC) was performed with silica gel (200–300 mesh; Qingdao Marine Chemical Inc. Qingdao, China) and Sephadex LH-20 (Pharmacia Biotech AB, Uppsala, Sweden). HPLC separation was performed on an instrument consisting of a Waters 600 controller, a Waters 600 pump, and a Waters 2487 dual λ absorbance detector (Waters Corporation, Milford, PA, USA), with an Prevail (250 × 10 mm i.d.) column packed with C_{18} (5 μm) (Alltech Associates Inc., Deerfield, IL, USA). TLC was carried out in glass precoated silica gel GF_{254} plates (Qingdao Marine Chemical Inc.). Spots were visualized under UV light or by spraying with 7% H_2SO_4 in 95% EtOH followed by heating. Unless otherwise noted, all chemicals were obtained from commercially available sources, and were used without further purification.

3.2 Plant material

The flower buds of *Lonicera japonica* were collected in May 2005 from Shangqiu, Henan Province, China. Plant identity was verified by Mr Lin Ma (Institute of Materia Medica, Beijing, China). A voucher specimen (No. ZH02273) was deposited at the herbarium of the Department of Chemistry of Natural Products, Institute of Materia Medica.

3.3 Extraction and isolation

The air-dried flower buds of *L. japonica* (40 kg) were extracted with H_2O (120 liters, 3 × 24 h) at room temperature, then the flower buds' residue was extracted by sonication with 95% EtOH (40 liters, 3 × 1 h). The ethanol extract was evaporated under reduced pressure to yield a dark brown residue (1536 g), which was suspended in H_2O (5 liters) and partitioned with EtOAc (6 × 5 liters). After removing the solvent, the EtOAc fraction (815 g) was chromatographed over silica gel, eluting with increasing amounts of acetone (0–100%) in petroleum ether, to afford 10 fractions (A–J) based on TLC analysis. Fraction I (90 kg) was separated by CC over MCI gel CHP 20P, successively eluted by H_2O (10 liters), 50% EtOH (20 liters), 90% EtOH (10 liters), and Me_2CO (8 liters), to give corresponding fractions I1 − I4. Fraction I1 (19 g) was separated via reverse phase medium pressure liquid chromatography (RP-MPLC) eluting with a gradient of MeOH (5–100%) in H_2O to give fractions I1-1 to I1-6. Fraction I1-4 (7.52 g) was chromatographed over Sephadex LH-20 eluting with chloroform/methanol (1:1) to yield fractions I1-4-1 to I1-4-4. Separation of fraction I1-4-3 (3.6 g) by a normal-phase silica gel column ($CHCl_3$/-MeOH, 40:1) yielded fractions I1-4-3-1 to I1-4-3-3. Fraction I1-4-3-2 (286 mg) was separated by RP flash CC (20–80% MeOH in H_2O) to afford fractions I1-4-3-2-1 to I1-4-3-2-3, and purification of fraction I1-4-3-2-1 (63 mg) by RP-HPLC (17% MeCN in

H_2O) gave **2** (4.2 mg). Fraction I1-4-3-2-2 (103 mg) was isolated by preparative TLC (mobile phase: $CHCl_3$/MeOH, 6:1), followed by RP HPLC separation (24% MeCN in H_2O) to yield **3** (5.7 mg) and **1** (3.6 mg). Fraction I1-5 (3.76 g) was chromatographed over Sephadex LH-20 eluting with methanol to yield fraction I1-5-1 to I1-5-4, of which I1-5-2 (1.63 g) was isolated by RP flash CC (20–90% MeOH in H_2O) to afford fractions I1-5-2-1 to I1-5-2-5. Fraction I1-5-2-3 (414 mg) was separated by silica gel CC ($CHCl_3$/MeOH, 20:1) to give fractions I1-5-2-3-1 to I1-5-2-3-7, of which fraction I1-5-2-3-6 (158 mg) was isolated by preparative TLC (mobile phase: $CHCl_3$/ MeOH, 4.5:1), followed by RP-HPLC separation (15% MeCN in H_2O) to yield **4** (3.7 mg), **5** (6.7 mg), and **6** (8.2 mg).

*3.3.1. (−)-2-Hydroxy-5-methoxybenzoic acid 2-O-β-D-(6-O-benzoyl)-glucopyranoside (**1**)*

White amorphous solid; $[\alpha]_D^{20}$ −51.5 (*c* 0.18, MeOH); UV (MeOH) λ_{max} (log ε) 206 (4.21), 232 (3.93), 284 (3.29) nm; IR ν_{max} 3307, 2919, 1708, 1603, 1516, 1452, 1415, 1346, 1315, 1281, 1224, 1185, 1116, 1070, 1027, 972, 914, 883, 822, 762, 710 cm^{-1}; ^1H NMR (CD_3OD, 600 MHz) and ^{13}C NMR (CD_3OD, 150 MHz) spectral data (see Table 1); ESIMS: *m/z* 457 $[M + Na]^+$, 433 $[M - H]^-$; (+)-HRESIMS: *m/z* 457.1106 $[M + Na]^+$ (calcd for $C_{21}H_{22}O_{10}Na$, 457.1105).

*3.3.2. (−)-4-Hydroxy-3,5-dimethoxybenzoic acid 4-O-β-D-(6-O-benzoyl)-glucopyranoside (**2**)*

White amorphous powder; $[\alpha]_D^{20}$ −31.8 (*c* 0.09, MeOH); UV (MeOH) λ_{max} (log ε) 207 (4.37), 230 (sh, 3.94), 255 (3.67) nm; IR ν_{max} 3394, 2938, 1717, 1566, 1502, 1454, 1394, 1319, 1280, 1224, 1189, 1125, 1071, 1026, 968, 920, 877, 790, 740, 717 cm^{-1}; ^1H NMR (CD_3OD, 500 MHz) and ^{13}C NMR (CD_3OD, 125 MHz) spectral data (see Table 1); ESIMS: *m/z* 487 $[M + Na]^+$, 463 $[M - H]^-$. (+)-HRESIMS: *m/z* 487.1212 $[M + Na]^+$ (calcd for $C_{22}H_{24}O_{11}Na$, 487.1211).

*3.3.3. (−)-(E)-3,5-Dimethoxyphenylpropenoic acid 4-O-β-D-(6-O-benzoyl)-glucopyranoside (**3**)*

White amorphous powder; $[\alpha]_D^{20}$ −10.1 (*c* 0.37, MeOH); UV (MeOH) λ_{max} (log ε) 202 (3.95), 227 (4.17), 288 (3.87) nm; IR ν_{max} 3394, 3188, 3010, 2921, 2850, 1719, 1646, 1587, 1503, 1468, 1420, 1393, 1333, 1279, 1246, 1124, 1070, 1026, 820, 719, 634 cm^{-1}; ^1H NMR (DMSO-d_6, 500 MHz) and ^{13}C NMR (DMSO-d_6, 125 MHz) spectral data (see Table 1); ESIMS: *m/z* 513 $[M + Na]^+$, 489 $[M - H]^-$. (+)-HRESIMS: *m/z* 513.1374 $[M + Na]^+$ (calcd for $C_{24}H_{26}O_{11}Na$, 513.1367).

*3.3.4. (−)-(7S,8R)-(4-Hydroxyphenylglycerol 9-O-β-D-[6-O-(E)-4-hydroxy-3,5-dimethoxyphenyl propenoyl]-glucopyranoside (**4**)*

Yellowish amorphous powder; $[\alpha]_D^{20}$ −10.5 (*c* 0.20, MeCN); UV (MeOH) λ_{max} (log ε) 204 (4.43), 225 (4.39), 329 (4.27) nm; CD (MeOH) $\Delta\varepsilon_{227\ nm}$ −3.97, $\Delta_{\varepsilon 252\ nm}$ −1.16, $\Delta\varepsilon_{275\ nm}$ −1.64, $\Delta\varepsilon_{306\ nm}$ −0.38; IR ν_{max} 3318, 2934, 1703, 1632, 1612, 1599, 1515, 1458, 1427, 1386, 1340, 1258, 1230, 1173, 1115, 1049, 1026, 1005, 835, 765 cm^{-1}; ^1H NMR (CD_3OD, 600 MHz) and ^{13}C NMR (CD_3OD, 150 MHz) spectral data (see Table 1); ESIMS: *m/z* 575 $[M + Na]^+$, 551 $[M - H]^-$, 587 $[M + Cl]^-$; HRESIMS: *m/z* 575.1745 $[M + Na]^+$ (calcd for $C_{26}H_{32}O_{13}Na$, 575.1735).

*3.3.5. (−)-(7S,8R)-(4-Hydroxy-3-methoxyphenylglycerol 9-O-β-D-[6-O-(E)-4-hydroxy-3,5-dimethoxy phenylpropenoyl]-glucopyranoside (**5**)*

Yellowish amorphous powder; $[\alpha]_D^{20}$ −25.1 (*c* 0.12, MeOH); UV (MeOH)

λ_{max} (log ε) 205 (4.80), 225 (4.87), 329 (4.79) nm; CD (MeOH) $\Delta\varepsilon_{218\ nm}$ −1.35, $\Delta\varepsilon_{230\ nm}$ −0.48, $\Delta\varepsilon_{277\ nm}$ −0.61, $\Delta\varepsilon_{329\ nm}$ −0.10, $\Delta\varepsilon_{324\ nm}$ −0.26; IR ν_{max} 3405, 2940, 1693, 1631, 1611, 1515, 1458, 1427, 1340, 1286, 1257, 1227, 1176, 1157, 1114, 1044, 835, 623 cm^{-1}; ^1H NMR (CD_3OD, 500 MHz) and ^{13}C NMR (CD_3OD, 125 MHz) spectral data (see Table 1); ESIMS: *m/z* 605 $[M + Na]^+$, 581 $[M - H]^-$, 617 $[M + Cl]^-$; (+)-HRESIMS: *m/z* 605.1854 $[M + Na]^+$ (calcd for $C_{27}H_{34}O_{14}Na$, 605.1841).

*3.3.6. (−)-4-Hydroxy-3-methoxyphenol β-D-{6-O-[4-O-(7S,8R)-(4-hydroxy-3-methoxyphenylglycerol-8-yl)-3-methoxybenzoyl]}-glucopyranoside (**6**)*

White amorphous powder; $[\alpha]_D^{20}$ −2.8 (*c* 0.18, MeOH); UV (MeOH) λ_{max} (log ε) 221 (5.16), 266 (4.89), 285 (4.82) nm; CD (MeOH) $\Delta\varepsilon_{228\ nm}$ −3.65, $\Delta\varepsilon_{251\ nm}$ +1.36; IR ν_{max} 3395, 3188, 3011, 2921, 2849, 1704, 1647, 1601, 1512, 1468, 1420, 1343, 1272, 1120, 1073, 1029, 980, 802, 763, 722, 647 cm^{-1}; ^1H NMR (CD_3OD, 600 MHz) and ^{13}C NMR (CD_3OD, 150 MHz) spectral data (see Table 1); ESIMS: *m/z* 671 $[M + Na]^+$, 647 $[M - H]^-$, 683 $[M + Cl]^-$; HRESIMS: *m/z* 671.1950 $[M + Na]^+$ (calcd for $C_{31}H_{36}O_{15}Na$, 671.1946).

*3.3.7. Acid hydrolysis of **1**–**6***

A solution of each compound (**1**–**6**, 2.0–5.0 mg) was hydrolyzed with 2 N HCl (5.0 ml) at 80°C for 6 h. The reaction mixture was extracted with EtOAc (3 × 5 ml). The H_2O phases of the hydrolysates of **1**–**6** were separately concentrated to dryness and chromatographed on a silica gel column, eluting with CH_3CN–H_2O (8:1), to yield glucose with $[\alpha]_D^{20}$ values ranging from +40.3 to +45.1 (*c* 0.08–0.16, H_2O). The solvent system $CHCl_3$–MeOH–H_2O (8:5:1) was used for TLC identification of glucose (retention factor, 0.31).

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