# Glycosides from the Root of *Iodes cirrhosa*

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Seven new neolignan glycosides (**1**–**7**), two arylglycerol glycosides (**8**, **9**), and 18 known glycosides have been isolated from an ethanolic extract of the root of *Iodes cirrhosa*. Their structures including absolute configurations were determined by spectroscopic and chemical methods. Based on analysis of the NMR data of *threo* and *erythro* 8–4′-oxyneolignans and arylglycerols in different solvents, the validity of $J_{7,8}$ and $\Delta\delta_{C8-C7}$ values to distinguish *threo* and *erythro* derivatives was discussed. In the *in vitro* assays, compound **4** and liriodendrin (**17**) both showed activity against glutamate-induced PC12 cell damage at $10^{-5}$ M.

*Iodes* (Icacinaceae) species are woody climber plants, widely distributed in southeastern Asia, especially in southern China. *Iodes cirrhosa* Turcz. is one of several *Iodes* species used in traditional Chinese medicine. Extracts of the root and stem are reported to improve general blood circulation and are used for treatment of inflammatory and rheumatic diseases.[1] There have been no previous reports concerning the secondary metabolites from this genus. As part of a program to assess the chemical and biological diversity of traditional Chinese medicines,[2,3] an ethanolic extract of the root of *I. cirrhosa* has been investigated. We describe herein isolation and structural elucidation of seven new neolignan glycosides (**1**–**7**), two new arylglycerol glycosides (**8**, **9**), and 18 known glycosides. Compounds **1**–**3** are unusual 8–4′-oxyneolignan glycosides with a glycosyloxy group at C-3′, while **7** is an unusual dihydro[*b*]benzofuran neolignan glycoside with an aromatic ring at C-8′. Since the assignment of some of the *erythro* and *threo* 8–4′-oxyneolignan and arylglycerol derivatives is ambiguous in the literature,[4–9] the validity of $J_{7,8}$ and $\Delta\delta_{C8-C7}$ values to distinguish *threo* and *erythro* 8–4′-oxyneolignan and arylglycerol derivatives is discussed on the basis of NMR data of *threo* and *erythro* 8–4′-oxyneolignans and arylglycerols in different solvents. Some *in vitro* bioassays are also reported.

## Results and Discussion

Compound **1** was obtained as an amorphous solid, and the presence of OH (3358 $cm^{-1}$) and aromatic (1602 and 1509 $cm^{-1}$) groups were indicated by its IR spectrum. The positive mode ESIMS of **1** gave a quasi-molecular ion peak at *m*/*z* 547 $[M + Na]^+$, and the molecular formula $C_{25}H_{32}O_{12}$ was indicated by HRFABMS at *m*/*z* 547.1801. The $^1H$ NMR of **1** in DMSO-$d_6$ showed signals attributed to two 1,3,4-trisubstituted aromatic rings at $\delta$ 6.97 (H-2), 6.68 (H-5), and 6.80 (H-6), and 7.19 (H-2′), 6.89 (H-5′), and 6.93 (H-6′), together with signals attributed to an aromatic methoxy at $\delta$ 3.72 and an exchangeable phenolic OH proton at $\delta$ 8.80 (O*H*-4). A *trans*-arylpropenoxy unit was indicated by signals at $\delta$ 6.41 (H-7′), 6.21 (H-8′), and 4.06 ($H_2$-9′). Meanwhile, an arylglyceryloxy unit was indicated by signals of a vicinal coupling system attributed to two oxymethines at $\delta$ 4.69 (H-7) and 4.27 (H-8) and an oxymethylene at $\delta$ 3.63 (H-9a) and 3.51 (H-9b). A doublet assignable to an anomeric proton at $\delta$ 4.79, partially overlapped signals attributed to oxymethylene and oxymethine protons between $\delta$ 3.13 and 3.70, and exchangeable OH protons between $\delta$ 4.50 and 5.35 suggested that there was a $\beta$-glycosyl moiety in **1**. Enzymatic hydrolysis of **1** produced **1a** and a sugar. The sugar gave a positive optical rotation, $[\alpha]$ +47.2, indicating that it was D-glucose.[10] The $^{13}C$ NMR and DEPT spectra of **1** showed carbon signals corresponding to the above units (Table 2). The presence of four oxygen-bearing aromatic carbons ($\delta$ > 145 ppm) in the $^{13}C$ NMR spectrum, in combination with the chemical shifts and coupling patterns of the protons of the two aromatic rings in the $^1H$ NMR spectrum and the molecular composition, suggested that **1** was a 8–4′-oxyneolignan $\beta$-D-glucopyranoside with one phenolic OH and one aromatic methoxy groups. However, the spectroscopic data of **1** were different from those of related known compounds.[11–13]

Extensive analysis of HMQC and $^1H-^1H$ COSY spectra of **1** provided unambiguous assignments of proton and carbon signals in the NMR spectra. In the HMBC spectrum of **1**, long-range correlations from H-7 to C-1, C-2, C-6, C-8, and C-9 and from H-7′ to C-1′, C-2′, C-6′, C-8′, and C-9′ (Figure S1, Supporting Information), in combination with chemical shifts and coupling patterns, confirmed the presence of 3,4-disubstituted phenylglyceryloxy and 3′,4′-disubstituted *trans*-phenylpropenoxy units. HMBC correlations of C-3 with H-2, H-5, and the methoxy protons and of C-4 with H-2 and H-6 proved that the methoxy was located at C-3. Correlations of C-3′ with H-2′, H-5′, and the anomeric proton and of C-4′ with H-2′ and H-6′, and the chemical shift of C-3′, indicated that the glucose was attached at C-3′. This conclusion was supported by NOE enhancements of H-7′, H-8′, and H-1″ by irradiation of H-2′ in the NOE difference experiment (Figure S1, Supporting Information). Although a correlation from H-8 to C-4′ was not observed in the HMBC spectrum of **1**, NOE enhancements of H-2, H-6, and H-5′ by irradiation of H-8 indicated a connection between C-8 and C-4′ in **1**.

The stereochemistry of **1** was elucidated by a comprehensive analysis of the NMR and CD data of **1**, **1a**, and the acetonide **1b**. The $^1H$ NMR spectra of **1a** and **1b** (Table S1, Supporting Information) showed $J_{7,8}$ values of 4.2 and 9.0 Hz, respectively. This suggested that **1** possessed an *erythro* relative configuration.[11,12,14–18] The CD spectra of **1** and **1a** showed negative Cotton effects at 233 and 236 nm (Figures S2 and S3, Supporting Information), respectively, indicating 8*R* configuration for these compounds.[18,19] Thus, **1** was determined to be (−)-(7*S*,8*R*,7′*E*)-4,7,9,3′,9′-pentahydroxy-3-methoxy-8–4′-oxyneolign-7′-ene-3′-*O*-$\beta$-D-glucopyranoside.

Compound **2** was obtained as an amorphous powder, and the molecular formula $C_{25}H_{34}O_{12}$ was indicated by HRESIMS at *m*/*z* 549.1963 $[M + Na]^+$. The UV, IR, and NMR spectroscopic data of **2** indicated that it was a diastereomer of (7*R*,8*R*)-4,7,9,9′-tetrahydroxy-3-methoxy-8-*O*-4′-neolignan-3′-*O*-$\beta$-D-glucopyranoside,[15] which was confirmed by the HMBC experiment of **2**. Hydrolysis of **2** with $\beta$-glucosidase librated **2a** and D-glucose. The

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**Table 1.** $^1$H NMR Data for Compounds **1**−**7**[a]

| no. | 1 | 2 | 3 | 4 | 5 | 6 | 7[b] |
|---|---|---|---|---|---|---|---|
| 1a | | | | | | 4.07 dd (10.5, 5.0) | |
| 1b | | | | | | 3.80 dd (10.5, 5.0) | |
| 2 | 6.97 brs | 6.97 brs | 6.88 brs | 6.95 d (1.5) | 6.96 d (1.5) | 4.54 quint (5.0) | 6.92 brs |
| 3a | | | | | | 3.77 dd (12.0, 5.0) | |
| 3b | | | | | | 3.74 dd (12.0, 5.0) | |
| 5 | 6.68 d (8.0) | 6.68 d (8.0) | 6.64 d (8.0) | 6.69 d (8.0) | 6.67 d (8.0) | | 7.04 d (8.0) |
| 6 | 6.80 brd (8.0) | 6.78 brd (8.0) | 6.67 brd (8.0) | 6.75 dd (8.0, 1.5) | 6.75 dd (8.0, 1.5) | | 6.80 brd (8.0) |
| 7 | 4.69 d (5.5) | 4.69 d (5.5) | 4.72 d (4.5) | 4.69 d (4.5) | 4.68 d (4.5) | | 5.46 d (5.5) |
| 8 | 4.27 ddd (6.0,5.5,3.0) | 4.18 ddd (5.5, 5.0, 3.5) | 4.24 ddd (6.0, 4.5, 3.0) | 3.93 ddd (6.5, 4.5, 3.5) | 4.16 ddd (5.5, 4.5, 4.0) | | 3.35 m |
| 9a | 3.63 dd (12.0, 6.0) | 3.62 dd (11.0, 5.0) | 3.70 dd (11.5, 6.0) | 3.58 dd (11.5, 6.5) | 3.55 dd (11.0, 4.0) | | 3.60 m |
| 9b | 3.51 dd (12.0, 3.0) | 3.48 dd (11.0, 3.5) | 3.38 dd (11.5, 3.0) | 3.48 d (11.5, 3.5) | 3.22 dd (11.0, 5.5) | | 3.49 m |
| 2′ | 7.19 d (1.0) | 6.94 d (2.0) | 6.90 d (1.5) | 6.60 d (1.5) | 6.81 d (1.5) | 7.24 d (2.0) | 6.55 brs |
| 5′ | 6.89 d (8.0) | 6.82 d (8.0) | | 6.65 d (8.0) | 6.90 d (8.5) | 7.13 d (8.0) | |
| 6′ | 6.93 dd (8.0, 1.0) | 6.70 dd (8.0, 2.0) | 6.77 d (1.5) | 6.39 dd (8.0, 1.5) | 6.66 dd (8.5, 1.5) | 7.20 dd (8.0, 2.0) | 6.60 brs |
| 7′ | 6.41 d (16.0) | 2.47 t (7.0) | 6.43 d (16.0) | 2.46 t (7.0) | 2.56 t (7.5) | 7.55 d (16.0) | 4.83 brs |
| 8′ | 6.21 dt (16.0, 5.0) | 1.66 quint (7.0) | 6.31 dt (16.0, 5.0) | 1.73 quint (7.0) | 1.78 m | 6.63 dd (16.0, 7.5) | 2.70 m |
| 9′a | 4.06 d (5.0) | 3.37 t (7.0) | 4.08 d (5.0) | 3.74 dt (10.0, 7.0) | 3.79 dt ((10.0, 6.0) | 9.54 d (7.5) | 3.69 m |
| 9′b | | | | 3.38 dt (10.0, 7.0) | 3.39 dt (10.0, 7.0) | | 3.47 m |
| 1″ | 4.79 d (7.0) | 4.75 d (7.5) | 4.80 d (7.0) | 4.09 d (8.0) | 4.09 d (8.0) | 4.29 d (7.5) | 4.87 d (7.5) |
| 2″ | 3.27 m | 3.26 m | 3.28 m | 2.94 dd (8.5, 8.0) | 2.95 dd (8.0, 8.0) | 3.14 dd (8.5, 7.5) | 3.23 m |
| 3″ | 3.29 m | 3.30 m | 3.26 m | 3.12 dd (8.5, 8.5) | 3.14 dd (8.0, 8.0) | 3.23 dd (8.5, 8.5) | 3.24 m |
| 4″ | 3.15 dd (9.0, 8.5) | 3.14 m | 3.12 m | 3.03 dd (8.5, 8.5) | 3.02 dd (8.0, 8.5) | 3.23 dd (9.0, 8.5) | 3.14 m |
| 5″ | 3.26 m | 3.25 m | 3.28 m | 3.04 m | 3.07 m | 3.29 m | 3.24 m |
| 6″a | 3.68 brd (11.5) | 3.66 brd (12.0) | 3.69 brd (11.5) | 3.64 brd (12.0) | 3.63 dd (12.0, 2.0) | 3.81 dd (11.5, 2.0) | 3.66 dd (12.0, 2.0) |
| 6″b | 3.46 dd (11.5, 5.5) | 3.46 dd (12.0, 5.5) | 3.48 dd (11.5, 5.5) | 3.43 dd (12.0, 5.0) | 3.41 dd (12.0, 6.0) | 3.60 dd (11.5, 5.0) | 3.46 dd (12.0, 5.0) |
| 3-OMe | 3.72 s | 3.72 s | 3.72 s | 3.71 s | 3.68 s | | 3.73 s |
| 3′/5′-OMe | | | 3.74 s | | 3.70 s | 3.84 s | 3.63 s |

[a] $^1$H NMR data ($\delta$) were measured in DMSO-$d_6$ for **1**−**5** and **7** and MeOH-$d_4$ for **6** at 500 MHz. Proton coupling constants ($J$) in Hz are given in parentheses. The assignments were based on DEPT, $^1$H−$^1$H COSY, HMQC, and HMBC experiments. [b] Data of the 3‴-methoxy-4‴-hydroxyphenyl unit at C-7′ of **7**, $\delta$ 6.56 (1H, brs, H-2‴), 6.58 (1H, d, $J$ = 8.0 Hz, H-5‴), 6.48 (1H, brd, $J$ = 8.0 Hz, H-6‴), 3.61 (3H, s, OMe-3‴).

**Table 2.** $^{13}$C NMR Data for Compounds **1**−**7**[a]

| no. | **1** | **2** | **3** | **4** | **5** | **6** | **7**[b] |
|---|---|---|---|---|---|---|---|
| 1 | 132.9 | 133.0 | 132.7 | 132.7 | 133.0 | 69.1 | 135.6 |
| 2 | 111.2 | 111.2 | 111.0 | 111.4 | 111.0 | 80.5 | 110.2 |
| 3 | 147.1 | 147.0 | 147.1 | 147.1 | 147.0 | 62.0 | 148.9 |
| 4 | 145.5 | 145.4 | 145.5 | 145.6 | 145.4 | | 146.1 |
| 5 | 114.8 | 114.8 | 114.8 | 114.8 | 114.6 | | 115.3 |
| 6 | 119.6 | 119.6 | 119.3 | 119.4 | 119.0 | | 117.8 |
| 7 | 71.4 | 71.3 | 71.6 | 71.8 | 71.0 | | 86.5 |
| 8 | 85.6 | 85.9 | 86.4 | 87.0 | 84.8 | | 53.5 |
| 9 | 59.9 | 59.8 | 59.6 | 60.2 | 60.1 | | 63.4 |
| 1′ | 131.0 | 136.0 | 132.4 | 136.5 | 134.8 | 129.6 | 138.4 |
| 2′ | 115.3 | 117.9 | 108.0 | 116.5 | 112.9 | 112.7 | 110.8 |
| 3′ | 148.4 | 148.2 | 151.2 | 149.7 | 149.5 | 151.9 | 142.8 |
| 4′ | 147.9 | 146.4 | 135.4 | 144.9 | 146.4 | 151.9 | 145.9 |
| 5′ | 118.5 | 118.8 | 153.0 | 119.3 | 116.0 | 117.2 | 127.6 |
| 6′ | 120.8 | 122.3 | 104.8 | 117.8 | 120.2 | 124.5 | 114.9 |
| 7′ | 128.2 | 31.1 | 128.4 | 31.0 | 31.1 | 155.4 | 72.5 |
| 8′ | 129.1 | 34.1 | 130.3 | 31.1 | 31.2 | 127.9 | 55.2 |
| 9′ | 61.5 | 60.1 | 61.5 | 67.8 | 67.9 | 196.1 | 62.6 |
| 1″ | 101.8 | 101.8 | 101.8 | 102.9 | 103.0 | 104.7 | 100.1 |
| 2″ | 73.6 | 73.6 | 73.6 | 73.4 | 73.5 | 75.0 | 73.2 |
| 3″ | 76.4 | 76.4 | 76.3 | 76.7 | 76.7 | 78.0 | 76.8 |
| 4″ | 69.8 | 69.7 | 69.9 | 70.0 | 70.0 | 71.6 | 69.6 |
| 5″ | 77.1 | 77.0 | 77.3 | 76.8 | 76.8 | 78.0 | 77.0 |
| 6″ | 60.8 | 60.7 | 60.8 | 61.0 | 61.1 | 62.8 | 60.6 |
| 3-OMe | 55.7 | 55.7 | 55.6 | 55.6 | 55.4 | 56.6 | 55.7 |
| 3′/5′-OMe | | | 56.0 | | 55.6 | | 55.5 |
| $\Delta\delta_{C8-C7}$ | 14.2 | 14.6 | 14.8 | 15.2 | 13.8 | | |

[a] $^{13}$C NMR data ($\delta$) were measured in DMSO-$d_6$ for **1**−**5** and **7** and MeOH-$d_4$ for **6** at 125 MHz. The assignments were based on DEPT, $^1$H−$^1$H COSY, HMQC, and HMBC experiments. [b] Data of the 3‴-methoxy-4‴-hydroxyphenyl unit at C-7′ of **7**, $\delta$ 131.2 (C-1‴), 113.9 (C-2‴), 146.5 (C-3‴), 144.7 (C-4‴), 114.5 (C-5‴), 121.8 (C-6‴), 55.4 (OMe-3‴).

$^1$H NMR spectra of **2a** and its acetonide **2b** in $CDCl_3$ showed $J_{7,8}$ values of 3.6 and 9.0 Hz (Table S1, Supporting Information), respectively, suggesting that **2** possessed an *erythro* relative configuration.[11,17] The CD spectra of **2** and **2a** displayed positive Cotton effects at 237 and 238 nm (Figures S2 and S3, Supporting Information), respectively, indicating that **2** and **2a** possessed an 8*S* configuration.[18,19] Therefore, **2** was determined to be (−)-(7*R*,8*S*)-4,7,9,3′,9′-pentahydroxy-3-methoxy-8–4′-oxyneolignan-3′-*O*-$\beta$-D-glucopyranoside.

Compound **3** was obtained as an amorphous powder, and its HRESIMS at *m*/*z* 577.1851 $[M + Na]^+$ indicated the molecular formula $C_{26}H_{34}O_{13}$. The UV, IR, and NMR spectroscopic features of **3** were similar to those of **1**, except that the NMR signals of the 3′,4′-disubstituted *trans*-arylpropenoxy unit in **1** were replaced by those attributed to a 3′,4′,5′-trisubstitued *trans*-arylpropenoxy unit and a methoxy in **3** (Tables 1 and 2). These data indicated that **3** was an analogue of **1** with an additional methoxy at C-5′. This was confirmed by the HMBC spectrum of **3**, which showed correlations of C-3′ with H-2′ and H-1″, C-4′ with H-2′ and H-6′, and C-5′ with H-6′ and the additional methoxy protons. An *erythro* configuration of **3** was confirmed by the $J_{7,8}$ (4.2 Hz) of **3a** in $CDCl_3$. In the CD spectra of **3** and **3a**, respective negative Cotton effects at 235 and 236 nm (Figures S2 and S3, Supporting Information) indicated that they had 8*R* configuration. Therefore, **3** was determined to be (−)-(7*S*,8*R*,7′*E*)-4,7,9,3′,9′-pentahydroxy-3,5′-dimethoxy-8–4′-oxyneolign-7′-ene-3′-*O*-$\beta$-D-glucopyranoside.

Compound **4** was assigned the molecular formula $C_{25}H_{34}O_{12}$, as indicated by HRESIMS. The NMR data suggested that **4** was an isomer of **2**. The anomeric proton of **4** was shielded $\Delta\delta$ 0.66 ppm by comparison with that of **2**, while two multiplets at $\delta$ 3.74 (H-9′a) and 3.38 (H-9′b) in **4** replaced the triplet of $H_2$-9′ in **2**. This indicated that the glucopyranosyloxy moiety was located at C-9′ in **4**, which was confirmed by the HMBC experiment of **4** showing correlations from both H-9′a and H-9′b to C-1″ and from H-1″ to C-9′. The $J_{7,8}$ (3.6 Hz) of **4a** in $CDCl_3$ (Table S1) suggested that **4** had an *erythro*-configuration. In the CD spectra of **4** and **4a**, a positive Cotton effect at 238 nm (Figures S2 and S3) suggested that they both had an 8*S* configuration. Consequently, **4** was determined to be (−)-(7*R*,8*S*)-4,7,9,3′,9′-pentahydroxy-3-methoxy-8–4′-oxyneolignan-9′-*O*-$\beta$-D-glucopyranoside.

The NMR data of **5** (Tables 1 and 2) were in good agreement with those of (−)-(7*R*,8*R*)-4,7,9,9′-tetrahydroxy-3,3′-dimethoxy-8-*O*-4′-neolignan-9′-*O*-$\beta$-D-glucopyranoside.[15] However, the optical rotation and CD data of **5** and its aglycone **5a** (Figures S2 and S3) were opposite of that reported. Therefore, the structure of **5** was assigned as (+)-(7*S*,8*S*)-4,7,9,9′-tetrahydroxy-3,3′-dimethoxy-8–4′-oxyneolignan-9′-*O*-$\beta$-D-glucopyranoside. This was supported by the $^1$H NMR data of **5a** and its acetonide **5b** in $CDCl_3$, which displayed $J_{7,8}$ values of 8.4 and ∼0.0 Hz (Table S2), respectively.

Compound **6** was obtained as an amorphous powder, and its IR spectrum showed absorption bands for OH (3379 cm$^{-1}$), conjugated CO (1662 cm$^{-1}$), and aromatic (1595 and 1511 cm$^{-1}$) groups. The molecular formula $C_{19}H_{26}O_{10}$ of **6** was indicated by HRFABMS. The NMR spectra of **6** in MeOH-$d_4$ displayed resonances due to glyceryl, 3′,4′-disubstituted *trans*-phenylpropenal, and $\beta$-glucopyranosyl moieties and to a methoxy signal (Tables 1 and 2). Enzymatic hydrolysis of **6** yielded D-glucose and **6a**. The $^1$H NMR and ESIMS spectroscopic data of **6a** were consistent with those of 2-{2-methoxy-4-[(*E*)-formylvinyl]phenoxyl}propan-1,3-diol.[20] The HMBC experiment of **6** revealed that it was 1-*O*-$\beta$-D-glucopyranosyl-2-{2-methoxy-4-[(*E*)-formylvinyl]phenoxyl}propan-3-ol. The negative optical rotation of **6** suggested that it had a 2*R* configuration.[21] Therefore, **6** was determined to be as (−)-(2*R*)-1-*O*-$\beta$-D-glucopyranosyl-2-{2-methoxy-4-[(*E*)-formylvinyl]phenoxyl}propane-3-ol.

Compound **7** was obtained as a gum, and its molecular formula $C_{33}H_{40}O_{14}$ was indicated by HRESIMS. The IR spectrum of **7** displayed absorption bands for OH (3426 cm$^{-1}$) and aromatic ring (1608 and 1514 cm$^{-1}$) groups. The $^1$H NMR spectrum of **7** showed signals due to two 1,3,4-trisubstituted phenyl groups, a 1,3,4,5-tetrasubstituted phenyl group, and three methoxy groups. In addition, it showed signals attributed to three oxymethine protons at $\delta$ 5.46 (H-7), 4.83 (H-7′), and 4.87 (H-1″), partially overlapped methylene and methine protons between $\delta$ 3.14 and 3.70, and broad signals due to eight OH protons between $\delta$ 4.10 and 5.30. In addition to carbon resonances corresponding to the above units, the $^{13}$C NMR and DEPT spectra of **7** displayed carbon signals attributed to two oxymethines at $\delta$ 86.5 (C-7) and 72.5 (C-7′), two oxymethylenes at $\delta$ 63.4 (C-9) and 62.6 (C-9′), and two aliphatic methines at $\delta$ 53.5 (C-8) and 55.2 (C-8′). These spectroscopic data suggested that **7** was a dihydro[*b*]benzofuran neolignan glycoside with an additional tri- or disubstituted aryl group.[22] Enzymatic hydrolysis of **7** produced D-glucose and **7a**, and the $^1$H NMR data of **7a** were in agreement with those of leptolepisol C isolated from *Larix leptolepis*.[23] This indicated that **7** was leptolepisol C $\beta$-D-glucoside, which was confirmed by HMQC and HMBC experiments of **7**. In the HMBC spectrum, a correlation from the anomeric proton to C-4 indicated unequivocally that the $\beta$-glycopyranosyl moiety was located at C-4. Consequently, **7** was determined to be leptolepisol C 4-*O*-$\beta$-D-glucopyranoside. The CD spectra of **7** and **7a** did not give any useful Cotton effect due to interaction among three aromatic rings, and the absolute configuration of **7** and **7a** remains to be determined.

Compound **8** showed IR absorption bands for OH (3374 cm$^{-1}$) and aromatic (1612 and 1513 cm$^{-1}$) functional groups. The ESIMS of **8** gave $[M + Na]^+$ and $[M + K]^+$ peaks at *m*/*z* 429 and 445. The $^1$H NMR spectrum of **8** in DMSO-$d_6$ showed a two-proton aromatic singlet at $\delta$ 6.64, a six-proton methoxy singlet at $\delta$ 3.75, two deshielded oxymethine doublets at $\delta$ 4.63 (H-7) and 4.25 (H-1′), and partially overlapped oxymethylene and/or oxymethine multiplets integrated for nine protons between $\delta$ 3.00 and 3.75

**Table 3.** NMR Data for Compounds **8** and **9**[a]

| | **8** | | **9** | |
|---|---|---|---|---|
| no. | H | C | H | C |
| 1 | | 131.9 | | 132.7 |
| 2 | 6.64 s | 104.6 | 6.97 d (1.2) | 111.5 |
| 3 | | 147.4 | | 147.0 |
| 4 | | 134.3 | | 145.4 |
| 5 | | 147.4 | 6.67 d (8.0) | 114.6 |
| 6 | 6.64 s | 104.6 | 6.73 dd (8.0, 1.2) | 119.4 |
| 7 | 4.63 d (4.4) | 72.6 | 4.63 d (4.0) | 72.5 |
| 8 | 3.75 m | 84.6 | 3.72 m | 84.9 |
| 9a | 3.42 dd (11.2, 4.4) | 61.1 | 3.39 dd (11.2, 5.2) | 61.1 |
| 9b | 3.29 dd (11.2, 4.8) | | 3.25 dd (11.2, 4.8) | |
| 1′ | 4.25 d (8.0) | 103.1 | 4.26 d (7.6) | 103.3 |
| 2′ | 3.00 dd (8.8,8.0) | 73.8 | 3.00 dd (8.8, 7.6) | 73.9 |
| 3′ | 3.10 dd (8.8, 8.8) | 76.4 | 3.13 dd (8.8,8.8) | 76.4 |
| 4′ | 3.02 dd (8.8, 8.8) | 70.1 | 3.02 dd (8.8, 8.4) | 70.1 |
| 5′ | 3.10 m | 76.9 | 3.10 m | 76.9 |
| 6′a | 3.66 dd (11.6, 1.6) | 61.0 | 3.64 brd (12.0) | 61.0 |
| 6′b | 3.38 dd (11.6, 5.2) | | 3.39 (12.0, 6.4) | |
| OMe | 3.73 s | 55.9 | 3.74 s | 55.6 |

[a] $^1$H NMR data ($\delta$) were measured in DMSO-$d_6$ at 400 MHz. Proton coupling constants ($J$) in Hz are given in parentheses. $^{13}$C NMR data ($\delta$) were measured in DMSO-$d_6$ at 125 MHz.

(Table 3). The $^{13}$C NMR and DEPT spectra of **8** displayed characteristic signals for 1-*C*-syringylglycerol and $\beta$-glucopyranosyl moieties (Table 3). Enzymatic hydrolysis of **8** with $\beta$-glucosidase yielded **8a** with [α] −19.6 (*c* 0.25, MeOH) and D-glucose. The NMR data of **8a** (Tables S11 and S13) were in good agreement with those of *erythro*-1-*C*-syringylglycerol,[24,25] indicating that it was (−)-*erythro*-1-*C*-syringylglycerol $\beta$-D-glucopyranoside. Comparison of the NMR data of **8** and **8a** indicated that C-8 of **8** was significantly deshielded by $\Delta\delta$ 9.3 ppm. This suggested that the $\beta$-D-glucopyranosyl moiety was located at C-8 of (−)-*erythro*-syringylglycerol in **8**. Since *erythro*-arylglycerols with 7*R*,8*S* configuration were reported to have negative $[\alpha]_D$ values,[26,27] the absolute configuration at C-7 and C-8 of **8a** was assigned as 7*R*,8*S*. Thus, the structure of **8** was determined to be (−)-(7*R*,8*S*)-syringylglycerol 8-*O*-$\beta$-D-glucopyranoside.

Compound **9** gave $[M + Na]^+$ and $[M + K]^+$ peaks at *m*/*z* 399 and 415 in the positive ESIMS. The IR and NMR data were similar to those of **8** except that the syringyl unit in **8** was replaced by a guaiacyl unit in **9**. This suggested that **9** was a demethoxy derivative of **8**. Hydrolysis of **9** yielded **9a**, with [α] –12.4 (*c* 0.33, MeOH), and D-glucose. The NMR data of **9a** (Tables S12 and S13) were identical to those of (7*S*,8*R*)-guaiacylglycerol except for an opposite optical rotation ([α] +11).[8,27] Therefore, **9** was determined to be (−)-(7*R*,8*S*)-guaiacylglycerol 8-*O*-$\beta$-D-glucopyranoside.

The known compounds were identified by comparison of spectroscopic data with those reported in the literature as (−)-(7*R*,8*S*,7′*E*)-4,7,9,9′-tetrahydroxy-3,3′-dimethoxy-8–4′-oxyneolign-7′-ene-9′-*O*-$\beta$-D-glucopyranoside (hyuganoside IIIa, **10**),[11] (−)-(7*S*,8*S*,7′*E*)-4,7,9,9′-tetrahydroxy-3,3′-dimethoxy-8–4′-oxyneolign-7′-ene-9′-*O*-$\beta$-D-glucopyranoside (hyuganoside IIIb, **11**),[11] (+)-(7*S*,8*S*)-syringylglycerol 8-*O*-$\beta$-D-glucopyranoside (**12**),[7] (+)-(7*S*,8*S*)-guaiacylglycerol 8-*O*-$\beta$-D-glucopyranoside (**13**),[30] (−)-(7*S*,8*R*)-guaiacylglycerol 9-*O*-$\beta$-D-glucopyranoside (**14**),[8] (−)-(7*R*,8*R*)-guaiacylglycerol 9-*O*-$\beta$-D-glucopyranoside (**15**),[8] (−)-(7*R*,8*R*)-syringylglycerol 9-*O*-$\beta$-D-glucopyranoside (**16**),[31] (−)-(7*R*,8*R*)-guaiacylglycerol 7-*O*-$\beta$-D-glucopyranoside,[32] (−)-tachioside,[33] (−)-liriodendrin (**17**),[34] (−)-sweroside,[35] (−)-11,13-dihydrodeacylcynaropicrin 3-*O*-$\beta$-D-glucopyranoside,[36] (−)-3,5-dimethoxy-4-hydroxyphenyl $\beta$-D-glucopyranoside (incorrect nomenclature was given in the literature[37]), (−)-(1′*R*)-1′-(3-hydroxy-4-methoxyphenyl)ethane-1′,2′-diol 3-*O*-$\beta$-D-glucopyranoside,[38] (−)-3-hydroxy-1-(4-hydroxy-3-methoxyphenyl)-1-propanone 3-*O*-$\beta$-D-glucopyranoside,[39] (−)-2-hydroxy-5-(2-hydroxyethyl)phenyl $\beta$-D-glucopyranoside,[32] (−)-2-methoxy-4-(1-propionyl)phenyl $\beta$-D-glucopyranoside,[40] and (−)-4-propionyl-2,6-dimethoxyphenyl $\beta$-D-glucopyranoside.[41] The absolute configurations of **10**, **11**, and **12** were determined on the basis of CD spectra and/or optical rotations of their aglycones, and **13** was obtained as a natural product for the first time.

Coupling constants ($J_{7,8}$) of the deshielded benzylic proton (H-7) signal in the $^1$H NMR spectra of the acetates of *erythro* ($J_{7,8} \leq 5.3$ Hz) and *threo* ($J_{7,8} \geq 6.3$ Hz) 8–4′-oxyneolignan aglycones and glycosides,[12,18] as well as acetonide derivatives of *threo* ($J_{7,8} < 2.0$ Hz) and *erythro* ($J_{7,8} > 8.0$ Hz) 8–4′-oxyneolignan aglycones,[11,17] unambiguously distinguished *erythro* and *threo* isomers in $CDCl_3$. However, there were several cases where the values of $J_{7,8}$ of 8–4′-oxyneolignans were directly applied for the differentiation of *erythro* and *threo* isomers without derivatization.[5,6,14a,15,42] A systematic analysis of $^1$H NMR data of **1**−**5** in DMSO-$d_6$ (Table 1) and **1a**−**5a**, **10a**, **11a**, **5**, **10**, and **11** in different solvents (Tables S1−S5, Supporting Information), in combination with the data of 8–4′-oxyneolignan derivatives in the literature,[11−15,17,44] indicated that the values of $J_{7,8}$ were variable in different solvents due to possible dynamic conformational changes.[43] In $Me_2CO$-$d_6$ + $D_2O$ or $CDCl_3$, the $J_{7,8}$ values of the *threo* 8–4′-oxyneolignan aglycones **5a** and **11a** (6.0 and 6.6 Hz in $Me_2CO$-$d_6$ + $D_2O$ and 8.4 and 8.4 Hz in $CDCl_3$, respectively) were larger than those of the *erythro* analogues **1a**, **2a** (**4a**), **3a**, and **10a** (4.8, 4.8, 3.0, and 5.4 Hz in $Me_2CO$-$d_6$ + $D_2O$ and 4.2, 3.6, 4.2, and 4.2 Hz in $CDCl_3$, respectively). Meanwhile, in $C_5D_5N$, the $J_{7,8}$ values of the glycosides **5** and **11** (6.0 and 5.4 Hz) were larger than that of the *erythro* analogue **10** (4.8 Hz). However, in $CD_3OD$, the $J_{7,8}$ values of the *threo* aglycones **5a** and **11a** (6.0 and 5.4 Hz) were smaller than or equal to the *erythro* aglycones **10a** (6.0 Hz), and in DMSO-$d_6$, $D_2O$, and $CD_3OD$, the $J_{7,8}$ values of *threo* glycosides **5** and **11** were smaller than or equal to those of the *erythro* glycosides. In addition, it was clear that the differences of the $J_{7,8}$ values between *threo* and *erythro* aglycones in $Me_2CO$-$d_6$ + $D_2O$ and glycosides in $C_5D_5N$ are small and close to the range of NMR instrument errors. Therefore, the direct application of the $J_{7,8}$ values was ambiguous to differentiate *erythro* and *threo* 8–4′-oxyneolignans with the exception of aglycone acetonides ($J_{7,8} > 8.0$ Hz for *erythro*, and $J_{7,8} < 2.0$ Hz for *threo*) and glycoside acetates ($J_{7,8} \leq 5.3$ Hz for *erythro*, and $J_{7,8} \geq 6.3$ Hz for *threo*) in $CDCl_3$, as well as aglycones in $CDCl_3$ ($J_{7,8} \leq 5.0$ Hz for *erythro*, and $J_{7,8} \geq 8.0$ Hz for *threo*).

In order to evaluate a possible application of the $^{13}$C NMR spectroscopic data for distinguishing *erythro* and *threo* 8–4′-oxyneolignan derivatives, the $^{13}$C NMR data of **1**−**5** in DMSO-$d_6$ (Table 2) and **1a**, **2a** (**4a**), **5a**, **10a**, **11a**, **2, 5**, **10**, and **11** in different solvents (Tables S6 and S7, Supporting Information), together with

the data of 8–4′-oxyneolignan derivatives in the literature,[11,15,17,44a] were investigated. The chemical shift difference between C-8 and C-7 ($\Delta\delta_{C8-C7}$) was used in the discussion to eliminate the systematic errors. The $\Delta\delta_{C8-C7}$ values of the *erythro* and *threo* isomers (**10** and **11**, and **10a** and **11a**) were variable in different solvents. In DMSO-$d_6$, $C_5D_5N$, or $CD_3OD$, the $\Delta\delta_{C8-C7}$ values of the *erythro* glycoside **10** (12.1, 12.6, or 12.1 ppm) were smaller than those of the *threo* isomer **11** (13.4, 13.9, or 13.1 ppm). Meanwhile, in $Me_2CO$-$d_6$, $CD_3OD$, or $CDCl_3$, the $\Delta\delta_{C8-C7}$ values of the *erythro* aglycone **10a** (12.9, 12.1, or 14.6 ppm) were smaller than those of the *threo* isomer **11a** (14.7, 13.2, or 15.5 ppm). These were fully consistent with data reported in the literature.[11,15,17,44a] However, in $D_2O$ the $\Delta\delta_{C8-C7}$ value of the *erythro* glycoside **10** (11.0 ppm) was larger than that of the *threo* isomer **11** (10.7 ppm), and in $CD_3OD$ the $\Delta\delta_{C8-C7}$ value of the *erythro* glycoside **2** (13.7 ppm) was larger than that of its *threo* isomer (12.9 ppm).[15] In the same solvent, the $\Delta\delta_{C8-C7}$ values of the 8–4′-oxyneolignan analogues [**1**−**5**, **1a**, **2a** (**4a**), and **10a**] were variable due to substituent differences at C-3′ and/or C-5′. Therefore, the $\Delta\delta_{C8-C7}$ values may be useful to distinguish the *erythro* and *threo* 8–4′-oxyneolignan isomers when the data are obtained in the same solvent.

The $J_{7,8}$ values of arylglycerol acetates were used to distinguish *threo*- ($J_{7,8} > 7.0$ Hz) and *erythro*- ($J_{7,8} < 6.5$ Hz) arylglycerols,[29,30,44b,45] and our previous investigation indicated that the chemical shift difference of C-7 and C-8 ($\Delta\delta_{C8-C7}$) was also applicable to differentiate *threo*- and *erythro*-arylglycerols without substituent(s) at C-7 and/or C-8 of the glycerol moiety.[25] A systematic analysis of the NMR data of **8**, **9**, **12**, and **13** in $D_2O$, DMSO-$d_6$, $C_5D_5N$, or $CD_3OD$ (Tables S8−S10, Supporting Information) and the data of *erythro*- and *threo*-arylglycerol 8-*O*-$\beta$-D-glucopyranosides in the literature[7–9,29–31] (the relative configurations at C-7 and C-8 were wrongly assigned in ref 9) indicated that the values of both $J_{7,8}$ and $\Delta\delta_{C8-C7}$ were variable in different solvents. However, in DMSO-$d_6$, $C_5D_5N$, or $CD_3OD$ except for $D_2O$, the differences of $J_{7,8}$ and $\Delta\delta_{C8-C7}$ between *erythro*- and *threo*-arylglycerol 8-*O*-$\beta$-D-glucopyranosides were significant and may be directly applicable to distinguish *erythro*- ($J_{7,8} \leq 4.4$ Hz in DMSO-$d_6$, $C_5D_5N$, or $CD_3OD$, $\Delta\delta_{C8-C7} \leq 12.5$ ppm in DMSO-$d_6$ or $C_5D_5N$, and $\Delta\delta_{C8-C7} < 11.0$ ppm in $CD_3OD$) and *threo*-arylglycerol 8-*O*-$\beta$-D-glucopyranosides ($J_{7,8} \geq 6.0$ Hz in DMSO-$d_6$, $C_5D_5N$, or $CD_3OD$, $\Delta\delta_{C8-C7} \geq 14.0$ ppm in DMSO-$d_6$ or $C_5D_5N$, and $\Delta\delta_{C8-C7} > 12.0$ ppm in $CD_3OD$). The $\Delta\delta_{C8-C7}$ of the aglycones **8a**, **9a**, **12a**, and **13a** in $Me_2CO$-$d_6$, $CD_3OD$, $C_5D_5N$, and DMSO-$d_6$ were consistent with those of our previously reported data for *erythro*- ($\Delta\delta_{C8-C7} < 1.0$ ppm in $Me_2CO$-$d_6$, $CD_3OD$, or $C_5D_5N$, and $\Delta\delta_{C8-C7} \leq 1.4$ in DMSO-$d_6$) and *threo*- ($\Delta\delta_{C8-C7} \geq 2.0$ ppm in all tested solvents) arylglycerols without substituent(s) at C-7 and/or C-8,[25] although the values of $J_{7,8}$ were indistinguishable among the aglycones without substituent(s) at C-7 and/or C-8 (Tables S11−S13). In addition, an investigation of the NMR data of three known *erythro*- and *threo*-arylglycerol 9-*O*-$\beta$-D-glucopyranosides (**14**−**16**) (Tables S14 and S15, Supporting Information) in different solvents, together with the data of *erythro*- and *threo*-arylglycerol 9-*O*-$\beta$-D-glucopyranosides[7–9,28,31] (the relative configurations at C-7 and C-8 were wrongly assigned in refs 7 and 9) indicated that there was no significant difference among $J_{7,8}$ values of *erythro*- and *threo*-arylglycerol 9-*O*-$\beta$-D-glucopyranosides. However, in DMSO-$d_6$ or $C_5D_5N$ except for in $D_2O$ or $CD_3OD$ $\Delta\delta_{C8-C7}$ values are applicable to distinguish *erythro*- ($\Delta\delta_{C8-C7} \leq 0.5$ ppm in DMSO-$d_6$ or $C_5D_5N$) and *threo*- ($\Delta\delta_{C8-C7} > 1.0$ ppm in DMSO-$d_6$ or $C_5D_5N$) arylglycerol 9-*O*-$\beta$-D-glucopyranosides.

The neuroprotective activity of the purified compounds against glutamate-induced neurotoxicity in cultures of PC12 cells was evaluated by the MTT assay. As shown in Table 4, treatment with glutamate (20 $\mu$M) resulted in significant inhibition of MTT reduction. However, exposure of compounds **4** and **17** at the concentration of $10^{-5}$ M for 24 h remarkably attenuated glutamate-induced cytotoxicity, whereas, **11**−**13** increased the cell damage. The other compounds were all inactive.

**Table 4.** Activities of Selected Compounds to Glutamate-Induced Neurotoxicity in PC12 Cells[a]

| compound | relative protection (%) |
|---|---|
| control | 100 ± 1.9 |
| glutamate-treated | 0.0 ± 2.8# |
| NGF | 108.2 ± 2.1** |
| **4** | 36.4 ± 3.1* |
| **9** | −46.6 ± 2.2** |
| **12** | −28.4 ± 1.9** |
| **13** | −32.9 ± 1.9** |
| **17** | 20.5 ± 1.8** |

[a] The data are expressed as mean ± SD of three independent experiments. (#$p < 0.05$ vs control; *$p < 0.05$, **$p < 0.01$ vs glutamate-treated group).

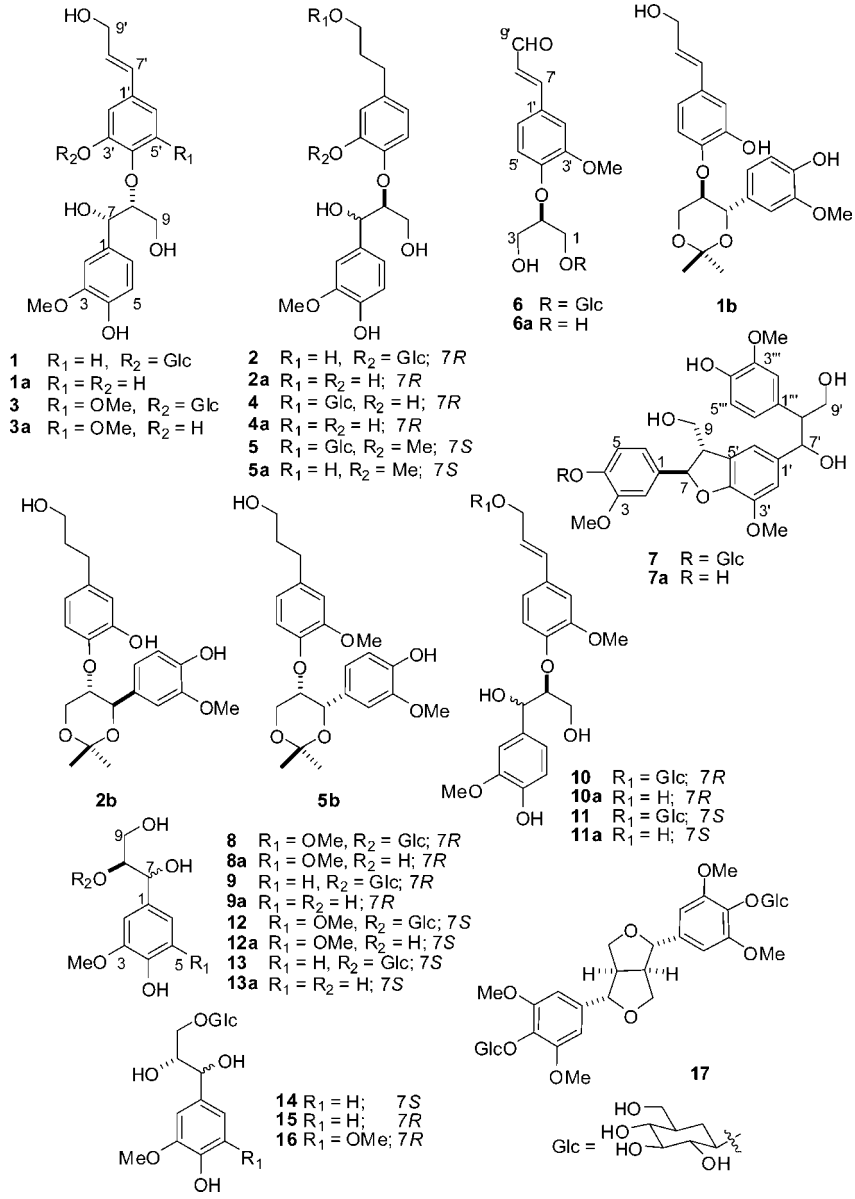
## Experimental Section

**General Experimental Procedures.** Optical rotations were measured on a Rudolph Research Autopol III automatic polarimeter. UV spectra were measured on a Cary 300 spectrophotometer. CD spectra were recorded on a JASCO J-810 spectropolarimeter. IR spectra were recorded as KBr disks on a Nicolet 5700 FT-IR instrument. NMR spectra were obtained at 400, 500, or 600 MHz for $^1$H and 100, 125, or 150 MHz for $^{13}$C, respectively, on Inova 400, 500, and 600 MHz spectrometers in DMSO-$d_6$, $C_5D_5N$, $CD_3OD$, $Me_2CO$-$d_6$, $D_2O$, or $CDCl_3$ with solvent peaks (or TMS, in the case of $D_2O$) used as references. ESIMS data were measured with a Q-Trap LC/MS/MS (Turbo Ionspray Source) spectrometer. HRFABMS and HRESIMS data were respectively measured using a Micromass Autospec-Ultima ETOF and an AccuToFCS JMS-T100CS spectrometer. Column chromatography (CC) was performed with silica gel (200–300 mesh, Qingdao Marine Chemical Inc. 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 $\lambda$ absorbance detector with an Alltima (250 × 10 mm i.d.) preparative column packed with $C_{18}$ (5 $\mu$M). TLC was carried out with glass precoated silica gel $GF_{254}$ plates. Spots were visualized under UV light or by spraying with 7% $H_2SO_4$ in 95% EtOH followed by heating.

**Plant Material.** The root of *I. cirrhosa* (6.0 kg) was collected at Dayao Moutain, Guangxi Province, China, in August 2002. The plant was identified by Mr. Guang-Ri Long (Guangxi Forest Administration, Guangxi 545005, China). A voucher specimen (no. YG02011) was deposited at the Herbarium of the Department of Medicinal Plants, Institute of Materia Medica, Beijing, China.

**Extraction and Isolation.** The air-dried root of *I. cirrhosa* (6 kg) was powdered and extracted with 95% EtOH (3 × 15 L) at room temperature for 3 × 48 h. The ethanolic extract was evaporated under reduced pressure to yield a dark brown residue (640 g). The residue was suspended in $H_2O$ (2000 mL) and then partitioned with EtOAc (5 × 2000 mL). The aqueous phase was applied to a HDP100 macroporous adsorbent resin (1000 g, dried weight) column. A successive elution of the column with $H_2O$, 30% EtOH, 60% EtOH, and 95% EtOH (5000 mL each) yielded four corresponding portions after removing solvents. The portion (24.0 g) eluted by 30% EtOH was separated by MPLC over reversed-phase silica gel eluting with a gradient of increasing MeOH (0−60%) in $H_2O$ to give four fractions (A−D) on the basis of TLC analysis. Subsequent separation of fraction A (7.5g) over Sephadex LH-20 eluted with $H_2O$ gave four subfractions ($A_1$−$A_4$). Subfraction $A_2$ (0.98 g) was further fractionated via silica gel CC, eluting with $CHCl_3$−MeOH (8:1), to yield two fractions ($A_{2-1}$, $A_{2-2}$). Fraction $A_{2-1}$ (0.31 g) was subjected to reversed-phase preparative HPLC, using a mobile phase of MeOH−$H_2O$−HOAc (6.5:93.5:0.5), to afford **8** (110 mg), **9** (51 mg), **12** (15 mg), and **13** (15 mg). Fraction B (13.0 g) was separated by normal silica gel CC, eluting with a gradient of increasing MeOH (10−50%) in $CHCl_3$, to afford five fractions ($B_1$−$B_5$). $B_1$ (4.60 g) was further separated into five subfractions ($B_{1-1}$−$B_{1-5}$) by reversed-phase flash chromatography using step-gradient elution with increasing MeOH (0−30%) in $H_2O$. Fraction $B_{1-2}$ (0.68 g) was subjected to CC over Sephadex LH-20, eluting with $H_2O$, to give four mixtures ($B_{1-2-1}$−$B_{1-2-4}$). $B_{1-2-2}$ (96 mg) was purified by reversed-phase prepara-

**Chart 1**




tive HPLC using the mobile phase of MeCN−$H_2O$−HOAc (10:90:0.5) to afford **6** (16 mg). Fraction $B_{1–2–3}$ (54 mg) was separated by reversed-phase preparative HPLC using MeOH−$H_2O$−-HOAc (21:79:0.5) to yield **1** (13 mg) and **2** (15 mg). $B_{1–4}$ (0.75 g) was subjected to CC over Sephadex LH-20 with $H_2O$ as eluent to give four mixtures ($B_{1–4–1}$−$B_{1–4–4}$). Fraction $B_{1–4–3}$ (243 mg) was separated by reversed-phase preparative HPLC with the mobile phase MeOH−$H_2O$ (30:70) to yield **3** (10 mg), **4** (12 mg), **5** (26 mg), **7** (11 mg), **10** (17 mg), and **11** (21 mg).

**(−)-(7*S*,8*R*,7′*E*)-4,7,9,3′,9′-Pentahydroxy-3-methoxy-8–4′-oxyneolign-7′-ene-3′-*O*-*β*-D-glucopyranoside (1):** amorphous solid; $[\alpha]^{20}_D$ −6.2 (*c* 0.08, MeOH); UV (MeOH) $\lambda_{max}$ (log $\epsilon$) 205 (4.3), 261 (3.9) nm; CD (MeOH) 221 ($\Delta\epsilon$ –0.82), 233 ($\Delta\epsilon$ –2.08), 250 ($\Delta\epsilon$ +0.07) nm; IR (KBr) $\nu_{max}$ 3358, 2920, 1602, 1509, 1453, 1267, 1072, 1024 $cm^{-1}$; $^1H$ NMR (DMSO-$d_6$, 500 MHz) data, see Table 1; $^{13}C$ NMR (DMSO-$d_6$, 125 MHz) data, see Table 2; ESIMS *m/z* 547 $[M + Na]^+$ and 563 $[M + K]^+$; HRFABMS *m/z* 547.1801 $[M + Na]^+$ (calcd for $C_{25}H_{32}O_{12}Na$, 547.1791).

**(−)-(7*R*,8*S*)-4,7,9,3′,9′-Pentahydroxy-3-methoxy-8–4′-oxyneolignan-3′-*O*-*β*-D-glucopyranoside (2):** amorphous powder; $[\alpha]^{20}_D$ −17.6 (*c* 0.45, MeOH); UV (MeOH) $\lambda_{max}$ (log $\epsilon$) 207 (4.4), 278 (3.6) nm; CD (MeOH) 228 ($\Delta\epsilon$ –2.34), 237 ($\Delta\epsilon$ +1.54), 244 ($\Delta\epsilon$ –0.18) nm; IR (KBr) $\nu_{max}$ 3381, 1604, 1509, 1270, 1026 $cm^{-1}$; $^1H$ NMR (DMSO-$d_6$, 500 MHz) data, see Table 1; $^{13}C$ NMR (DMSO-$d_6$, 125 MHz) data, see Table 2; ESIMS *m/z* 549 $[M + Na]^+$ and 565 $[M + K]^+$; HRESIMS *m/z* 549.1963 $[M + Na]^+$ (calcd for $C_{25}H_{34}O_{12}Na$ 549.1948).

**(−)-(7*S*,8*R*,7′*E*)-4,7,9,3′,9′-Pentahydroxy-3,5′-dimethoxy-8–4′-oxyneolign-7′-ene-3′-*O*-*β*-D-glucopyranoside (3):** amorphous powder; $[\alpha]^{20}_D$ –9.0 (*c* 0.10, MeOH); UV (MeOH) $\lambda_{max}$ (log $\epsilon$) 204 (4.3), 221 (4.2), 269 (3.8) nm; CD (MeOH) 226 ($\Delta\epsilon$ –0.46), 235 ($\Delta\epsilon$ –2.84), 250 ($\Delta\epsilon$ +0.72) nm; IR (KBr) $\nu_{max}$ 3422, 1590, 1508,1076, 1037 $cm^{-1}$; $^1H$ NMR (DMSO-$d_6$, 500 MHz) data, see Table 1; $^{13}C$ NMR (DMSO-$d_6$, 125 MHz) data, see Table 2; ESIMS *m/z* 577 $[M + Na]^+$, 593 $[M + K]^+$, and 553 $[M - H]^-$; HRESIMS *m/z* 577.1851, $[M + Na]^+$ (cacld for $C_{26}H_{34}O_{13}Na$ 577.1897).

**(−)-(7*R*,8*S*)-4,7,9,3′,9′-Pentahydroxy-3-methoxy-8–4′-oxyneolignan-9′-*O*-*β*-D-glucopyranoside (4):** colorless gum; $[\alpha]^{20}_D$ −12.5 (*c* 0.08, MeOH); UV (MeOH) $\lambda_{max}$ (log $\epsilon$) 207 (4.4), 280 (3.7) nm; CD (MeOH) 226 ($\Delta\epsilon$ –1.52), 238 ($\Delta\epsilon$ +1.14), 250 ($\Delta\epsilon$ –0.12) nm; IR (KBr) $\nu_{max}$ 3413, 1604, 1512, 1277, 1030 $cm^{-1}$; $^1H$ NMR (DMSO-$d_6$, 500 MHz) data, see Table 1; $^{13}C$ NMR (DMSO-$d_6$, 125 MHz) data, see Table 2; ESIMS *m/z* 549 $[M + Na]^+$, 565 $[M + K]^+$, and 525 $[M - H]^-$; HRESIMS *m/z* 549.1963 $[M + Na]^+$ (cacld for $C_{25}H_{34}O_{12}Na$ 549.1948).

**(+)-(7*S*,8*S*)-4,7,9,9′-Tetrahydroxy-3,3′-dimethoxy-8–4′-oxyneolignan-9′-*O*-D-glucopyranoside (5):** amorphous solid; $[\alpha]^{20}_D$ +2.0 (*c* 1.20, MeOH); UV (MeOH) $\lambda_{max}$ (log $\epsilon$) 230 (4.1), 280 (3.4) nm; CD (MeOH) 226 ($\Delta\epsilon$ +0.48), 236 ($\Delta\epsilon$ +1.75), 250 ($\Delta\epsilon$ +0.15); IR (KBr) $\nu_{max}$ 3283,

1558, 1513, 1259, 1032 $cm^{-1}$; $^1H$ NMR (DMSO-$d_6$, 500 MHz) data, see Table 1; $^{13}C$ NMR (DMSO-$d_6$, 125 MHz) data, see Table 2; ESIMS *m/z* 563 $[M + Na]^+$ and 539 $[M - H]^-$; HRESIMS *m/z* 563.2097 $[M + Na]^+$ (cacld for $C_{26}H_{34}O_{12}Na$ 563.2104).

**(−)-(2*R*)-1-*O*-*β*-D-Glucopyranosyl-2-{2-methoxy-4-[(*E*)-formylvinyl]phenoxyl}propane-3-ol (6):** yellowish, amorphous powder; $[\alpha]^{20}_D$ −7.5 (*c* 0.08, MeOH); UV (MeOH) $\lambda_{max}$ (log $\epsilon$) 202 (4.4), 224 (4.3), 237 (4.3), 334 (4.5) nm; IR (KBr) $\nu_{max}$ 3379, 2921, 1662, 1595, 1511, 1272, 1137, 1078 $cm^{-1}$; $^1H$ NMR ($CD_3OD$, 500 MHz) data, see Table 1; $^{13}C$ NMR ($CD_3OD$, 125 MHz) data, see Table 2; ESIMS *m/z* 437 $[M + Na]^+$ and 453 $[M + K]^+$; HRFABMS *m/z* 437.1406 $[M + Na]^+$ (calcd for $C_{19}H_{26}O_{10}Na$, 437.1424).

**Leptolepisol C 4-*O*-*β*-D-glucopyranoside (7):** colorless gum; $[\alpha]^{20}_D$ −6.0 (*c* 0.10, MeOH); UV (MeOH) $\lambda_{max}$ (log $\epsilon$) 208 (4.8), 279 (4.0) nm; IR (KBr) $\nu_{max}$ 3426, 1608, 1514, 1268, 1030 $cm^{-1}$; $^1H$ NMR (DMSO-$d_6$, 500 MHz) data, see Table 1; $^{13}C$ NMR (DMSO-$d_6$, 125 MHz) data, see Table 2; ESIMS *m/z* 683 $[M + Na]^+$, 699 $[M + K]^+$, and 659 $[M - H]^-$; HRESIMS *m/z* 683.2338 $[M + Na]^+$ (calcd for $C_{33}H_{40}O_{14}Na$ 683.2316).

**(−)-(7*R*,8*S*)-Syringylglycerol 8-*O*-*β*-D-glucopyranoside (8):** white, amorphous powder; $[\alpha]^{20}_D$ −14.3 (*c* 0.30, MeOH); UV (MeOH) $\lambda_{max}$ (log $\epsilon$) 211 (4.2), 232 (3.7), 271 (3.4) nm; IR (KBr) $\nu_{max}$ 3374, 1612, 1513, 1228, 1073, 1043 $cm^{-1}$; $^1H$ NMR (DMSO-$d_6$, 400 MHz) and $^{13}C$ NMR (DMSO-$d_6$, 125 MHz) data, see Table 3; ESIMS *m/z* 429 $[M + Na]^+$ and 445 $[M + K]^+$.

**(−)-(7*R*,8*S*)-Guaiacylglycerol 8-*O*-*β*-D-glucopyranoside (9):** white, amorphous powder; $[\alpha]^{20}_D$ –21.5 (*c* 0.55, MeOH); UV (MeOH) $\lambda_{max}$ (log $\epsilon$) 230 (3.8), 278 (3.2) nm; IR (KBr) $\nu_{max}$ 3383, 1605, 1517, 1272, 1073, 1028 $cm^{-1}$; $^1H$ NMR (DMSO-$d_6$, 400 MHz) and $^{13}C$ NMR (DMSO-$d_6$, 125 MHz) data, see Table 3; ESIMS *m/z* 399 $[M + Na]^+$ and 415 $[M + K]^+$.

**Enzymatic Hydrolysis of 1–13.** A solution of each compound in $H_2O$ (3 mL) was individually hydrolyzed with *β*-glucosidase (10 mg, Almonds Lot 1264252, Sigma-Aldrich) at 37 °C for 24 or 36 h. Each reaction mixture was extracted with EtOAc (3 × 3 mL) to yield the individual EtOAc extract and $H_2O$ phase after removing the solvents.

The EtOAc extracts were separately chromatographed over silica gel, eluting with $CH_3Cl$−MeOH (12:1) for the hydrolysates from **1**−**7** (3–12 mg), **10** (6 mg), and **11** (11 mg) to yield aglycones **1a**–**7a**, **10a**, and **11a**, respectively, and eluting with $CH_3Cl$−MeOH (10:1) for the hydrolysates from **8**, **9**, **12**, and **13** (each 10 mg) to yield **8a**, **9a**, **12a**, and **13a**.

Compound **1a** (2.0 mg): $[\alpha]^{20}_D$ +1.5 (*c* 0.20, MeOH), CD (MeOH) 236 ($\Delta\epsilon$ –1.26); **2a** (2.1 mg): $[\alpha]^{20}_D$ –7.9 (*c* 0.21, MeOH), CD (MeOH) 238 ($\Delta\epsilon$ +1.46); **3a** (1.4 mg): $[\alpha]^{20}_D$ –2.8 (*c* 0.14, MeOH), CD (MeOH) 236 ($\Delta\epsilon$ –0.58); **4a** (1.8 mg): $[\alpha]^{20}_D$ –4.2 (*c* 0.18, MeOH), CD (MeOH) 238 ($\Delta\epsilon$ +0.58); **5a** (7.4 mg): $[\alpha]^{20}_D$ +16.9 (*c* 0.67, MeOH), CD (MeOH) 235 ($\Delta\epsilon$ +2.59); **6a** (3.2 mg); **7a** (2.2 mg): $[\alpha]^{20}_D$ +4.0 (*c* 0.22, MeOH); **8a** (4.0 mg): $[\alpha]^{20}_D$ –19.6 (*c* 0.25, MeOH); **9a** (4.0 mg): $[\alpha]^{20}_D$ –12.4 (*c* 0.33, MeOH); **10a** (2.1 mg): $[\alpha]^{20}_D$ –4.0 (*c* 0.21, MeOH), CD (MeOH) 236 ($\Delta\epsilon$ +0.50); **11a** (3.8 mg): $[\alpha]^{20}_D$ +2.4 (*c* 0.38, MeOH), CD (MeOH) 236 ($\Delta\epsilon$ +0.58); **12a** (4.5 mg): $[\alpha]^{20}_D$ +25.1 (*c* 0.30, MeOH); **13a** (4.5 mg): $[\alpha]^{20}_D$ +17.0 (*c* 0.30, MeOH). $^1H$ NMR and $^{13}C$ NMR data of **1a**−**5a** and **8a**−**13a** in different solvents, see Tables S1−S3, S6, and S11−S13 in the Supporting Information. $^1H$ NMR ($Me_2CO$-$d_6$, 600 MHz) and $^{13}C$ NMR ($Me_2CO$-$d_6$, 150 MHz) data of **6a** and **7a** were identical with that reported in the literature.[20,23]

The aqueous phases of the hydrolysates of **1**−**13** were separately subjected to CC over silica gel eluted with MeCN−$H_2O$ (8:1) to yield glucose with positive optical rotations, and the $[\alpha]^{20}_D$ values ranged from +42.5 to +49.7 (*c* in a range of 0.11 to 0.31, $H_2O$). The solvent system MeCN−$H_2O$ (6:1) was used for TLC identification of glucose ($R_f$, 0.33).

**Preparation of Acetonide Derivatives (1b, 2b, and 5b).** A solution of **1a** (0.8 mg) in dry acetone (1 mL) was treated with 2,2-dimethoxypropane (0.1 mL) and (1*S*)-(+)-camphorsulforic acid (CSA) (1 mg, 0.004 mmol), and the mixture was stirred at ambient temperature for 4 h. The reaction mixture was quenched by addition of triethylamine and then evaporated *in vacuo* to give a crude product. The residue was purified by reversed-phase preparative HPLC using MeOH−$H_2O$ (64:36) to afford acetonide **1b** (0.6 mg). Similarly, **2a** (1.7 mg) and **5a** (2.1 mg) were transformed into acetonide derivatives **2b** (1.5 mg) and **5b** (1.6 mg), respectively. For $^1H$ NMR data (600 MHz) of **1b**, **2b**, and **5b** in $CDCl_3$ see Tables S1 and S2 in the Supporting Information.

**Cell Culture and MTT Assay.** PC12 cells at a density of $5 \times 10^3$ cells per well in 96-well plates were suspended in Dulbecco's modified Eagle's medium (DMEM, Gibico) supplemented with 5% fetal bovine serum (FBS, Hyclone), 5% horse serum, penicillin (100 IU/mL), streptomycin (100 $\mu$g/mL), and L-glutamine (2 $\mu$M) and incubated in a $CO_2$ incubator (5%) at 37 °C for 24 h. Then the cells were pretreated with test compounds ($10^{-5}$ M) and NGF (50 ng/mL), respectively, for another 24 h before exposed to glutamate (20 $\mu$M). After incubation for an additional 24 h, MTT (0.5 mg/mL) was added to the medium and incubated for 4 h. Absorbance was measured at 570 nm using a microplate reader, and the cell viability was evaluated by relative protection, which was calculated as 100 × [optical density (OD) of test compound + glutamate-treated culture − OD of glutamated-treated culture]/[OD of control culture − OD of glutamated-treated culture].[46]

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**Supporting Information Available:** Detailed extraction and isolation procedure including known compounds. Figure S1 for HMBC correlation scheme of compounds **1** and **7**. Figures S2 and S3 of the CD spectra of compounds **1**−**5**, **10**, **11**, **1a**−**5a**, **10a**, and **11a**. 1D NMR spectra of compounds **1**−**9**. Tables S1−S15 of the NMR data for **1a**−**5a**, **8a**−**13a**, **1b**, **2b**, **5b**, **5**, and **8**−**16** in different solvents. This material is available free of charge via the Internet at http://pubs.acs.org.

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