New Lignan Glycosides from Chinese Medicinal Plant, *Sinopodophillum emodi*

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Two new podophillotoxin glucosides, L-picropodophillotpxin 7′-*O*-(β-D-glucopyranosyl-(1→6)-β-D-glucopyranoside) (2) and L-picropodophillotpxin 7′-*O*-β-D-glucopyranoside (3), were isolated from Chinese medicinal plant, *Sinopodophillum emodi*, together with 4 known compounds, podophillotoxin (1), podorhizol 4′-*O*-β-D-glucopyranoside (4), deoxypodophillotoxin (5), and dehydropodophillotoxin (6). The structures of 2 and 3 were finally determined by the extensive decouping and nuclear Overhauser effect (NOE) experiments in NMR spectra and circular dichroism (CD) spectra. Compound 2 is the second example of podophillotoxin diglucoside, and both the first one and 2 were isolated from *S. emodi.* X-ray crystal structure analysis of 1, 5, and 6 was carried out. Compounds 1 and 5 showed the different conformations from those reported.

Key words podophillotoxin; *Sinopodophillum emodi*; picropodophillotoxin diglucoside; crystal structure

Podophillotoxins are a group of important natural products with significant biological properties such as antitumor and antiviral activities. Podophillotoxin (**1**) had already been described by Podwyssotzki in the 1880s.[1] From the middle of the previous century, a number of podophillotoxin derivatives and their glycosides were isolated from the plants of *Podophillum* species.[2,3] The successful chemical modification of a major congener, podophillotoxin (**1**), into the clinically useful anticancer drugs, etoposide and teniposide, has triggered further research in this area.[4]

In the course of our investigation of Chinese medicinal plants, we have reported some new podophillotoxin derivatives from *Sinopodophillum emodi* (WALL.) YING.[5] *S. emodi* is mainly distributed over the western regions of the QinLing Mountains, China, and is used as folk medicine, but had not been previously investigated. As the plant seemed to contain other podophillotoxin derivatives, we continued the investigation. In this paper, we describe the isolation and identification of two new podophillotoxin derivatives (**2**, **3**) together with 4 known ones (**1**, **4**—**6**) from *S. emodi.* The stereochemistry of podophillotoxin congeners (**2**, **3**) was established by the coupling constants in the ^{1}H-NMR spectra, nuclear Overhauser effects (NOE), and the cotton curves in the circular dichroism (CD) spectra.

As **1**, **5**, and **6** were isolated as crystals and the solvents for recrystallization of **1** and **5** were different from the reported solvent,[6,7] X-ray crystal structure analysis of **1**, **5**, and **6** was carried out. We also report the conformations in the crystals of these compounds.

Isolation Dried root and rhizomes of *S. emodi* were extracted with 95% EtOH. The EtOH extract was partitioned between water and Et_2O, and the water layer was further partitioned with $CHCl_3$, AcOEt, and *n*-BuOH. The *n*-BuOH extract was separated on a silica gel column, reversed-phase middle-pressure liquid chromatography (MPLC), Sephadex LH-20 column, and reversed-phase HPLC as described in the Experimental to give compounds **2** (0.028%), **3** (0.0014%), and **4** (0.0056%).

The $CHCl_3$ extract obtained in the partition was chromatographed on silica gel, Sephadex LH-20 column, and reversed-phase HPLC as described also in the Experimental to give compounds **1**,[5,8] **5**,[5,9] and **6** (0.0018%).

Structure Elucidation of the Compounds Compound **2** was obtained as a colorless needle. The molecular formula of **2** was determined as $C_{34}H_{42}O_{18}$ by high-resolution (HR) FAB-MS spectrum. The ^{1}H-NMR spectrum of 2 exhibited methyl signals due to three methoxy groups (δ 3.72, 6H, s and δ 3.90, 3H, s), the signals due to six aromatic protons (δ 6.56, 2H, s, δ 7.06, 2H, s, and δ 8.02, 2H, s), the signals of a methylenedioxy group (δ 6.02, 6.15, each 1H, both d), and other aliphatic proton signals. The ^{13}C-NMR and distortionless enhancement by polarization transfer (DEPT) spectra of **2** showed 9 quaternary carbons including one carbonyl at (δ 178.9), 18 methins and 4 methylenes and 3 methoxy methyl carbons. Out of these 22 methin and methylene carbons, 12 were assignable to the carbons of two sugar moieties. The connection of aliphatic carbons in aglycone was established by correlation spectroscopy (COSY), and the planar structure was determined as podophillotoxin-like structure by 2D-NMR analysis as shown in Chart 1.

Establishment of the relative configurations of the aglycone was tried from the ^{1}H coupling constants (*J* values). The $J_{\text{H-7/H-8}}$, $J_{\text{H-8/H-8'}}$ and $J_{\text{H-8'/H-7'}}$ were 7.7, 9.8, and 10.1 Hz, respectively. These large values seemed to indicate all *trans*, and H-7′/H-8′ was determined as *trans* by comparison with the known *trans* compounds.[10] However, we could not decide the relative configuration of H-8/H-8′ and H-7/H-8, because most H-8/H-8′-*trans* derivatives showed the $J_{\text{H-8/H-8'}}$ values of *ca.* 14 Hz, and some H-7/H-8-*trans* and -*cis* derivatives showed $J_{\text{H-7/H-8}}$ values of *ca.* 5 Hz.[11—13] Thus the configuration have to be confirmed by NOE experiments. Figure 1 shows the NOE observed by the irradiation of each proton at H-7, H-8, H-8′, and H-7′. NOE observed between H-8 and H-8′ indicated that these protons were at *cis* direction and the dihedral angle of H-8–C-8–C-8′–H-8′ was almost 0 deg., which was in good agreement with the $J_{\text{H-8/H-8'}}$ value of 9.8 Hz. NOE observed between H-7 and H-7′ indicated these two protons were at *cis*-direction. Therefore H-8 and H-7

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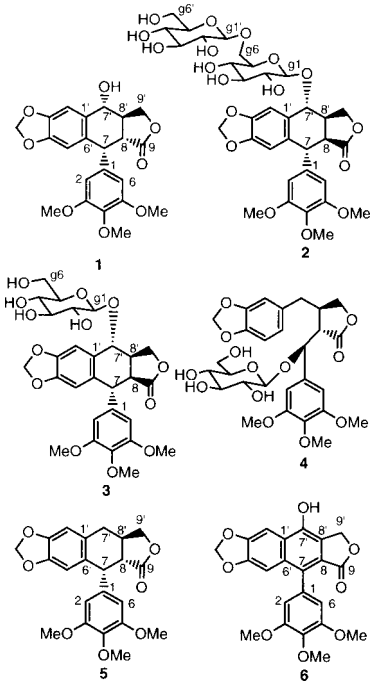

Chart 1

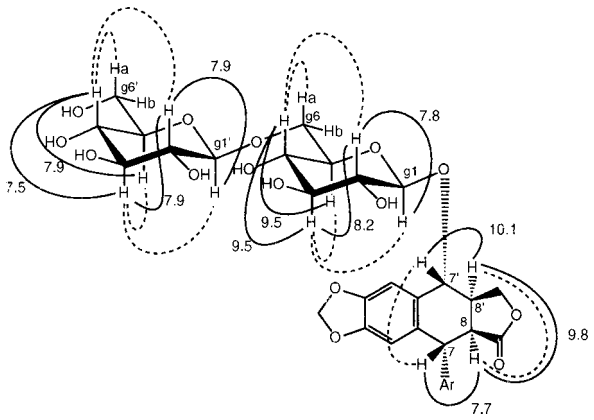

Fig. 1. Selected H–H Coupling Constants and NOEs in **2**

Coupling constants (solid lines, Hz) were obtained by decoupling experiments. NOEs (dotted lines) were detected by NOE difference spectra.

were resulted in *trans*, which agreed with the large $J_{H\text{-}7/H\text{-}8}$ value of 10.1 Hz.

The absolute configuration was established by CD spectra. Swan, Klyne and co-workers reported on the CD spectra of 7-aryltetraline lignans that all 7-α-aryl (L-type) derivatives showed positive Cotton effect around 280—290 nm, while 7-β-aryl (D-type) ones showed negative effect.[14,15] As compound **2** showed positive Cotton effect at 289 nm, C-7 of **2** was determined as α (*R*, L-type). We also measured CD of aglycone of **2**, picropodophyllotoxin, obtained by enzymatic hydrolysis of 2. The aglycone showed also positive Cotton effect at 289 nm and the value was in agreement with the reported value. Thus 7, 8, 8′, and 7′ of **2** were deduced as *R*, *S*, *S*, and *R*, respectively.

The sugar moieties were determined by NMR spectra. Chemical shifts in the ^{13}C-NMR spectrum suggested the sugars were both glucoses. The cross peaks of the anomeric proton at δ 5.13/C-7′ (δ 77.7) and the other anomeric proton at δ 5.03/methylene at δ 69.5 in the HMBC experiment indicated one glucose was at C-7′ of the aglycone and the other was substituted at C-6 of the inner glucose. In order to confirm the structure of the glucoses, NOE difference and ^{1}H decoupling experiments were thoroughly carried out. The NOE were observed in H-g1/H-g3, H-g3/H-g5, H-g4/H-g6, H-g1′/H-g3′, H-g3′/H-g5′ and H-g4′/H-g6′ by irradiation of the corresponding protons. $J_{H\text{-}H}$ values of the sugar protons except for H-g5/H-g6 and H-g5′/H-g6′b were 7.4—9.5 Hz (Fig. 1, Table 1). These large values indicated that the protons at H-g1-5 and H-g1′-5′ were all axial, and the sugars were unambiguously determined as chair-formed β-glucopyranoses (Fig. 1). Thus compound **2** was deduced as a novel diglycoside of podophillotoxins, L-picropodophillotoxin 7′-*O*-(β-D-glucopylanosyl-(1→6)-β-D-glucopylanoside).

Fortunately, the protons of all hydroxy groups were detected in the ^{1}H-NMR spectrum of **2**. All these signals were assigned by heteronuclear multiple bond connectivity (HMBC) correlation. This should be a rare case in that all hydroxy proton signals of the sugar moieties in a glycoside were detected and assigned. H-8/H-8′ *cis* configuration and NOE between H-7 and H-7′ in **2** indicated that its B ring was boat form as conformation A in Fig. 2. However, we could also detect NOE of H-7/H-8, H-7′/H-8′, H-7′/H-2′, and H-7/H-5′, which suggested that compound **2** can also be at the other boat conformation B. These facts indicated that compound **2** was in an equilibrium between the boat forms A and B, as shown in Fig. 2. To confirm the difference in the energy level of these isomers, a computation method was carried out with GAUSSIAN 98[16] on these conformers of the aglycone. The distances between the protons are indicated in Table 2. In form I, the distances of H-7/H-7′ were less than 3Å, which is reasonable to detect NOE. The distances of H-7/H-5′ and H-7′/H-2′ were much more than 3Å, while those in form II were much less than 3Å. The boat form I (=A) was more stable than II (=B) but the difference was small, only 2.18 kcal/mol. Thus, these two conformers were easily interconvertible, and the NOE of H-7/H-7′, H-8/H-8′, H-7/H-8, H-7′/H-8′, H-7′/H-2′, and H-7/H-5′ can be detected at the same time.

Compound **3** obtained as a colorless needle were formulated from the HR-FAB-MS spectrum as $C_{28}H_{32}O_{13}$, which was smaller than **2** by $C_6H_{10}O_5$. The ^{1}H- and ^{13}C-NMR spectra and the cross peaks detected in the 2D-NMR spectra of **3** were extremely similar to that of **2**, except for the disappearance of the signals due to one glucose. The CD spectrum of **3** exhibited positive Cotton effect at 289 nm. As the hydrolysate of **3** was completely identical with that from **2**, the aglycone was determined as picropodophyllotoxin. The sugar moiety was determined as β-glucopyranose by a similar spectral method to **2**. Thus, **3** was determined as a new podophillotoxin congener, L-picropodophillotoxin 7′-*O*-β-D-glucopyranoside.

The FAB-MS spectrum of **4** showed a quasi-molecular ion peak at 539 $[M+H]^+$, and ^{1}H-, ^{13}C- and other 2D-NMR spectra revealed that **4** was the known podorhizol 4′-*O*-β-D-glucopyranoside.[17] The FAB-MS spectrum of 4 showed a quasi-molecular ion peak at 410 $[M]^+$. Compound **6** showed no optical rotation, and all signals in the ^{1}H-NMR spectrum were singlet. By comparison of the ^{1}H- and ^{13}C-NMR spectra with reported data, **6** was deduced as dehydropodophillo-

Table 1. NMR Spectral Data of **2** and **3** (δ ppm, in Pyridine-d_5, ^{13}C: 125 MHz, ^{1}H: 500 MHz)

Position	**2**		**3**	
	^{13}C	^{1}H	^{13}C	^{1}H
1	139.2		139.4	
2,6	107.4	7.06 (2H, s)	107.1	6.90 (2H, s)
3,5	154.2		154.3	
4	137.7		137.9	
7	45.1	4.62 (1H, d, J=7.7 Hz)	44.9	4.31 (1H, d, J=6.9 Hz)
8	45.0	3.80 (1H, dd, J=7.7, 9.8 Hz)	45.4	3.64 (1H, dd, J=6.9, 8.8 Hz)
9	178.9		178.3	
1′	132.9		132.8	
2′	107.2	8.02 (1H, s)	108.1	7.96 (1H, s)
3′	147.0		147.0	
4′	147.6		147.5	
5′	108.6	6.56 (1H, s)	108.4	6.53 (1H, s)
6′	133.7		132.2	
7′	77.7	5.32 (1H, d, J=10.1 Hz)	78.1	5.20 (1H, d, J=8.8 Hz)
8′	43.6	3.14 (1H, m)	42.9	3.20 (1H, m)
9′	69.4	4.56 (1H, dd, J=1.2, 9.8 Hz)	70.2	4.54 (1H, dd, J=6.6, 9.4 Hz)
		5.22 (1H, dd, J=6.4, 9.8 Hz)		5.26 (1H, overlapped with Glc-1)
3,5-MeO	56.1	3.72 (6H, s)	56.2	3.76 (6H, s)
4-MeO	60.6	3.90 (3H, s)	60.6	3.91 (3H, s)
O–CH_2–O	101.5	6.02 (1H, d, J=1.2 Hz)	101.4	5.88 (1H, d, J=1.2 Hz)
		6.15 (1H, d, J=1.2 Hz)		5.90 (1H, d, J=1.2 Hz)
Glc (inner)				
g1	106.0	5.18 (overlapped with HOD)	105.8	5.25 (1H, d, J=7.6 Hz)
g2	75.3	4.16 (1H, m)	75.5	4.18 (1H, m)
g3	78.0	3.72 (1H, m)	78.7	4.30 (1H, m)
g4	71.8	4.08 (1H, m)	71.7	4.30 (1H, m)
g5	78.7	4.17 (1H, m)	78.8	4.04 (1H, m)
g6	69.5	4.76 (1H, dd, J=1.5, 12.4 Hz)	62.9	4.58 (1H, dd, J=2.1, 11.8 Hz)
		4.27 (1H, dd, J=7.8, 12.4 Hz)		4.38 (1H, dd, J=5.4, 11.8 Hz)
g2-OH		7.50 (1H, br-s)		
g3-OH		7.30 (1H, br-s)		
g4-OH		7.38 (1H, br-s)		
Glc (terminal)				
g1′	105.8	5.03 (1H, d, J=7.9 Hz)		
g2′	75.2	3.90 (1H, m)		
g3′	78.0	3.52 (1H, t-like, J=8.9 Hz)		
g4′	71.8	3.95 (1H, m)		
g5′	78.5	4.09 (1H, m)		
g6′	62.8	4.48 (1H, dd, J=2.1, 11.8 Hz)		
		4.31 (1H, dd, J=5.2, 11.8 Hz)		
g2′-OH		6.95 (1H, br-s)		
g3′-OH		6.49 (1H, br-s)		
g4′-OH		6.88 (1H, br-s)		
g6′-OH		6.33 (1H, br-s)		

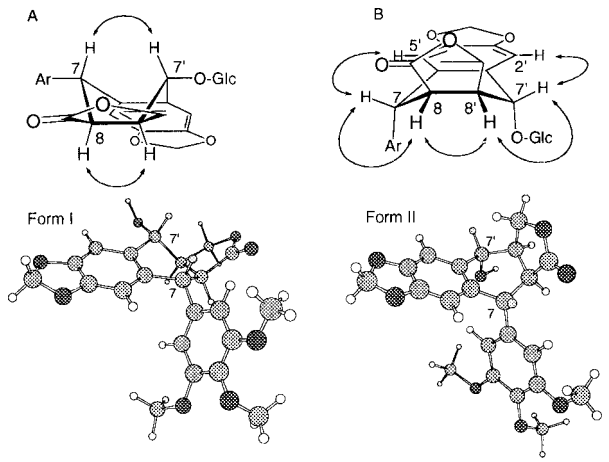

Fig. 2. Conformations at B Ring of **2**

Form A and B represent the conformers of **2**, and Form I and II are the corresponding conformers of the aglycone obtained by GAUSSIAN 98.

Table 2. Atomic Distance and Dihedral Angles of the Conformers I and II

	Form I	Form II
Atomic distance (Å)		
H7–H7′	2.88	4.86
H7–H5′	3.20	2.44
H7–H2 or H6	2.26 (3.77)	2.32 (3.71)
H7–H8	2.93	2.75
H7′–H2′	3.39	2.38
H7′–H8′	3.04	2.54
H5′–H2 or H6	2.83 (3.93)	3.84 (4.08)
H8–H8′	2.40	2.31
Dihedral angles (deg)		
H7′–C7′–C8′–H8′	177	71
H7–C7–C8–H8	133	103
Relative energy (kcal/mol)	0.00	2.18

Table 3. Selected Dihedral Angles in **1**, **5**, and **6** (deg)

Dihedral angles	Podophyllotoxin (**1**)				**5**			**6**
			Reported data[5]			Reported data[6]		
	Form A	Form B	Form A	Form B		Form A	Form B	
C2–C1–C7–C8	78.4	71.0	85.6	73.3	86.5	99.8	79.2	81.8
C6–C1–C7–C8	−100.9	−109.5	−96.3	−106.6	−89.3	−81.7	−100.4	−99.9
C2–C1–C7–C6′	−161.2	−168.5	−153.4	−165.2	−149.6	−136.3	−158.6	−103.5
C6–C1–C7–C6′	19.4	11.1	24.7	15.0	34.0	42.3	21.9	74.8
C1–C7–C8–C8′	68.7	67.6	67.7	71.5	76.4	78.2	67.2	171.9
C1–C7–C8–C9	−54.8	−55.1	−52.8	−53.8	−45.4	−49.1	−53.9	7.2
C7–C8–C8′–C7′	72.3	73.5	72.4	71.8	71.1	70.6	72.1	1.0
C7–C8–C8′–C9′	−155.0	−154.2	−161.5	−163.1	−164.4	−163.9	−161.7	−178.2
C8–C8′–C7′–O	−170.5	−169.7	−176.7	−174.3	*	*	*	−178.7
C9′–C8′–C7′–O	73.3	74.3	67.0	70.1	*	*	*	0.3
C8–C8′–C7′–C1′	−46.9	−46.3	−50.6	−46.8	−45.5	−49.3	−44.7	1.9
C9′–C8′–C7′–C1′	−163.0	−162.3	−166.9	−162.4	−160.4	−163.8	−158.2	−179.1

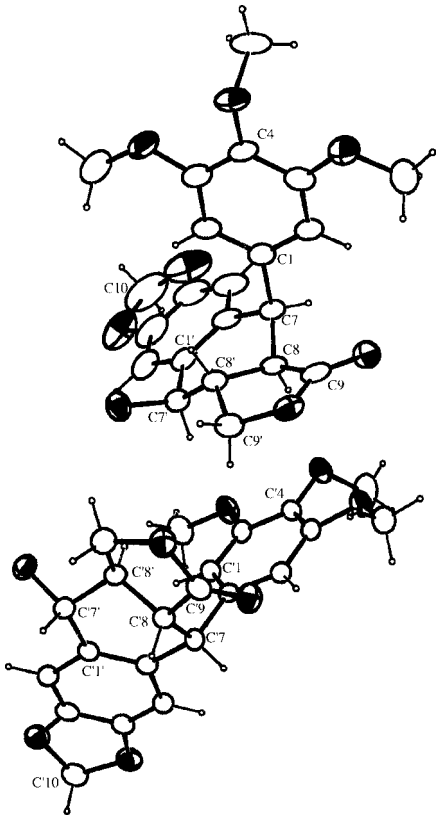

Fig. 3. ORTEP Drawing of Podophillotoxin (**1**)
The solvent molecules are omitted.

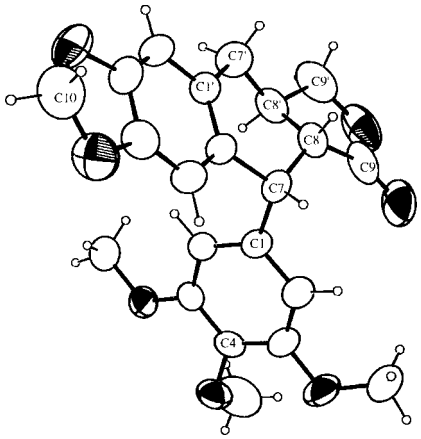

Fig. 4. ORTEP Drawing of **5**

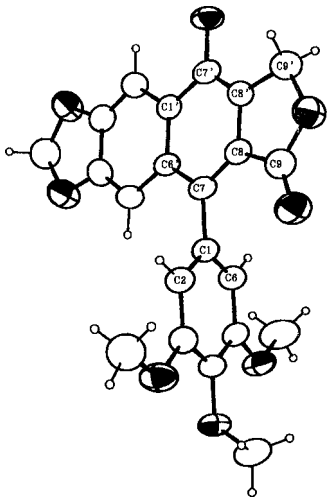

Fig. 5. ORTEP Drawing of **6**
The solvent molecule is omitted.

toxin. The structures of compounds **1** and **5** were also determined by comparison of their spectra and optical rotations with those of reported data.[10,18]

Although the crystal structures of **1** and **5** were reported,[6,7] X-ray structure analysis of **1** and **5** was carried out on the crystals we obtained from other recrystallizing solvents than those reported. Compound **1** was crystallized from MeOH and the crystal contained a pair of different conformational isomers, MeOH and H_2O (Fig. 3). The packing and the conformations of **1** in the crystal were essentially similar to the reported crystals containing nitrobenzene. However, by comparison of the dihedral angles around the B ring of the two conformers, the conformational difference in the pair was smaller than the reported ones[6] (Table 3). In the case of **5**, the reported crystal contained two conformers, while our crystal consisted of one conformer (Fig. 4), which was a rare case for podophillotoxin derivatives. The conformation in the

crystal we obtained was the average of two conformers in the reported crystal.[7)]

The crystal of **6** contained one conformer because of the planar aromatic B ring. However, the angle between the plane of the B ring and the C ring was not exactly 90°. The dihedral angle of C6–C1–C7–C8 (or C2–C1–C7–C6′) was *ca.* 100° and the crystal of **6** contained atrope isomers (Table 3, Fig. 5).

In summary, we have isolated two new lignanes. Compound **2** was the second example of a natural diglycoside of a podophyllotoxin derivative, and both the first one reported by the authors and **2** were isolated from *Sinopodophillum emodi.* This class of compounds may characterize this plant distributed only in the QuinLing Mountains. The biological activities of the compounds are now under investigation.

Experimental

General The FAB-MS and HR FAB-MS were measured with JEOL JMS DX-505 or SX-102 mass spectrometer, and the NMR spectra were measured with a JEOL Lambda-500 or Alpha-500 spectrometer using tetramethylsilane as an internal standard. The following abbreviations are used: s, singlet; d, doublet; t, triplet; dd, doublet-of-doublets; m, multiplet; br, broad. The IR spectra was measured with a JASCO IRA-2 spectrometer. Optical rotations were measured on a JASCO DIP-140 or DIP-1000 digital polarimeter, and CD spectra were on a JASCO J-725 spectrometer. TLC was carried out on precoated plates (Kieselgel 60 F254, 0.25 mm thick, Merck no. 5715), and spots were detected by illumination with an ultraviolet lamp or by spraying 1% $Ce(SO_4)_2$–10% H_2SO_4, followed by heating. Column chromatography was performed on Silica gel BW-200 (Fuji Devison Chemicals Co. Ltd.). MPLC was carried out on Develosil ODS (22 mm i.d.×250 mm, Nomura Chemical) using a Kusano KPW-20. HPLC was carried out on TSKgel ODS-80Ts (20 mm i.d.×250 mm, TOSOH Co.) using a Shimadzu LC-10A pump and SPD-10A UV detector.

Plant Material The rhizomes of *S. emodi* (WALL.) YING were collected from the QuinLing mountain area, China, in 1999, by the author. This plant was botanically identified by Prof. Li Guangming (Biology Department, Northwest University, China).

Extraction and Isolation Dried roots and rhizomes of *S. emodi* (2.47 kg) were extracted three times with 95% EtOH (each time 60 min in 50 °C) and EtOH was removed under reduced pressure to give EtOH extract (240 g). The EtOH extract (40 g) was dissolved in water, and then extracted with Et_2O, $CHCl_3$, AcOEt and *n*-BuOH, followed by removal of the solvents, to give Et_2O extract (6.89 g), $CHCl_3$ extract (1.33 g), AcOEt extract (3.75 g), *n*-BuOH extract (18.6 g), and water extract (12.4 g), respectively. The AcOEt extract (3.75 g) was subjected to silica gel column chromatography and eluted successively by a solvent system of $CHCl_3$ and $CHCl_3$–MeOH (20 : 1—10 : 1) to give five fractions (Fr. A-1—5). Fraction A-2 (1.17 g) was separated by chromatography on a silica gel column and Sephadex LH-20 (Pharmacia Biotech) column with $CHCl_3$–MeOH (16 : 1—8 : 1) and MeOH as the eluant to give **4** (23 mg). The *n*-BuOH extract (18.6 g) was subjected to a silica gel column and eluted with a solvent system of $CHCl_3$–MeOH–H_2O (40 : 1 : 0.1—6 : 4 : 0.5) to give nine fractions (B-l—9) according to their TLC analysis. Fractions B-3 (340 mg) and B-5 (344 mg) were subjected to the Sephadex LH-20 column with MeOH as the eluant to give **1** (214 mg) and **5** (207 mg). Fraction B-4 (310 mg) was isolated by Sephadex column and MeOH as the eluant to give B-4-1 (126 mg) and B-4-2 (187 mg). The B-4-2 was separated by by MPLC (ODS, 30% MeOH) to give B-4-2-1 (49 mg), B-4-2-2 (17 mg), B-4-2-3 (70 mg) and B-4-2-4 (53 mg). Fractions B-4-2-2 and B-4-2-3 were finally purified by preparative HPLC (ODS, 25% MeCN) to give compound **3** (5.7 mg). Fraction B-7 (997 mg) was separated by MPLC (ODS, 40% MeOH) to give three fractions (B-7-1—3). Fraction B-7-1 (210 mg) was separated by chromatography on a Sephadex LH-20 column and eluted with MeOH to give B-7-2-1 (167 mg). B-7-2-1 was separated by chromatography on a Sephadex LH-20 column and eluted with MeOH, and finally purified by preparative HPLC (ODS, 20% MeCN) to give compound **2** (117 mg).

The $CHCl_3$ extract (1.33 g) was subjected to column chromatography on silica gel ($CHCl_3$–MeOH=100 : 0—20 : 1) to give four fractions (Fr. C-1—4). Fraction C-2 (0.42 g) was separated by Sephadex LH-20 column and eluted with MeOH followed by MPLC (ODS, 60% MeOH) to give **6** (7.4 mg).

2: A colorless needle. $[\alpha]_D^{29}$ −46° (c=0.6, MeOH). HR-FAB-MS: m/z 761.2243 $[M+Na]^+$ (Calcd for $C_{34}H_{42}O_{18}Na$: 761.2269). UV: λ_{max} (MeOH) nm: 208, 287. IR (KBr) cm^{-1}: 3550, 3350, 1780. CD (c=0.0050, MeOH), $\Delta\varepsilon$: 290.3 (+1.8), 270 (−2.1), 245.3 (−0.60), 218.3 (−1.1), 202.5 (+12). ^{1}H- and ^{13}C-NMR (pyridine-d_5) Spectral data are given in Table 1.

3: A colorless needle. $[\alpha]_D^{26}$ −91° (c=0.08, EtOH–H_2O=1 : 1). HR-FAB-MS: m/z 585.1566 $[M+Na]^+$ (Calcd for $C_{27}H_{30}O_{13}Na$: 585.1584). UV: λ_{max} (MeOH) nm: 287, 208. IR (KBr) cm^{-1}: 3430, 1770. CD (c=0.0045, MeOH) $\Delta\varepsilon$: 289 (+4.4), 273 (−1.4), 239 (+0.87), 224 (−1.9), 212 (+14). ^{1}H- and ^{13}C-NMR (pyridine-d_5) Spectral data are given in Table 1.

Table 4. Summary of Crystal Data and Intensity Collection Parameters for **1**, **5**, and **6**

	1	**5**	**6**
Formula	$C_{22}H_{22}O_8\cdot H_2O\cdot CH_3OH$	$C_{22}H_{22}O_7$	$C_{22}H_{18}O_8\cdot CH_3OH$
F.W.	464.5	398.4	442.4
Crystal size	0.15×0.18×0.45	0.12×0.18×0.30	0.06×0.18×0.18
Crystal system	Orthorhombic	Orthorhombic	Triclinic
Space group	$P2_12_12_1$	$P2_12_12_1$	P-1
T/°K	293	293	293
a/Å	10.160 (1)	10.580 (1)	8.856 (1)
b/Å	17.295 (1)	11.952 (1)	9.819 (1)
c/Å	26.101 (1)	15.304 (1)	12.796 (1)
α/deg			92.67 (1)
β/deg			108.59 (1)
γ/deg			99.42 (1)
U/Å^3	4586.2 (9)	1935.2 (5)	1034.6 (6)
Z	8	4	2
F(000)	1968	840	464
D_c/g cm^{-3}	1.345	1.367	1.420
μ/cm^{-1}	0.99	0.95	1.03
2θ range/deg	4—52	4—52	4—52
No. of measured data	5228	3964	7200
No. of unique obsd data [$F_0>2.0\sigma(F_0)$]	2103	2339	1923
$R^{a)}$	0.042	0.043	0.047
$R_w{}^{a)}$	0.040	0.047	0.047
No. of variables	587	262	289

a) $R=\Sigma||F_O|-|F_C||/\Sigma|F_O|$. $R_w=[\Sigma_w(|F_O|-|F_C|)^2/\Sigma_w(F_O)^2]^{1/2}$.

Table 5. Positional Parameters and Their Estimated Standard Deviations of **1**

Atom	x	y	z	B_{eq} (Å^2)	Atom	x	y	z	B_{eq} (Å^2)
O1	0.5506 (5)	1.2991 (3)	0.9112 (2)	5.5 (1)	C9′	0.2624 (8)	1.3429 (5)	0.9416 (3)	4.9 (2)
O2	0.1231 (5)	1.3503 (3)	0.9301 (2)	5.0 (1)	C10	0.7489 (9)	1.3432 (5)	0.6800 (3)	5.7 (2)
O3	−0.0067 (6)	1.3661 (3)	0.8621 (2)	5.3 (2)	C20	−0.2006 (8)	1.1797 (5)	0.8164 (3)	5.0 (2)
O4	0.6145 (6)	1.3195 (3)	0.6729 (2)	5.7 (2)	C21	0.017 (1)	0.9771 (5)	0.7481 (5)	9.7 (4)
O5	0.7761 (5)	1.3408 (3)	0.7339 (2)	5.6 (2)	C22	0.4589 (8)	1.0396 (5)	0.7831 (4)	6.2 (3)
O6	−0.1067 (5)	1.1191 (3)	0.8083 (2)	4.6 (1)	C′1	−0.0399 (9)	0.9469 (4)	1.0137 (3)	4.6 (2)
O7	0.0627 (5)	1.0035 (3)	0.7976 (2)	4.9 (1)	C′2	−0.176 (1)	0.9440 (5)	1.0230 (3)	5.5 (3)
O8	0.3207 (5)	1.0291 (3)	0.7891 (2)	5.3 (2)	C′3	−0.219 (1)	0.9452 (5)	1.0734 (4)	5.4 (3)
Ow1	0.7772 (6)	1.3757 (5)	0.9288 (2)	8.2 (2)	C′4	−0.130 (1)	0.9452 (5)	1.1141 (3)	5.0 (2)
O11	0.3177 (6)	1.1109 (3)	0.9685 (2)	6.1 (2)	C′5	0.002 (1)	0.9442 (5)	1.1046 (3)	5.2 (3)
O12	−0.1105 (6)	1.1504 (4)	0.9415 (2)	5.8 (2)	C′6	0.0487 (9)	0.9463 (5)	1.0537 (3)	5.1 (2)
O13	−0.2470 (7)	1.0512 (4)	0.9306 (3)	7.0 (2)	C′7	0.0051 (9)	0.9507 (5)	0.9572 (3)	4.8 (2)
O14	0.355 (1)	0.7504 (5)	0.9466 (4)	12.2 (4)	C′8	−0.0071 (8)	1.0309 (4)	0.9344 (3)	4.4 (2)
O15	0.5233 (9)	0.8407 (6)	0.9341 (3)	10.5 (3)	C′9	−0.136 (1)	1.0744 (6)	0.9352 (3)	5.6 (3)
O16	−0.3485 (7)	0.9473 (4)	1.0879 (2)	7.7 (2)	C′1′	0.249 (1)	0.9803 (5)	0.9430 (3)	4.8 (2)
O17	−0.1760 (7)	0.9498 (3)	1.1645 (2)	6.3 (2)	C′2′	0.178 (1)	0.8455 (6)	0.9524 (3)	7.4 (3)
O18	0.0802 (8)	0.9432 (4)	1.1469 (2)	7.3 (2)	C′3′	0.305 (2)	0.8246 (7)	0.9466 (4)	8.2 (4)
Ow2	0.5296 (6)	1.1419 (4)	0.9108 (3)	7.7 (2)	C′4′	0.403 (1)	0.8773 (8)	0.9392 (4)	7.9 (4)
C1	0.2018 (7)	1.2304 (4)	0.7991 (2)	3.2 (2)	C′5′	0.380 (1)	0.9543 (6)	0.9365 (3)	6.6 (3)
C2	0.0679 (7)	1.2156 (4)	0.8016 (3)	3.4 (2)	C′6′	0.147 (1)	0.9272 (5)	0.9507 (3)	5.1 (3)
C3	0.0208 (7)	1.1399 (4)	0.8023 (3)	3.4 (2)	C′7′	0.2254 (9)	1.0670 (5)	0.9397 (3)	4.6 (2)
C4	0.1091 (8)	1.0785 (4)	0.7970 (3)	3.5 (2)	C′8′	0.0879 (8)	1.0860 (4)	0.9580 (3)	4.0 (2)
C5	0.2430 (8)	1.0932 (4)	0.7933 (3)	3.5 (2)	C′9′	0.031 (1)	1.1642 (5)	0.9448 (3)	5.4 (2)
C6	0.2875 (7)	1.1686 (4)	0.7955 (3)	3.5 (2)	C′10	0.491 (2)	0.759 (1)	0.9345 (6)	13.8 (7)
C7	0.2500 (7)	1.3140 (4)	0.7999 (3)	3.5 (2)	C′20	−0.446 (1)	0.9487 (8)	1.0488 (4)	9.9 (4)
C8	0.2333 (7)	1.3534 (4)	0.8525 (3)	3.4 (2)	C′21	−0.216 (1)	0.8778 (6)	1.1856 (3)	8.9 (3)
C9	0.101 (1)	1.3566 (4)	0.8787 (3)	4.4 (2)	C′22	0.217 (1)	0.9417 (8)	1.1399 (4)	9.4 (4)
C1′	0.4919 (8)	1.3354 (4)	0.8224 (3)	3.4 (2)	Om1[a]	0.734 (4)	0.957 (2)	0.831 (1)	23 (1)*
C2′	0.4298 (8)	1.3165 (4)	0.7338 (3)	4.1 (2)	Om2[a]	0.926 (3)	0.730 (2)	0.841 (1)	19 (1)*
C3′	0.5588 (9)	1.3241 (4)	0.7209 (3)	4.1 (2)	Cm[a]	0.908 (7)	0.746 (3)	0.897 (2)	25 (3)*
C4′	0.6540 (8)	1.3384 (4)	0.7570 (3)	4.4 (2)	Cm2[a]	0.218 (5)	0.617 (3)	0.121 (2)	23 (2)*
C5′	0.6253 (7)	1.3439 (4)	0.8077 (3)	3.8 (2)	Cm3	0.338 (2)	0.576 (1)	0.1601 (7)	16.9 (7)*
C6′	0.3954 (8)	1.3224 (4)	0.7862 (3)	3.4 (2)	Cm1[a]	0.432 (5)	0.706 (2)	0.142 (2)	18 (1)*
C7′	0.4641 (8)	1.3436 (4)	0.8802 (3)	3.9 (2)	Cm4[a]	0.321 (5)	0.732 (3)	0.120 (2)	18 (2)*
C8′	0.3241 (8)	1.3181 (4)	0.8915 (3)	3.8 (2)					

Starred atoms were not anisotropically refined. Anisotropically refined atoms are given in the form of the isotropic equivalent displacement parameter defined as: $(4/3)\times[a^2\times B(1,1)+b^2\times B(2,2)+c^2\times B(3,3)+ab(\cos\gamma)\times B(1,2)+ac(\cos\beta)\times B(1,3)+bc(\cos\alpha)\times B(2,3)]$. *a*) Occupancy of 0.5. * Not anisotropically refined.

Table 6. Positional Parameters and Their Estimated Standard Deviations of **5**

Atom	x	y	z	B_{eq} (Å^2)	Atom	x	y	z	B_{eq} (Å^2)
O1	0.6338 (4)	1.1394 (3)	0.2154 (3)	7.4 (1)	C9	0.5506 (6)	1.1227 (4)	0.1493 (5)	6.2 (2)
O2	0.4958 (4)	1.1973 (3)	0.1134 (3)	7.7 (1)	C1′	0.4489 (4)	0.8145 (3)	0.0964 (2)	3.16 (9)
O3	0.3266 (3)	0.5572 (3)	−0.0078 (2)	6.11 (9)	C2′	0.3658 (4)	0.7495 (4)	0.0467 (3)	3.63 (9)
O4	0.5062 (4)	0.4792 (3)	0.0535 (3)	6.3 (1)	C3′	0.3936 (4)	0.6392 (4)	0.0363 (3)	4.2 (1)
O5	0.1207 (3)	0.9143 (2)	0.3485 (2)	4.39 (7)	C4′	0.5011 (5)	0.5920 (3)	0.0733 (3)	4.4 (1)
O6	−0.0108 (3)	1.0869 (3)	0.2851 (2)	4.85 (8)	C5′	0.5816 (4)	0.6510 (4)	0.1241 (3)	4.3 (1)
O7	0.0783 (3)	1.2075 (3)	0.1503 (2)	5.34 (8)	C6′	0.5552 (4)	0.7667 (3)	0.1368 (3)	3.49 (9)
C1	0.3090 (4)	0.9767 (3)	0.1516 (3)	3.21 (9)	C7′	0.6478 (4)	0.8299 (4)	0.1938 (3)	4.9 (1)
C2	0.2690 (4)	0.9205 (3)	0.2264 (3)	3.23 (9)	C8′	0.5971 (4)	0.9455 (4)	0.2136 (3)	4.3 (1)
C3	0.1660 (3)	0.9605 (3)	0.2724 (3)	3.37 (9)	C9′	0.6919 (5)	1.0335 (5)	0.2436 (4)	6.4 (2)
C4	0.1008 (3)	1.0548 (3)	0.2441 (3)	3.53 (9)	C10	0.4089 (6)	0.4638 (5)	−0.0115 (4)	7.5 (2)
C5	0.1434 (4)	1.1119 (3)	0.1714 (3)	3.9 (1)	C11	0.1893 (4)	0.8216 (4)	0.3833 (3)	4.7 (1)
C6	0.2470 (4)	1.0725 (4)	0.1251 (3)	3.9 (1)	C12	−0.0011 (5)	1.1764 (5)	0.3454 (4)	7.1 (2)
C7	0.4244 (4)	0.9414 (3)	0.0993 (2)	3.43 (9)	C13	0.1374 (5)	1.2841 (4)	0.0919 (3)	5.8 (1)
C8	0.5467 (4)	0.9972 (3)	0.1300 (3)	4.0 (1)					

Anisotropically refined atoms are given in the form of the isotropic equivalent displacement parameter defined as: $(4/3)\times[a^2\times B(1,1)+b^2\times B(2,2)+c^2\times B(3,3)+ab(\cos\gamma)\times B(1,2)+ac(\cos\beta)\times B(1,3)+bc(\cos\alpha)\times B(2,3)]$.

4: A colorless needle. $[\alpha]_D^{26}$ −50° (c=0.20, MeOH). CD (c=0.020, MeOH) $\Delta\varepsilon$: 279 (−1.5), 258 (0.0), 238 (−5.1), 211 (+1.5). ^{1}H-NMR[19] (CD_3OD) δ ppm: 2.25 (1H, dd, J=13.7, 9.1 Hz, 7′-H), 2.60 (1H, dd, J=13.7, 6.7 Hz, 7′-H), 2.69 (1H, dd, J=4.6, 2.7 Hz, 8-H), 2.94 (1H, m, 8′-H), 3.10 (1H, m, g5-H), 3.28 (3H, m, g2, g3, g4-H), 3.62 (1H, dd, J=12.2, 6.4 Hz, g6-H), 3.75 (3H, s, 4-OC$\underline{H}_3$), 3.80 (6H, s, 3,5-OC$\underline{H}_3$), 3.83 (1H, dd, J=12.2, 2.7 Hz, g6-H), 4.06 (1H, dd, J=8.5, 4.0 Hz, 9′-H), 4.20 (1H, d, J=6.7 Hz, g1-H), 4.49 (1H, dd, J=8.5, 7.6 Hz, 9′-H), 5.43 (1H, d, J=2.7 Hz, 7-H), 5.86, 5.89 (each 1H, both d, J=1.5 Hz, OCH_2O), 6.34 (1H, dd, J=7.9, 1.7 Hz, 6′-H), 6.54 (1H, d, J=7.9 Hz, 5′-H), 6.67 (2H, s, 2, 6-H). ^{13}C-

Table 7. Positional Parameters and Their Estimated Standard Deviations of **6**

Atom	x	y	z	B_{eq} (Å^2)	Atom	x	y	z	B_{eq} (Å^2)
Om	0.8858 (4)	−0.2823 (4)	0.6512 (3)	5.8 (1)	C7	0.9021 (5)	0.0927 (4)	1.0887 (3)	3.1 (1)
O1	0.9581 (4)	0.0706 (3)	1.1982 (2)	4.13 (8)	C8	0.9709 (5)	0.1987 (4)	1.0434 (3)	2.9 (1)
O2	1.1290 (4)	0.3904 (3)	1.0043 (2)	4.28 (9)	C9	1.1160 (5)	0.3118 (5)	1.0955 (4)	3.8 (1)
O3	1.0001 (4)	0.3873 (3)	0.8231 (3)	5.2 (1)	C1′	0.7645 (5)	−0.0022 (4)	1.0191 (3)	2.8 (1)
O4	0.4808 (4)	−0.3218 (3)	1.0198 (2)	4.69 (9)	C2′	0.6951 (5)	−0.1169 (4)	1.0644 (3)	3.4 (1)
O5	0.3652 (4)	−0.2921 (3)	0.8350 (2)	4.65 (9)	C3′	0.5651 (5)	−0.2038 (4)	0.9958 (3)	3.3 (1)
O6	0.6948 (4)	0.1273 (3)	0.4585 (2)	4.55 (9)	C4′	0.4946 (5)	−0.1858 (4)	0.8840 (3)	3.3 (1)
O7	0.4337 (3)	0.2446 (3)	0.4267 (2)	3.68 (8)	C5′	0.5553 (5)	−0.0801 (4)	0.8360 (3)	3.1 (1)
O8	0.3297 (4)	0.3080 (3)	0.5914 (2)	4.08 (9)	C6′	0.6951 (5)	0.0166 (4)	0.9043 (3)	2.7 (1)
Cm	0.9561 (8)	−0.3944 (6)	0.6227 (5)	7.0 (2)	C7′	0.7661 (5)	0.1307 (4)	0.8598 (3)	2.9 (1)
C1	0.6842 (5)	0.1609 (4)	0.7440 (3)	2.8 (1)	C8′	0.9039 (5)	0.2158 (4)	0.9308 (3)	2.9 (1)
C2	0.7380 (5)	0.1272 (4)	0.6575 (3)	3.2 (1)	C9′	1.0062 (6)	0.3360 (5)	0.9076 (4)	3.8 (1)
C3	0.6543 (5)	0.1560 (4)	0.5509 (3)	3.1 (1)	C10	0.3481 (6)	−0.3750 (5)	0.9212 (4)	4.6 (1)
C4	0.5204 (5)	0.2186 (4)	0.5325 (3)	2.9 (1)	C11	0.8319 (6)	0.0637 (6)	0.4716 (4)	5.0 (2)
C5	0.4665 (5)	0.2502 (4)	0.6192 (3)	2.9 (1)	C12	0.5138 (6)	0.3594 (5)	0.3863 (4)	5.0 (1)
C6	0.5481 (5)	0.2214 (4)	0.7255 (3)	3.2 (1)	C13	0.2639 (7)	0.3361 (6)	0.6773 (4)	5.9 (2)

Anisotropically refined atoms are given in the form of the isotropic equivalent displacement parameter defined as: $(4/3)\times[a^2\times B(1,1)+b^2\times B(2,2)+c^2\times B(3,3)+ab(\cos\gamma)\times B(1,2)+ac(\cos\beta)\times B(1,3)+bc(\cos\alpha)\times B(2,3)]$.

NMR[19] (CD_3OD) δ ppm: 38.0 (C-8′), 40.5 (7′-C), 53.3 (C-8), 56.6 (3,5-OCH_3), 61.1 (4-OCH_3), 63.0 (C-g6), 71.7 (C-g4), 75.0 (C-9′), 75.4 (C-g2), 78.0 (C-g5), 78.1 (C-g3), 78.3 (C-7), 101.5 (C-g1), 102.3 (OCH_2O), 104.7 (C-2, 6), 108.7 (C-5′), 109.7 (C-2′), 122.8 (C-6′), 133.2 (C-1′), 135.3 (C-1), 138.3 (C-4), 147.6 (C-3′), 149.2 (C-4′), 154.4 (C-3,5), 180.8 (C-9).

Computational Method Calculations were performed with GAUSSIAN 98[16] on the IBM RS-6000SP computer and the SGI Origin 2000 system of Library and Information Processing Center of Nagoya City University. The Becke3LYP hybrid functional was used throughout this work. It consists of the nonlocal exchange functional of Becke's three-parameter set[20] and the nonlocal correlation functional of Lee, Yang, and Parr.[21] All the functionals are applied to the self-consistent-field HF densities. The 6-31G* basis set was used in all calculations.[22] Full geometry optimizations were performed with GAUSSIAN 98 default criteria.

X-Ray Crystal Structure Analysis of 1, 5, and 6 Crystals of all compounds suitable for X-ray analysis were obtained by slow evaporation of the $CHCl_3$–MeOH solution. Crystals were examined on a Nonius Kappa CCD diffractometer using Mo$K\alpha$ radiation. The crystal data and data collection parameters of all compounds are summarized in Table 4. The CCD image intensities were reduced to a set of relative structure factors by the DENZO programs.[23] No absorption correction was made. The structures were solved by the direct method,[24] then refined by the difference Fourier (DF) and least-squares techniques.[25] DF syntheses revealed all non-hydrogen atomic positions of the molecule of all compounds. The non-hydrogen atoms of the molecules were refined with anisotropic thermal parameters, but atoms of disordered solvents were refined isotropically. Hydrogen atoms bound to carbons of the molecules were included in calculated positions as fixed parameters. Final cycles of full-matrix least-squares refinement of the models shown in Figs. 3 and 4 were carried to convergence at $R=0.042$ (**1**) and $R=0.043$ (**5**), respectively, whereas the enantiomers converged at the same value. Thus, the absolute structure could not be determined by the X-ray diffractions; however, other evidences show the correct choice of the models in Figs. 3 and 4. The final DF map showed residual peaks of the 0.2 e/Å^3 level, but those were judged to be essentially featureless. Atomic coordinates, bond lengths, and angles, thermal parameters, and calculated hydrogen atom positions have been submitted as the supplementary supporting data in the format of Cambridge Crystallographic Data Centre. A CIF will be available on request to K.H.

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References and Notes

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