Studies on the Constituents of *Cimicifuga* Species. XVI.[1] Three New Cycloartane Xylosides from the Aerial Parts of *Cimicifuga simplex* WORMSKJORD

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Three new cycloartane xylosides were isolated from the aerial parts of *Cimicifuga simplex*, and their structures were elucidated as 23-*O*-acetyl-1α-hydroxyshengmanol 3-*O*-β-D-xylopyranoside, 25-*O*-acetyl-1α-hydroxycimigenol 3-*O*-β-D-xylopyranoside and 1α-hydroxycimigenol 3-*O*-β-D-xylopyranoside.

Key words *Cimicifuga simplex*; Ranunculaceae; cycloartane; triterpene xyloside; Cellulase

Previously, we reported the isolation and structural characterization of two new triterpenic glucosyl xylosides from the aerial parts of *Cimicifuga simplex*.[1] Further investigation of the glycosides obtained from the same herb has led to the isolation of three new related glycosides (I—III).

The glycoside I was obtained as colorless needles, mp 245—247 °C, $[\alpha]_D^{23}$ −20.0°, $[M]_{290}^{23}$ +13560.0°, and $[M]_{338}^{23}$ −11119.0° (MeOH), and its molecular formula was determined as $C_{37}H_{58}O_{11}$ on the basis of the high resolution (HR) positive FAB-MS ($[M+H]^+$, *m/z* 679.3562 and $[M+Li]^+$, *m/z* 685.3615) and the ^{13}C-NMR spectrum. The IR spectrum showed hydroxyl bands at 3500—3250 cm^{-1}, and acetoxyl and five-membered ketone bands at 1735 cm^{-1}. The ^{1}H-NMR spectrum showed the presence of a cyclopropane methylene group at δ 0.43 and 0.73 (each 1H, d, J=4 Hz, 19-H_2), an acetyl group at δ 2.06, a secondary and six tertiary methyl groups at δ 1.11—1.40 and an anomeric proton at δ 4.86 (1H, d, J=8 Hz).

The ^{1}H- and ^{13}C-NMR signals were assigned by the use of ^{1}H–^{1}H shift correlation spectroscopy (^{1}H–^{1}H COSY) coupled with ^{13}C–^{1}H COSY and by comparison with the data for 23-*O*-acetylshengmanol 3-*O*-β-D-xylopyranoside,[1] indicating the presence of the partial structures shown in Fig. 1.

The heteronuclear multiple bond connectivity (HMBC) spectrum of I clarified the connection of these partial structures shown in Fig. 2. These results are also summarized in Tables I and II.

On hydrolysis with cellulase T[Amano]4, I afforded an aglycone (Ia), mp 110—112 °C, $[\alpha]_D^{20}$ −38.9°. The HR positive electron impact mass spectra (EI-MS) showed the M^+ ion at *m/z* 546.3556 suggesting the molecular formula $C_{32}H_{50}O_7$. The ^{1}H- and ^{13}C-NMR signals were similar to those of I except for the loss of the xylose moiety and the diamagnetic shift of C-3 shown in Tables I and II. The above data suggested that Ia is the genuine aglycone, 23-*O*-acetyl-1α-hydroxyshengmanol.

The second glycoside II was obtained as colorless needles, mp 175—176 °C, $[\alpha]_D^{20}$ +26.2°, and its molecular formula $C_{37}H_{58}O_{11}$ was determined on the basis of the

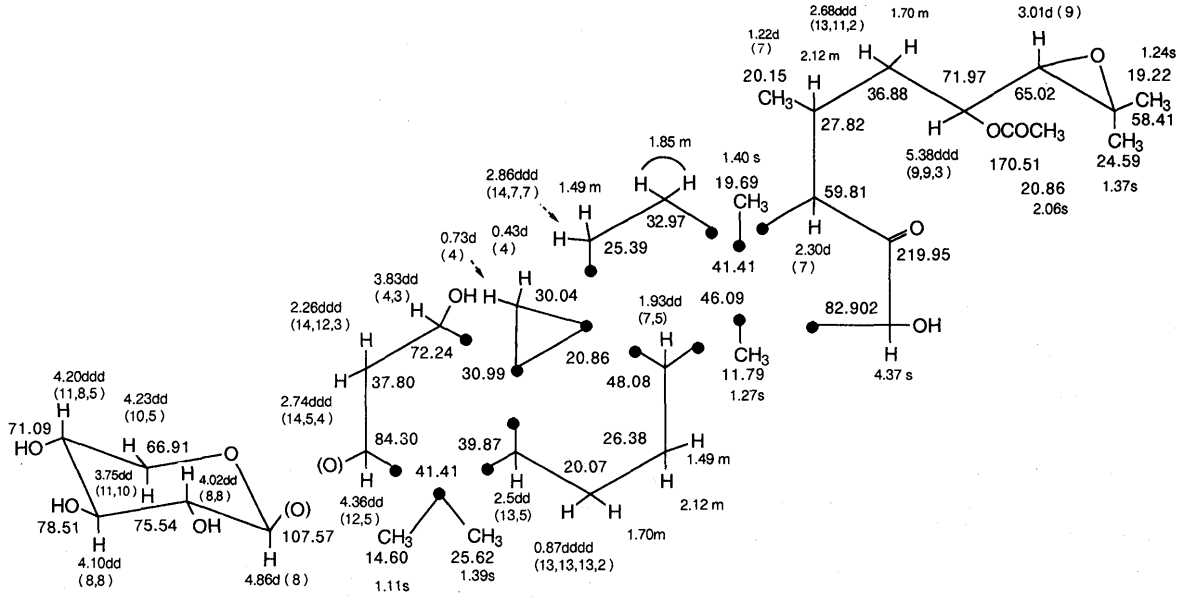

Fig. 1. Partial Structures of I Deduced from the ^{1}H- and ^{13}C-NMR Data

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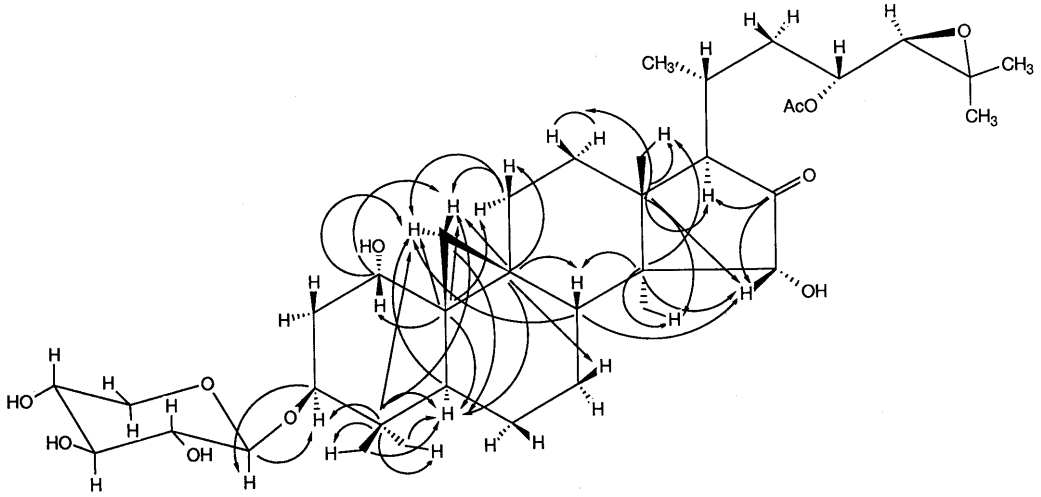

Fig. 2. The Structure I Built up by Connection of the Partial Structures with the HMBC Data

TABLE I. ^{1}H-NMR Chemical Shifts of the Glycosides (I—III) and Their Aglycones (Ia—IIIa) in Pyridine-d_5

	I[a]	Ia[b]	II[c]	IIa[c]	III[c]	IIIa[c]
1	3.83 dd (3, 4)	3.88 dd (3, 3)	3.82 br s	3.84 br s	3.82 br s	3.84 br s
2	2.26 ddd (14, 12, 3)	2.72 dd (14, 12)	2.25, 2.68	2.25, 2.50	2.30, 2.72	2.25, 2.50
	2.74 ddd (14, 5, 4)	2.58 m				
3	4.36 dd (12, 5)	4.43 dd (12, 4)	4.32 dd (12, 4)	4.40 dd (12, 5)	4.32 dd (12, 4)	4.40 dd (12, 4)
5	2.50 dd (13, 5)	2.48 dd (12, 5)	2.47 dd (12, 5)	2.45	2.43	2.41
6	0.87 dddd (13, 13, 13, 2)	0.88 ddd (13, 13, 13)	0.85	0.85, 1.70	0.85, 2.70	0.85, 1.70
	1.70	1.75				
7	1.49, 2.12	1.53, 2.17	1.45, 2.20	1.50, 2.28	1.50, 2.30	1.50, 2.25
8	1.93 dd (7, 5)	1.97 dd (7, 4)	1.73	1.75	1.75	1.75
11	1.49	1.53, 2.94	1.45, 2.88	1.45, 2.91	1.45, 2.87	1.45, 2.90
	2.86 ddd (14, 7, 7)					
12	1.85 (2H)	1.89 (2H)	1.60, 1.75	1.60, 1.75	1.60, 1.75	1.60, 1.78
15	4.37 s	4.42 s	4.29 s	4.32 s	4.30 s	4.32 s
17	2.30 d (7)	2.35 d (7)	1.45 d (11)	1.45 d (11)	1.45 d (11)	1.48 d (11)
18	1.40 s	1.44 s	1.19 s	1.21 s	1.20 s	1.21 s
19	0.43 d (4)	0.52 d (4)	0.43 d (4)	0.48 d (4)	0.42 d (4)	0.47 d (4)
	0.73 d (4)	0.80 d (4)	0.70 d (4)	0.75 d (4)	0.68 d (4)	0.74 d (4)
20	2.12	2.17	1.70	1.70	1.68	1.70
21	1.22 d (7)	1.27 d (7)	0.85 d (7)	0.85 d (7)	0.84 d (7)	0.85 d (7)
22	1.70	1.75	1.05, 2.24	0.98, 2.28	1.05, 2.25	1.03, 2.25
	2.68 ddd (13, 11, 2)	2.70 dd (13, 13)				
23	5.38 ddd (9, 9, 3)	5.40 ddd (9, 9, 3)	4.61 d (9)	4.61 d (9)	4.74 d (9)	4.75 d (9)
24	3.01 d (9)	3.06 d (9)	4.10 s	4.11 s	3.79 s	3.80 s
26	1.24 s	1.28 s	1.66 s	1.67 s	1.47 s	1.48 s
27	1.37 s	1.42 s	1.68 s	1.69 s	1.50 s	1.50 s
28	1.27 s	1.29 s	1.29 s	1.27 s	1.30 s	1.27 s
29	1.39 s	1.33 s	1.38 s	1.30 s	1.39 s	1.31 s
30	1.11 s	1.18 s	1.11 s	1.15 s	1.11 s	1.15 s
$COCH_3$	2.06 s	2.09 s	1.99 s	1.98 s		
1′-H	4.86 d (8)		4.84 d (8)		4.84 d (8)	
2′-H	4.02 dd (8, 8)		4.01 dd (8, 8)		4.02 dd (8, 8)	
3′-H	4.10 dd (8, 8)		4.14 dd (8, 8)		4.12 dd (8, 8)	
4′-H	4.20 ddd (11, 8, 5)		4.20 ddd (11, 8, 4)		4.20 ddd (11, 8, 4)	
5′-H	3.57 dd (11, 10)		3.57 dd (10, 9)		3.57 dd (10, 9)	
5′-H	4.23 dd (10, 5)		4.25 dd (9, 4)		4.25 dd (9, 4)	

a) Measured on a 500 MHz machine. *b*) 400 MHz. *c*) 300 MHz.

elemental analysis, the FAB-MS (*m/z* 679.2 $[M+H]^+$, 701.2 $[M+Na]^+$ and the ^{13}C-NMR spectrum. The ^{1}H-NMR spectrum showed the presence of a cyclopropane methylene at δ 0.43 and 0.70 (each 1H, d, $J=4$ Hz, 19-H_2), a secondary and six tertiary methyl groups at δ 1.11—1.68 and an anomeric proton at δ 4.84 (1H, d, $J=8$ Hz). The ^{1}H- and ^{13}C-NMR signals were assigned with the aid of ^{1}H–^{1}H and ^{13}C–^{1}H COSY spectra and by comparison with those for 25-*O*-acetylcimigenol 3-*O*-β-D-xylopyranoside.[1] The ^{1}H-NMR signals at δ 4.10 (1H, s, 24-H), 4.61 (1H, d, $J=9$ Hz, 23-H), 4.28 (1H, s, 15-H), 1.66, 1.69 (each 3H, s, 26-H, 27-H) and 1.99 (3H, s, an

TABLE II. ^{13}C-NMR Chemical Shifts of I—III and Their Aglycones in Pyridine-d_5

	I[a]	Ia[b]	II[c]	IIa[c]	III[c]	IIIa[c]
1	72.24	72.24	72.44	72.61	72.42	72.58
2	37.80	38.77	37.91	38.80	38.16	38.64
3	84.30	73.23	84.58	73.23	84.60	73.22
4	41.41	41.28	41.48	41.29	41.48	41.22
5	39.87	39.96	39.96	39.94	39.97	39.88
6	20.07	21.16	20.93	21.27	20.93	21.21
7	26.38	26.63	26.25	26.44	26.29	26.39
8	48.08	48.39	48.81	48.91	48.83	48.89
9	20.86	21.03	20.93	20.94	20.93	20.91
10	30.99	31.44	30.83	31.27	30.82	31.18
11	25.39	25.64	25.73	25.84	25.73	25.78
12	32.97	33.23	34.03	34.11	34.06	34.09
13	41.41	41.66	41.78	41.83	41.78	41.79
14	46.09	46.31	47.24	47.32	47.29	47.32
15	82.90	82.99	80.19	80.25	80.23	80.25
16	219.95	220.01	112.40	112.28	111.95	111.96
17	59.81	60.06	59.38	59.46	59.54	59.57
18	19.69	19.92	19.50	19.67	19.59	19.60
19	30.04	30.45	30.93	31.02	30.90	30.97
20	27.82	28.06	23.92	23.99	24.05	24.06
21	20.15	20.38	19.50	19.56	19.59	19.60
22	36.88	37.09	37.70	37.96	37.70	38.16
23	71.97	72.57	71.66	71.71	71.85	71.85
24	65.02	65.27	86.74	86.28	90.17	90.19
25	58.41	58.73	83.24	83.28	71.06	71.06
26	19.22	19.43	21.51	21.51	26.69	26.72
27	24.59	24.77	23.32	23.41	25.29	25.29
28	11.79	11.93	11.69	11.77	11.71	11.75
29	25.62	26.17	25.73	26.23	25.73	26.16
30	14.60	14.12	14.67	14.16	14.67	14.11
$\underline{C}OCH_3$	170.51	170.80	170.39	170.37		
$COC\underline{H}_3$	20.86	21.09	22.34	22.37		
1′	107.57		107.49		107.50	
2′	75.54		75.36		75.36	
3′	78.51		78.27		78.23	
4′	71.09		70.98		70.96	
5′	66.91		66.82		66.82	

Measured at *a*) 125 MHz, *b*) 100.5 MHz, *c*) 75.4 MHz.

acetyl group) and the ^{13}C-NMR signals at δ 112.40 (a quaternary carbon bearing two oxygens, C-16), 80.19, 71.66, 86.74 (methine carbons bearing oxygen, C-15, C-23, C-24), 83.28 (a quaternary carbon bearing oxygen, C-25), 21.51, 23.32 (methyl carbons, C-26, C-27), and 22.34, 170.39 (an acetyl group) were similar to those of 25-*O*-acetylcimigenol derivatives.[1,2,3] The ^{1}H-signals at δ 3.82 (1H, unresolved s, 1-H) and δ 4.32 (1H, dd, J=12, 4 Hz, 3-H) and the ^{13}C-signals at δ 72.44, 84.58 (methine carbons bearing oxygen, C-1, C-3) and δ 37.91 (a methylene carbon, C-2) were assigned to a 1α-hydroxy-3-*O*-moiety as in I.

On hydrolysis with Cellulase T[Amano]4, II afforded an aglycone (IIa), mp 127—128 °C, $[\alpha]_D^{20}$ +36.7°. The HR positive FAB-MS showed $[M+Na]^+$ at m/z 569.3483, suggesting the molecular formula $C_{32}H_{50}O_7$. Based on the ^{1}H- and ^{13}C-NMR spectra (Tables I and II), the structure of IIa was suggested to be 25-*O*-acetyl-1α-hydroxycimigenol.

Compound III was obtained as colorless needles, mp 187—188 °C, $[\alpha]_D^{20}$ +24.6°, and its molecular formula, $C_{35}H_{56}O_{10}$, was determined on the basis of the elemental analysis, the FAB-MS (m/z 637.3 $[M+H]^+$) and the ^{13}C-NMR spectrum. The ^{1}H- and ^{13}C-NMR spectra were analyzed in the same way as those of II and they were similar to those of II except for the loss of an acetyl methyl and the diamagnetic shift of 26,27-H_3 (Δ, 0.19, 0.18 ppm) and 24-H (Δ, 0.31 ppm). The above data suggested that III is a desacetyl derivative of II.[2]

On hydrolysis with Cellulase T[Amano]4, III afforded an aglycone (IIIa), mp 145—146 °C, $[\alpha]_D^{20}$ +50.8°. The HR-MS showed the $[M+Na]^+$ ion at m/z 527.3363, suggesting the molecular formula $C_{30}H_{48}O_6$. Based on the ^{1}H- and ^{13}C-NMR spectra (Tables I and II), the structure of IIIa was suggested to be 1α-hydroxy cimigenol.

Compound I was transformed to II on reflux in 2.5%

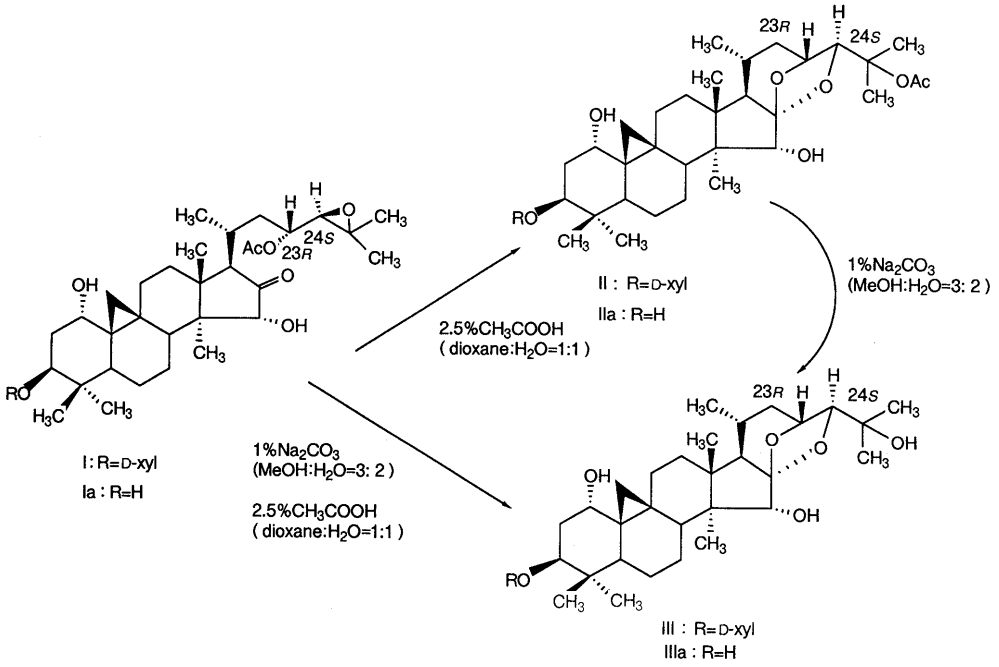

Fig. 3. The Structure of I, Ia, II, IIa, III, IIIa and Transformation of I into II and III

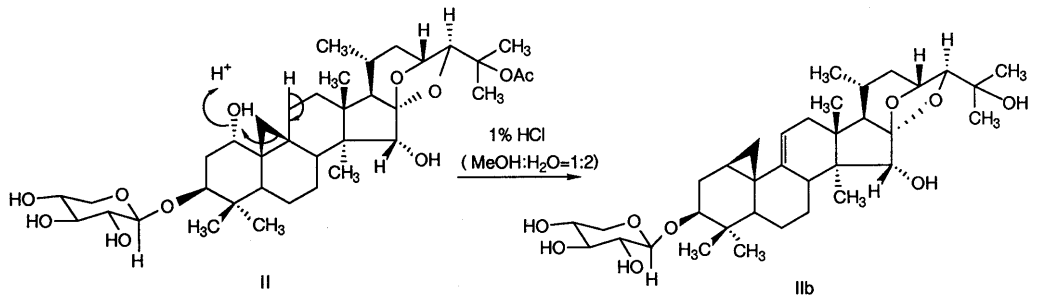

Fig. 4. Chemical Conversion of II into IIb

acetic acid (dioxane : H_2O = 1 : 1) and to III on treatment with 1% sodium carbonate (MeOH : H_2O = 3 : 2) followed by refluxing in 2.5% acetic acid. Compound III was also produced from II on treatment with 1% sodium carbonate. These results established the correlation among I—III and the stereo structure of the side chain (23*R* and 24*S*) of I as shown in Fig. 3.[2)]

D-Xylose was identified on the basis of isolation by HPLC, identification from the ^{1}H-NMR spectrum and $[\alpha]_D^{20}$ values (+38.1° from I, +23.6° from II, +17.4° from III) of the sugar on hydrolysis of I, II, and III with 1% hydrochloric acid (H_2O : MeOH = 2 : 1). The structure of the 3-*O*-*β*-D-xylopyranosyl moiety of I, II and III was established on the basis of the ^{1}H- and ^{13}C-NMR spectra (1′-H: *δ* 4.84—4.86, d, *J*=8 Hz; 2′-H: *δ* 4.01—4.02, dd, *J*=8, 8 Hz; 3'-H: *δ* 4.10—4.14, dd, *J*=8, 8 Hz; 4′-H: *δ* 4.20, ddd, *J*=11, 8, 4—5 Hz; 5′-H_2: *δ* 3.57, dd, *J*=10—11, 9—10 Hz; *δ* 4.23—4.25, dd, *J*=9—10, 4—5 Hz. C-1′: *δ* 107.49—107.57; C-2′: *δ* 75.36—75.54; C-3′: *δ* 78.23—78.51; C-4′; *δ* 70.96—71.09; C-5′: *δ* 66.82—66.91; C-3: *δ* 84.30—84.60; glycosylation shift: 11.07—11.38 ppm) as shown in Tables I and II. This was also supported by the nuclear Overhauser effect (NOE) between 3-H (*δ* 4.36) and the anomeric proton (*δ* 4.86) in the NOE difference spectrum of I and the cross peaks between 3-H and C-1′ and C-3 and 1′-H in the HMBC spectrum of I.

The 1α-hydroxy group was eliminated easily and the rearranged compound IIb was obtained on treatment of II with 1% hydrochloric acid, as shown in Fig. 4. A concerted elimination-rearrangement mechanism from 1α-hydroxy-9,19-cyclolanostane glycosides to 1,19-cyclolanost-9(11)-ene derivatives has been proposed by Pegel and Rogers[4)] and the characteristic ^{1}H-NMR signals due to the 1,19-cyclopropane methylene group and 11-olefinic proton were found in the spectrum of IIb. The production of IIb from II is additional evidence in favor of the 3-*O*-xylosyl structure in the glycosides I,II and III.

Thus, the first glycoside (I) should be formulated as 23-*O*-acetyl-1α-hydroxyshengmanol 3-*O*-*β*-D-xylopyranoside, the second (II) as 25-*O*-acetyl-1α-hydroxycimigenol 3-*O*-*β*-D-xylopyranoside, and the third (III) as 1α-hydroxycimigenol 3-*O*-*β*-D-xylopyranoside, as shown in Fig. 3.

Experimental

General The instruments used in this work were as follows: Yanagimoto micro melting apparatus (melting points), JASCO DIP digital polarimeter (specific rotation), JASCO ORD/UV-5 spectrometer (optical rotatory dispersion (ORD)), Perkin–Elmer 1720X-FT IR spectrometer (IR spectra), Varian Gemini-200, Varian XL-300, General Electric GN-300, GN-400 and GN-500 spectrometers (NMR spectra), and Hitachi M-400H and JEOL JMS-DX-300 spectrometers (mass spectra). Melting points are uncorrected. NMR spectra were measured in pyridine-d_5, and chemical shifts are expressed on the *δ* scale using tetramethyl silane as an internal standard. Column chromatography was carried out on silica gel (Wakogel C-200) and ODS-A YMC. HPLC was conducted on a Gilson 305 pump with a Shodex refractometer as a detector.

Extraction and Isolation of I, II and III The dried aerial parts (6.1 kg) of *Cimicifuga simplex*, which were collected in August 1991 in Sendai, were extracted with MeOH (80 l × 3) at room temperature overnight. The MeOH solution was subjected to activated charcoal column chromatography (250 g, 6.7 cm i.d. × 37 cm) and the pass-through fraction was concentrated *in vacuo* to yield a gummy extract (500 g). The concentrated extract was shaken with *n*-BuOH (300 ml) three times and the combined *n*-BuOH layer was washed with water (200 ml) twice, then concentrated *in vacuo* (fraction A). The water layer and water washing were combined and concentrated *in vacuo* (fraction B).

The *n*-BuOH extract (fraction A, 270 g) was chromatographed on SiO_2 and octadecyl silica (ODS)-A as described under the isolation of 7*β*-hydroxy-23-*O*-acetylshengmanol 3-*O*-*β*-D-xylopyranoside.[2)] The residue after the removal of the above compound afforded I (120 mg) as colorless needles after recrystallization from MeOH. I showed mp 245—247 °C, $[\alpha]_D^{23}$ −20.0° (*c*=0.50, MeOH), ORD: $[M]_{290}^{23}$ +13560.0°, $[M]_{338}^{23}$ −11119.0° (*c*=0.50, MeOH). The HR positive FAB-MS *m/z*: Calcd for $C_{37}H_{58}O_{11}$: 678.4057 ([M+H]$^+$), 685.4139 ([M+Li]$^+$. Found: *m/z* 679.3562 ([M+H]$^+$), 685.3615 ([M+Li]$^+$). IR (KBr): 3500—3250 (OH), 1735 (AcO- and five membered ketone) cm^{-1}. ^{1}H- and ^{13}C-NMR: Tables I and II.

The aqueous fraction (fraction B) was chromatographed on a Diaion HP-20 column (6.5 cm i.d. × 36 cm) and after washing with water, elution with MeOH afforded a gummy material. On storage at room temperature, this afforded precipitates (23-*O*-acetylshengmanol 3-*O*-xyloside, 3-*O*-glucosyl xyloside, 25-*O*-acetylcimigenol 3-*O*-xyloside, 3-*O*-glucosyl xyloside).[1)] The residue (185 g) after the concentration of the mother solution was chromatographed on SiO_2 (1 kg, 6.5 cm i.d. × 40 cm) and fractions eluted with $CHCl_3$–MeOH (10 : 1) and (9 : 1) were rechromatographed on ODS. Elution with MeOH–H_2O (2 : 1) afforded I (5 mg), and elution with MeOH–H_2O (4 : 1) afforded II (174 mg). The latter was purified by HPLC [column, CrestPak C18T-5 (5*μ*,i.d. 5 mm × 250 mm); solvent, MeOH–H_2O–CH_3CN (10 : 7 : 3); rate, 1 ml/min; column temperature, 40 °C, t_R, 18 min] followed by recrystallization from a mixture of MeOH and isopropyl ether. The fractions eluted with $CHCl_3$–MeOH (7 : 1) in the above SiO_2 chromatography were rechromatographed on the ODS column and elution with MeOH–H_2O (4 : 1) afforded III (45 mg). This was purified by HPLC (t_R, 10 min) as in the case of II.

II: Colorless needles, mp 175—176 °C, $[\alpha]_D^{20}$ +26.2° (*c*=0.97, MeOH). *Anal.* Calcd for $C_{37}H_{58}O_{11}\cdot H_2O$: C, 64.15; H, 8.68. Found: C, 64.15; H, 8.78. FAB-MS *m/z*: 679.2 ([M+H]$^+$), 701.2 ([M+Na]$^+$). IR (KBr): 3520—3200 (OH), 1736 (AcO) cm^{-1}. ^{1}H- and ^{13}C-NMR: Tables I and II.

III: Colorless needles, mp 187—188 °C, $[\alpha]_D^{20}$ +24.6° (*c*=0.68, MeOH). *Anal.* Calcd for $C_{35}H_{56}O_{10}\cdot 2H_2O$ C, 63.99; H, 9.21. Found: C, 63.33; H, 9.12. FAB-MS *m/z*: 637.3 ([M+H]$^+$). IR (KBr): 3580—3200 (OH) cm^{-1}. ^{1}H- and ^{13}C-NMR: Tables I and II.

Hydrolysis of I with Cellulase I (20 mg) was dissolved in 1% ethanolic AcOH (20 ml), then water (40 ml) was added with stirring and the solution

was adjusted to pH 4.5 by the dropwise addition of AcOH. Cellulase T[Amano]4 (from *Trichoderma viride*, 200 mg) was added. The solution was stirred for 4 d at room temperature, then shaken with EtOAc (20 ml × 3). The combined EtOAc layer was washed with water, dried over Na_2SO_4, and evaporated *in vacuo*. Recrystallization from a mixture of MeOH and isopropyl ether afforded Ia (5 mg) as colorless needles, mp 110—112 °C. $[\alpha]_D^{20}$ −38.9° (c=0.18, MeOH). HR-positive EI-MS m/z: Calcd for $C_{32}H_{50}O_7$: 546.3554 ($[M]^+$). Found m/z: 546.3556 ($[M]^+$). ^{1}H- and ^{13}C-NMR (pyridine-d_5): Tables I and II.

Hydrolysis of II with Cellulase II (50 mg) was treated as mentioned above and the product was purified by recrystallization from EtOAc to yield IIa (15 mg), mp 127—128 °C, $[\alpha]_D^{20}$ +36.7° (c=0.35, MeOH). IR (KBr): 3500—3200 (OH), 1739 (AcO) cm^{-1}. Calcd for $C_{32}H_{50}NaO_7$ m/z: 569.3505 ($[M+Na]^+$). Found: HR-FAB-MS m/z: 569.3483 ($[M+Na]^+$). ^{1}H- and ^{13}C-NMR (pyridine-d_5): Tables I and II.

Hydrolysis of III with Cellulase III (21.6 mg) was treated as mentioned above and the product was purified by recrystallization from EtOAc to yield IIIa (10.5 mg), mp 145—146 °C, $[\alpha]_D^{20}$ +50.8° (c=0.78, MeOH). IR (KBr): 3440—3200 (OH) cm^{-1}. Calcd for $C_{30}H_{48}NaO_6$ m/z: 527.3349 ($[M+Na]^+$). Found: HR-FAB-MS m/z: 527.3367 ($[M+Na]^+$).

Transformation of I to II I (20 mg) was stirred in 2.5% AcOH (dioxane : H_2O = 1 : 1, 2.0 ml) on a hot water bath for 5 h, then the reaction solution was shaken with EtOAc (20 ml × 3). After evaporation of the EtOAc extract, the residue was chromatographed on SiO_2 (5 g). Elution with $CHCl_3$–MeOH (4 : 1) afforded II (5 mg), which was identified by comparison of TLC behavior and ^{1}H-NMR spectra with those of the isolated glycoside II.

Transformation of I and II into III I (10 mg) was stirred in 1% Na_2CO_3 (MeOH : H_2O = 3 : 2, 5.0 ml) for 3 h at room temperature and after evaporation of the MeOH, the residue was extracted with EtOAc (20 ml × 3). The extract was dissolved in 2.5% AcOH (dioxane : H_2O = 1 : 1, 2.0 ml) and heated on a boiling water bath for 3 h, then shaken with EtOAc (20 ml × 3). The residue after evaporation of the solvent was chromatographed on SiO_2 (5 g). Elution with EtOAc–MeOH (2 : 1) yielded III (5 mg), which was purified by HPLC as above and identified by comparison of its ^{1}H-NMR spectra with that of the isolated glycoside III. II (25.6 mg) was dissolved in 1% Na_2CO_3 (MeOH : H_2O = 3 : 2, 5.0 ml) and the solution was stirred for 3 d at room temperature, then shaken with EtOAc (20 ml × 3). The products were chromatographed on SiO_2 (5 g). Elution with $CHCl_3$–MeOH (4 : 1) yielded III (15.0 mg) and the ^{1}H-NMR spectra was identical with that of the isolated glycoside III.

Hydrolysis of I, II and III with 0.5 N HCl I (10 mg) was dissolved in 0.5 N HCl (MeOH : H_2O = 1 : 2, 3.0 ml) and the solution was refluxed in a water bath for 2 h. After it had cooled , the solution was shaken with EtOAc (10 ml × 2). The EtOAc layer was washed with water (10 ml × 2), and the water layer and water washings were combined. After removal of the MeOH by evaporation, the residual solution was heated on a boiling water bath for 1 h, then chromatographed on an Amberlite IR-35 column. Elution with water afforded D-xylose, $[\alpha]_D^{20}$ +38.1° (c=0.25, 50% MeOH), which was identified by HPLC [column: LiChrosorb NH_2 (5 μ, i.d. 4.6 mm × 25 mm); solvent, CH_3CN–H_2O (4 : 1); rate, 1 ml/min; column temperature, 40 °C; t_R, 5.50 min]. Similarly, D-xylose of $[\alpha]_D^{20}$ +23.6° (c=0.33, 50% MeOH) was obtained from II and D-xylose of $[\alpha]_D^{20}$ +17.4° (c=0.20, 50% MeOH) from III.

Transformation of II to IIb with 1% HCl II (20 mg) was dissolved in 1% HCl (MeOH : H_2O = 1 : 2, 3.0 ml) and the solution was refluxed for 1 h. It was concentrated to half the initial volume *in vacuo* and shaken with EtOAc (30 ml × 3). The EtOAc layer was chromatographed on SiO_2 (10 g) and elution with $CHCl_3$–MeOH (4 : 1) afforded IIb (7 mg), mp over 300 °C. $C_{35}H_{54}O_9$. FAB-MS m/z: 619.2 ($[M+H]^+$), 641.2 ($[M+Na]^+$). ^{1}H-NMR (pyridine-d_5) δ: −0.04 (1H, dd, J=4, 7 Hz, 19-H), 0.85 (3H, d, J=7 Hz, 21-H), 1.54 (3H, s, 26-H), 1.60 (3H, s, 27-H), 3.50 (1H, dd, J=10, 5 Hz, 3-H), 3.58 (1H, s, 24-H), 4.00 (1H, dd, J=6, 6 Hz, 5′-H), 4.35 (1H, s, 15-H), 4.70 (1H, d, J=9 Hz, 23-H), 4.77 (1H, d, J=7 Hz, 1′-H), 5.12 (1H, d, J=5 Hz, 11-H).

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