Studies on the Constituents of *Clematis* Species. VIII.[1] Triterpenoid Saponins from the Aerial Part of *Clematis tibetana* KUNTZ[2]

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From the aerial part of *Clematis tibetana*, two new hederagenin 3,28-*O*-bisdesmosides called clematibetosides A and C, and a new gypsogenin 3,28-*O*-bisdesmoside called clematibetoside B, have been isolated together with ten known saponins. The structures of the new saponins have been elucidated based on chemical and spectral evidence as follows: clematibetoside A, 3-*O*-(2-*O*-caffeoyl)-β-D-glucopyranosyl-(1→4)-β-D-glucopyranosyl-(1→4)-β-D-ribopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl hederagenin 28-*O*-α-L-rhamnopyranosyl-(1→4)-β-D-glucopyranosyl-(1→6)-β-D-glucopyranoside; clematibetoside B, 3-*O*-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl gypsogenin 28-*O*-α-L-rhamnopyranosyl-(1→4)-β-D-glucopyranosyl-(1→6)-β-D-glucopyranoside; clematibetoside C, 3-*O*-β-D-ribopyranosyl hederagenin 28-*O*-α-L-rhamnopyranosyl-(1→4)-β-D-glucopyranosyl-(1→6)-β-D-glucopyranoside.

Key words *Clematis tibetana*; clematibetoside; saponin; hederagenin bisdesmoside; gypsogenin bisdesmoside; Ranunculaceae

As a continuation of our study on the constituents from *Clematis* species,[1] the dried aerial part of *C. tibetana* KUNTZ that was collected in central Nepal has been investigated. The water-soluble portion of a hot MeOH extract was successively extracted with hexane, EtOAc, and *n*-BuOH. The *n*-BuOH-soluble fraction was subjected to repeated chromatography to give thirteen compounds (**1**—**13**), including three new ones, named clematibetosides A (**1**), B (**6**) and C (**12**) as described in the experimental section.

Compounds **2**, **3**, **4**, **5**, **7**, **8**, **9**, **10**, **11** and **13** were identified as huzhangoside D,[3] hederasaponin C,[4] CP_6,[5] α-hederin,[6] CP_{10},[7] hederasaponin B,[4] HN saponin H,[8] kizutasaponin K_{10},[9] CP_8[3] and dipsacoside B,[9,10] respectively, by direct comparison with the respective authentic samples.

Clematibetoside A (**1**) was obtained as a yellow amorphous powder and gave hederagenin, glucose, rhamnose, arabinose and ribose on acid hydrolysis. The ^{13}C-NMR spectrum of **1** showed eight anomeric carbon signals together with signals assignable to a 3,4-dihydroxycinnamoyl (caffeoyl) group. The ^{1}H-NMR and UV spectra of **1** also supported the presence of a caffeoyl group. The molecular formula of **1** was determined as $C_{85}H_{130}O_{43}$ from high resolution (HR) FAB-MS and ^{13}C-NMR spectral data. Compound **1** was hydrolyzed with mild alkali, 0.1 N KOH aq., at room temperature to give a deacylated compound (**1a**) and caffeic acid. Compound **1a** was identified as the deacylated compound of clematernoside C by direct comparison.[1] Therefore, **1** was a monocaffeate of deacylated clematernoside C. The assignments of proton and carbon signals due to the sugar moiety of **1** and **1a** were determined by comparison with those of the clematernoside group[1] and were confirmed based on the ^{1}H–^{1}H correlation spectroscopy (COSY) and ^{1}H–^{13}C COSY spectral data. In comparison of the ^{1}H- and ^{13}C-NMR data for **1** with those for **1a**, the H-2 and C-2 signals of the terminal glucose moiety (Glc^4) in **1** were observed at a lower field by 1.68 and 0.3 ppm, respectively, and the C-1 and C-3 signals of Glc^4 at a higher field by 2.3 and 1.8 ppm, respectively, than the corresponding signals in **1a** (Table 1). These results show that the caffeoyl group in **1** is connected to the C-2 position of the terminal glucose moiety. Furthermore, the ^{13}C-NMR signals due to the terminal glucose unit of **1** were found at almost the same positions with those in clematernoside C. From these facts, the structure of clematibetoside A (**1**) was concluded to be 3-*O*-(2-*O*-caffeoyl)-β-D-glucopyranosyl-(1→4)-β-D-glucopyranosyl-(1→4)-β-D-ribopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl hederagenin 28-*O*-α-L-rhamnopyranosyl-(1→4)-β-D-glucopyranosyl-(1→6)-β-D-glucopyranoside.

Clematibetoside B (**6**), $C_{59}H_{94}O_{26}$, showed signals due to a formyl group [δ 9.96 (d, J=2 Hz) in ^{1}H-NMR and δ 207.6 (d) in ^{13}C-NMR] and five anomeric carbon signals in its ^{13}C-NMR spectrum. **6** gave gypsogenin,[11] glucose, rhamnose and arabinose on acid hydrolysis; moreover the proton and carbon signals due to the sugar moieties in **6** were observed at almost the same positions as those in **3** and **8**. Therefore, **6** was concluded to be a gypsogenin bisdesmoside possessing the same sugar moieties as **3** and **8**.

Clematibetoside C (**12**), $C_{53}H_{86}O_{22}$, gave hederagenin, glucose, rhamnose and ribose on acid hydrolysis. Comparison of the ^{1}H- and ^{13}C-NMR data for **12** with those for **2** and **9**, suggested that **12** possessed an α-L-rhamnopyranosyl-(1→4)-β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyloxy group at the C-28 position and a ribopyranosyloxy group at C-3. (Tables 1, 2) In order to clarify the anomeric configuration of the ribopyranosyl unit, **12** was hydrolyzed by alkaline. In the ^{1}H-NMR spectrum of the resulting prosapogenin (**12a**), the coupling constants $J_{H\text{-}1,H\text{-}2}$, $J_{H\text{-}4,H\text{-}5}$ and $J_{H\text{-}4,H\text{-}5'}$ in the ribopyranosyl unit were observed as 4, 3 and 5 Hz, respectively. These data were in agreement with those of methyl β-D-ribopyranoside (Table 3).[12] In addition, the carbon signals assignable to the ribopyranosyl unit in **12a** were observed at almost the same positions as methyl β-D-ribopyranoside (Table 3). Based on these results, the structure of clematibetoside C (**12**) was concluded to be 3-*O*-β-D-ribopyranosyl hederagenin 28-*O*-α-L-rhamnopyranosyl-(1→4)-β-D-glucopyranosyl-(1→6)-β-D-glucopyranoside (Chart 1).

Experimental

General Procedures NMR spectra were taken in pyridine-d_5 on a JEOL GSX-400 spectrometer (^{1}H-NMR at 400 MHz and ^{13}C-NMR at 100 MHz), using the residual signals of the solvent as an internal standard: pyridine-d_5,

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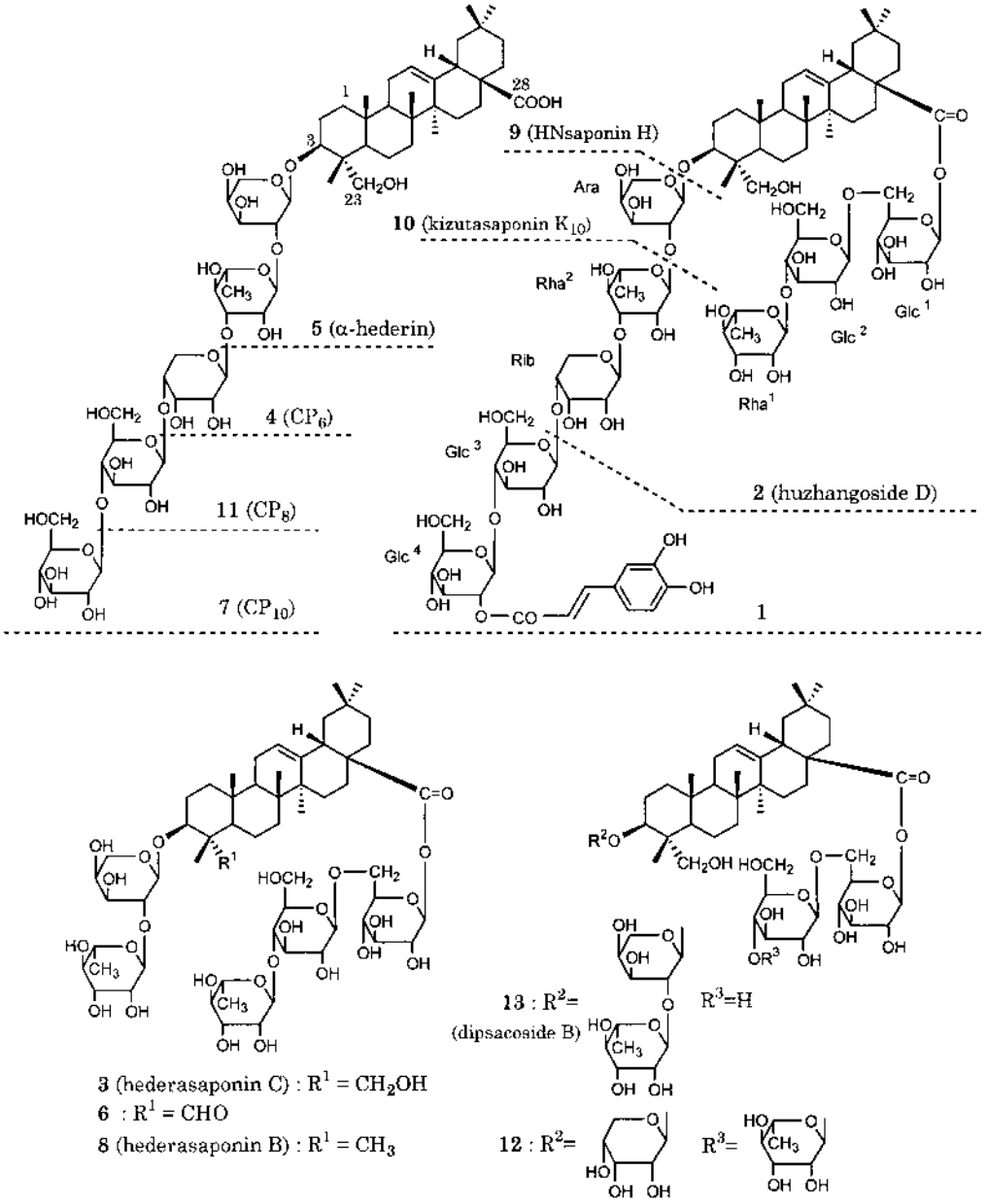

Chart 1

δ_C 123.5, δ_H 7.20 (β-OH). MS were taken on a JEOL JMS-SX-102A mass spectrometer, using triethanolamine as a matrix. UV spectra were taken in MeOH on a Shimadzu dual-wavelength/doublebeam recording spectrophotometer. Samples for IR spectra were prepared as a KBr disk and the spectra were taken on a HORIBA FT-720 FT-IR spectrophotometer. Optical rotation was measured by a JASCO DIP-370 digital polarimeter. The HPLC system was composed of TOSO CCPE pump with recycling valve and a JASCO 875 UV detector. For TLC, pre-coated plates of Silica gel $60F_{254}$, RP-18 and HP Silica gel $60F_{254}$ (Merck) were used.

Medium pressure liquid chromatography (MPLC) was conducted on an octadecyl silica (ODS) column [stuffed Cosmosil $140C_{18}$OPN in 500 mm×45 mm i.d., mobile phase, MeOH–propanol–H_2O (5 : 1 : 6) (MPLC-1); stuffed Cosmosil $40C_{18}$OPN in 500 mm×32 mm i.d., mobile phase, CH_3CN–propanol–H_2O (2.5 : 1 : 7.5) (MPLC-2)]. Purification by preparative recycling HPLC was carried out under the following conditions: column A, COSMOSIL $5C_{18}$-AR (250 mm×20 mm i.d.); column B, YMC-Pack Polyamine-II (250 mm×20 mm i.d.); mobile phase, CH_3CN : propanol : H_2O=2.3 : 1 : 7.7 (solv. 1), 2.4 : 1 : 7.6 (solv. 2), 2.4 : 1.2 : 7.6 (solv. 3), 2.5 : 1.5 : 7.5 (solv. 4), 2.6 : 1 : 7.4 (solv. 5), 2 : 3 : 6 (solv. 6), 6 : 1 : 2 (solv. 7); detection, UV 210 nm.

Materials The aerial part of *C. tibetana* Kuntz was collected in central Nepal in August. A voucher specimen is deposited at the Herbarium of the Faculty of Pharmaceutical Sciences, Hokuriku University, Kanazawa, Japan.

Extraction and Isolation The dried aerial part (6.1 kg) of the plant was extracted by boiling with MeOH. The MeOH extract was concentrated to dryness under reduced pressure. The residue (1150 g) was suspended in H_2O and successively extracted with hexane, EtOAc, and *n*-BuOH. The *n*-BuOH layer was concentrated and the residue was dissolved in a small amount of MeOH. This solution was poured into EtOAc and the resulting precipitate (80 g) was collected. The precipitate was subjected to silica gel column chromatography eluting with $CHCl_3$–MeOH (100 : 2→100 : 4→100 : 6→100 : 8) and $CHCl_3$–MeOH–H_2O (25 : 3 : 0.3→25 : 5 : 0.5→25 : 7 : 0.9→25 : 10 : 1.8→25 : 12 : 2.5→25 : 14 : 3→25 : 16 : 4) to give sixteen fractions.

Fraction 13 (6.5 g) was separated by MPLC-1 and MPLC-2 to give crude **1** (1.8 g), which was purified by preparative recycling HPLC (column A, solv. 1) to give pure **1**. By MPLC-1 and MPLC-2, fraction 12 (3.7 g) gave crude **2** (1.5 g) which was purified by preparative recycling HPLC (column A, solv. 2) to give pure **2**. Fraction 10 (14.5 g) was separated by MPLC-1 and MPLC-2 to give crude **3** (7.5 g). This was purified by preparative recycling HPLC (column A, solv. 5) to give pure **3**. By MPLC-1 and MPLC-2, fraction 6 (5.5 g) gave a mixture of **4** and **5**. It was separated by HPLC (column A, solv. 6) to give **4** (63 mg) and **5** (38 mg). In the same manner as for

Table 1. ^{13}C-NMR Chemical Shifts of Glycosyl Moieties in Pyridine-d_5

	1	1a	6	12
28-*O*-Glycosyl moieties				
Glc^1-1	95.6	95.7	95.5	95.6
2	73.8	73.9	73.7	73.8
3	78.7	78.7	78.6	78.7
4	70.8	70.9	70.7	70.9
5	78.0	78.1	77.9	78.0
6	69.2	69.2	69.1	69.2
Glc^2-1	104.8	104.9	104.7	104.8
2	75.3	75.3	75.2	75.3
3	76.5	76.5	76.4	76.5
4	78.2	78.2	78.2	78.2
5	77.1	77.1	77.0	77.1
6	61.3	61.3	61.2	61.3
Rha^1-1	102.7	102.7	102.6	102.7
2	72.5	72.6	72.4	72.5
3	72.7	72.8	72.7	72.8
4	73.9	74.0	73.9	74.0
5	70.3	70.3	70.2	70.3
6	18.5	18.5	18.4	18.5
3-*O*-Glycosyl moieties				
Ara-1	104.6	104.7	102.0	
2	75.3	75.3	75.2	
3	75.1	75.2	73.7	
4	69.7	69.7	68.7	
5	66.3	66.3	65.0	
Rha^2-1	101.4	101.4	101.3	
2	71.9	72.0	72.3	
3	82.0	82.1	72.4	
4	72.8	72.8	74.1	
5	69.7	69.7	69.6	
6	18.4	18.4	18.4	
Rib-1	104.6	104.7		104.2
2	72.5	72.6		72.9
3	69.5	69.6		68.4
4	76.4	76.4		70.6
5	61.5	61.7		65.0
Glc^3-1	102.8	103.1		
2	74.2	74.2		
3	76.4	76.6		
4	81.2	81.0		
5	76.3	76.6		
6	60.7	61.9		
Glc^4-1	102.6	104.9		
2	75.1	74.8		
3	76.4	78.2		
4	71.9	71.5		
5	78.6	78.5		
6	62.4	62.4		

Table 2. ^{1}H-NMR Chemical Shifts of Glycosyl Moieties in Pyridine-d_5[a)]

	1	6	12
28-*O*-Glycosyl moieties			
Glc^1-1	6.22 d (8)	6.18 d (8)	6.23 d (8)
2	4.10 dd (9, 8)	4.07 dd (9, 8)	4.11 dd (9, 8)
3	4.18 dd (9, 9)	4.18 dd (9, 9)	4.19 dd (9, 9)
4	4.29 dd (9, 9)	4.27 dd (9, 9)	4.30 dd (9, 9)
5	4.09[b)]	4.05[b)]	4.07[b)]
6	4.33[b)]	4.30[b)]	4.32[b)]
	4.67[b)]	4.63[b)]	4.66[b)]
Glc^2-1	4.98 d (8)	4.95 d (8)	4.98 d (8)
2	3.93 dd (9, 8)	3.90 dd (9, 8)	3.93 dd (9, 8)
3	4.16 dd (9, 9)	4.10 dd (9, 9)	4.14 dd (9, 9)
4	4.40 dd (9, 9)	4.37 dd (9, 9)	4.41 dd (9, 9)
5	3.65 m	3.62 m	3.64 m
6	4.07[b)]	4.03[b)]	4.08 dd (12, 4)
	4.20[b)]	4.18[b)]	4.20[b)]
Rha^1-1	5.84 br s	5.81 br s	5.85 d (1.5)
2	4.67 m	4.63 m	4.67 m
3	4.54 dd (9, 3.5)	4.51 dd (9, 3.5)	4.64 dd (9, 3.5)
4	4.32 dd (9, 9)	4.29 dd (9, 9)	4.32 dd (9, 9)
5	4.95 m	4.91 m	4.96 m
6	1.69 d (6)	1.65 d (6.5)	1.69 d (6)
3-*O*-Glycosyl moieties			
Ara-1	5.05 d (6.5)	4.70 d (5.5)	
2	4.54 dd (6.5, 6.5)	4.42 dd (6, 5.5)	
3	4.02 m	4.15 m	
4	4.11[b)]	4.17[b)]	
5	3.67 br d	3.70 br d (11)	
	4.27[b)]	4.25[b)]	
Rha^2-1	6.28 br s	6.05 br s	
2	4.84 m	4.63 m	
3	4.68[b)]	4.56[b)]	
4	4.40 dd (9, 9)	4.20 dd (9, 9)	
5	4.65[b)]	4.50[b)]	
6	1.55 d (6)	1.65 d (6.5)	
Rib-1	5.80 d (5.5)		5.59 d (4)
2	4.07[b)]		4.24[b)]
3	4.64[b)]		4.42 dd (4, 3)
4	4.28[b)]		4.26 m
5	4.20[b)]		4.14 br d (12)
	4.26[b)]		4.22 br d (12)
Glc^3-1	4.87 d (8)		
2	3.88 dd (9, 8)		
3	4.10 dd (9, 9)		
4	4.29 dd (9, 9)		
5	3.64[b)]		
6	4.14[b)]		
	4.23[b)]		
Glc^4-1	5.35 d (8)		
2	5.77 dd (8, 8)		
3	4.32 dd (9, 8)		
4	4.18 dd (9, 9)		
5	4.06 m		
6	4.22[b)]		
	4.55[b)]		

a) Coupling constants (*J*) in Hz are given in parentheses. *b*) Overlapped signals.

fraction 6, fraction 9 (9.1 g) was separated to give four crude compounds. These compounds were purified by HPLC to give **6** (102 mg) (column A, solv. 3), **7** (52 mg) (column A, solv. 6), **8** (61 mg) (column A, solv. 4) and **13** (25 mg) (column B, solv. 7). Fraction 8 (11 g) was separated by MPLC-2 to give four crude compounds. These compounds were purified by repeated preparative recycling HPLC to give **9** (25 mg) (column A, solv. 3), **10** (31 mg) (column A, solv. 6), **11** (26 mg) (column A, solv. 4) and **12** (25 mg) (column B, solv. 7).

Acid-Hydrolysis of Saponins A few milligrams of each sample was dissolved in 2 N H_2SO_4–50% dioxane (2—4 ml) and heated at 90 °C for 2 h. After cooling, the reaction mixture was diluted with H_2O and concentrated to about half volume to yield precipitates, which were collected by filtration. The precipitates were examined by TLC (solv., benzene : EtOAc=1 : 1), which revealed the presence of hederagenin (in the cases of **1** and **12**) or gypsogenin (in the case of **6**). The filtrate was neutralized with saturated $Ba(OH)_2$ aq. and centrifuged. The supernatant was evaporated and the residue was dissolved in H_2O (*ca.* 0.5 ml) and subjected to HPLC analysis [column, YMC-Pack Polyamine II (250 mm×4.6 mm i.d.); solv., CH_3CN : H_2O : H_3PO_4=86 : 14 : 0.05; detector, Shimadzu RID-2A refractive index detector and JASCO OR-990 optical rotation detector; temperature, 50 °C], which revealed the presence of L-rhamnose (t_R 6.3 min, α_D −), D-ribose (t_R 7.2 min, α_D −), L-arabinose (t_R 10.5 min, α_D +) and D-glucose (t_R 16.9 min, α_D +).

Clematibetoside A (1) Yellowish amorphous powder, $[\alpha]_D^{28}$ −59.2° (*c*=0.33, MeOH). IR (KBr) cm^{-1}: 3440, 2931, 1700, 1633, 1527. FAB-MS (negative ion mode) *m/z*: 1837.8 [(M−H)$^-$], 1659.8 [(M−caffeoyloxy−H)$^-$], 1365.7 [(M−Rha−Glc−Glc−H)$^-$], 1189.6, 1027.6, 865.5, 733.5, 587.4. HR-FAB-MS: 1837.7908 (Calcd for $C_{85}H_{129}O_{43}$ 1837.7908). UV λ_{max} nm (log ε): 216 (4.33), 244 (4.19), 297 (4.31), 326 (4.38). ^{13}C-NMR: Tables 1 and 4. ^{13}C-NMR data for a caffeoyl moiety: 166.9 (C-1), 115.0 (C-2), 146.3 (C-3), 126.9 (C-4), 115.8 (C-5), 147.6 (C-6), 150.4 (C-7), 116.8 (C-8),

Table 3. NMR Data for Ribopyranosyl Moiety (**12**, **12a**) and Methyl α- and β-D-Ribopyranoside in Pyridine-d_5[a)]

	12	**12a**	Methyl β-D-ribopyranoside[b)]	Methyl α-D-ribopyranoside[b)]
^{1}H-NMR				
Rib-1	5.59 d (4)	5.60 d (4)	5.08 d (4)	4.73 d (3)
2	4.24[c)]	4.25 dd (4, 3)	4.13 ddd (4, 3, 1)	4.10 dd (3, 3)
3	4.42 dd (4, 3)	4.43 dd (3, 3)	4.36 dd (3, 3)	4.32 dd (3, 3)
4	4.26 m	4.27 m	4.20 dddd (5, 3, 2.5, 1)	4.07 ddd (8, 3.5, 3)
5	4.14 br d (12)	4.16 dd (11, 5)	4.00 dd (11.5, 2.5)	3.71 dd (11, 3.5)
	4.22 br d (12)	4.24 dd (11, 3)	4.08 dd (11.5, 5)	4.19 dd (11, 8)
^{13}C-NMR				
Rib-1	104.2	104.2	103.2	101.6
2	72.9	72.9	72.3	70.7
3	68.4	68.4	68.2	71.8
4	70.6	70.6	70.3	68.8
5	65.0	65.1	64.8	62.1

a) Coupling constants (*J*) in Hz are given in parentheses. *b*) Data were taken in our laboratory and assignments were confirmed by ^{1}H–^{1}H homo decoupling experiment and ^{1}H–^{13}C COSY. *c*) Overlapped signals.

Table 4. ^{13}C-NMR Chemical Shifts of Aglycone Moieties in Pyridine-d_5

C No.	Ole.	Hed.	Gyp.	**1**	**6**	**12**
1	38.9	38.9	38.5	39.1	38.2	38.7
2	28.2	27.6	27.1	26.4	25.2	25.7
3	78.0	73.7	71.7	81.0	80.2	81.4
4	39.4	42.9	56.3	43.5	55.4	43.4
5	55.8	48.8	48.0	47.6	48.3	47.3
6	18.8	18.7	21.1	18.1	20.5	18.2
7	33.3	33.0	32.6	32.7	32.4	32.7
8	39.8	39.8	40.1	39.9	40.1	40.0
9	48.1	48.2	47.7	48.2	47.9	48.1
10	37.4	37.3	36.2	36.8	36.0	37.0
11	23.8	23.8	23.7	23.8	23.6	23.8
12	122.5	122.7	122.3	122.9	122.5	122.9
13	144.8	145.0	144.9	144.1	144.1	144.1
14	42.0	42.2	42.2	42.1	42.1	42.1
15	28.3	28.4	28.3	28.3	28.1	28.3
16	23.8	23.8	23.8	23.3	23.2	23.3
17	46.7	46.7	46.7	47.0	46.9	47.0
18	42.7	42.0	42.0	41.6	41.6	41.5
19	46.7	46.5	46.5	46.1	46.1	46.2
20	31.0	31.0	31.0	30.7	30.7	30.7
21	34.3	34.3	34.2	33.9	33.9	34.0
22	33.3	33.3	33.2	32.5	32.4	32.5
23	28.7	68.2	207.1	63.8	207.6	64.4
24	16.5	13.1	9.7	14.0	10.6	13.5
25	15.5	16.0	15.7	16.2	15.6	16.2
26	17.5	17.5	17.4	17.5	17.4	17.5
27	26.2	26.2	26.2	26.0	26.0	26.0
28	180.2	180.4	180.1	176.5	176.4	176.5
29	33.3	33.3	33.3	33.0	33.1	33.1
30	23.8	23.8	23.8	23.6	23.6	23.7

Ole.: oleanolic acid, Hed.: hederagenin, Gyp.: gypsogenin.

122.0 (C-9), ^{1}H-NMR: Table 2 (sugar). ^{1}H-NMR data for a caffeoyl moiety: 6.70 (1H, d, J=16 Hz), 7.11 (1H, br d, J=8 Hz), 7.19 (1H, br d, J=8 Hz), 7.56 (1H, br s), 8.08 (1H, d, J=16 Hz).

Mild Alkaline-Hydrolysis of 1 A solution of **1** (17 mg) in 0.1 N KOH (5 ml), was left to stand for 1 h at room temperature. The reaction mixture was neutralized with dil. H_2SO_4 and extracted with *n*-BuOH (5 ml×4). The *n*-BuOH layer was concentrated and purified by preparative HPLC (column A, solv. 6) to give caffeic acid and **1a** (11 mg). Compound **1a**: ^{1}H-NMR: 4.82 (1H, d, J=6 Hz, H-1 of Ara), 4.97 (1H, d, J=8 Hz, H-1 of Glc^3), 4.98 (1H, d, J=8 Hz, H-1 of Glc^2), 5.16 (1H, d, J=7.5 Hz, H-1 of Glc^4), 5.81 (1H, d, J=5 Hz, H-1 of Rib), 5.83 (1H, br s, H-1 of Rha^1), 6.22 (1H, d, J=8 Hz, H-1 of Glc^1), 6.25 (1H, br s, H-1 of Rha^2). ^{13}C-NMR: Table 1 (sugar moiety). The NMR data of **1a** agreed with those of the clematernoside C deacylate obtained in the same manner from clematernoside C.[1)]

Clematibetoside B (6) White amorphous powder, $[\alpha]_D^{28}$ −13.0° (c=0.51, MeOH). IR (KBr) cm^{-1}: 3500, 2939, 1722, 1645, 1550. FAB-MS (negative ion mode) *m/z*: 1217.6 [(M−H)$^-$], 747.4 [(M−Rha−Glc−Glc−H)$^-$], 1189.6, 1027.6, 865.5, 733.5, 587.4. HR-FAB-MS: 1217.5953 (Calcd for $C_{59}H_{93}O_{26}$ 1217.5955). ^{1}H-NMR: Table 2. ^{13}C-NMR: Tables 1 and 4.

Clematibetoside C (12) White amorphous powder, $[\alpha]_D^{28}$ −25.7° (c=0.67, MeOH). IR (KBr) cm^{-1}: 3456, 2939, 1747, 1639, 1550. FAB-MS (negative ion mode) *m/z*: 1073.5 [(M−H)$^-$], 603.4 [(M−Rha−Glc−Glc−H)$^-$], HR-FAB-MS: 1073.5531 (Calcd for $C_{53}H_{85}O_{22}$ 1073.5533). ^{1}H-NMR: Table 2. ^{13}C-NMR: Tables 1 and 4.

Alkaline-Hydrolysis of 12 Compound **12** (13 mg) was dissolved in 0.5 N KOH (5 ml) and heated at 90 °C for 1 h. After cooling, the reaction mixture was neutralized with dil. H_2SO_4 and extracted with *n*-BuOH (5 ml×4). The *n*-BuOH layer was concentrated and purified by preparative HPLC (column A, solv. 6) to give **12a** (5 mg). ^{1}H- and ^{13}C-NMR: Table 3 (sugar moiety).

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