Triterpene Saponins from *Clematis chinensis* and Their Potential Anti-inflammatory Activity

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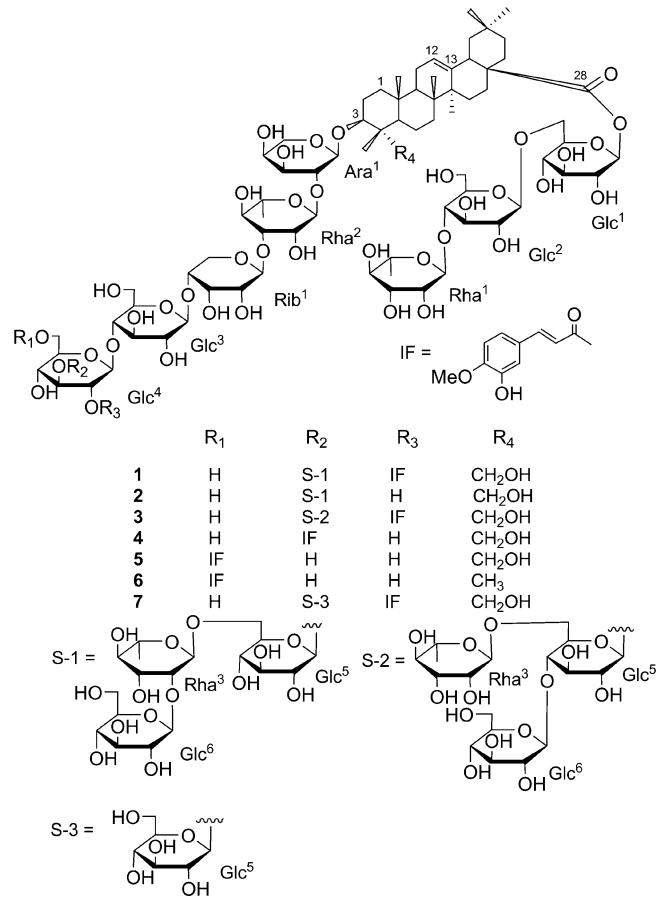
Seven new triterpene saponins, clematochinenosides A−G (**1**−**7**), together with 17 known saponins (**8**−**24**), were isolated from the roots and rhizomes of *Clematis chinensis*. Their structures were elucidated on the basis of spectroscopic evidence and hydrolysis products. Compounds **1**, **3**−**7**, and **20**−**24** showed inhibitory activities against COX-1 and COX-2 enzymes.

Clematis chinensis Osbeck (Ranunculaceae) is distributed widely in the south of the People's Republic of China. According to the *Chinese Pharmacopoeia*,[1] the roots and rhizomes of *C. chinensis*, *C. hexapetala* Pall., and *C. mandshurica* Rupr. are collectively termed "Weilingxian", a traditional Chinese herbal drug that is commonly used as an anti-inflammatory, antitumor, and analgesic agent. However, previous phytochemical investigations have revealed that the chemical constituents of these three species are quite different. The roots and rhizomes of *C. mandshurica* and *C. chinensis* are rich in saponins,[2−13] whereas the major components of *C. hexapetala* are flavonoids.[14] The aim of the present study was to explore the similarities in chemical constituents and bioactivity of tissues from these three species. Previously, we reported on the isolation and structural elucidation of flavonoids from *C. hexapetala*[14] and seven bidesmosides of oleanolic acid of and lignans from *C. mandshurica*.[13,15] Herein we report on the isolation and structure elucidation of seven new saponins based upon hederagenin and oleanolic acid from the roots and rhizomes of *C. chinensis* and on the inhibitory activity of all compounds isolated against the cyclooxygenase-1 (COX-1) and cyclooxygenase-2 (COX-2) enzymes.

Results and Discussion

A 50% EtOH extract of the dried roots and rhizomes of *C. chinensis* (4 kg) was suspended in H_2O and successively extracted with petroleum ether, EtOAc, and *n*-BuOH. The *n*-BuOH-soluble fraction was subjected to column chromatography over porous polymeric resin (D101), silica gel, and C_{18} silica gel, to yield compounds **1**−**24**. Comparison of their NMR and MS data with reported values revealed the structures of the known compounds **8**−**24** to be cirensenoside O,[16] kizutasaponin K_3,[9] ciwujianoside C_3,[17] kizutasaponin K_{10},[18] hederasaponin B,[7] kizutasaponin K_{12},[7] huzhangoside B,[7] huzhangoside D,[7] clematichinenoside C,[10] clematichinenoside B,[9] clematomanshurica saponin C,[13] 3-*O*-*β*-D-glucopyranosyl-(1→4)-*β*-D-glucopyranosyl-(1→4)-*β*-D-ribopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosylhederagenin 28-*O*-α-L-rhamnopyranosyl-(1→4)-*β*-D-glucopyranosyl-(1→6)-*β*-D-glucopyranoside,[19] clematernosides B−E,[8] and clematernoside K,[8] respectively.

Compounds **1**−**7** were isolated as white, amorphous powders. The monosaccharides obtained after aqueous acid hydrolysis of each compound were identified as glucose, rhamnose, arabinose, and ribose by TLC comparison with authentic samples. The absolute configuration of the monosaccharides was determined to be D for glucose and ribose and L for rhamnose and arabinose by GC analysis of chiral derivatives of the monosaccharides in the hydrolysate of each compound (see Experimental Section). The relatively large coupling constants (5.0−8.0 Hz) for the anomeric protons in the



	R_1	R_2	R_3	R_4
1	H	S-1	IF	CH_2OH
2	H	S-1	H	CH_2OH
3	H	S-2	IF	CH_2OH
4	H	IF	H	CH_2OH
5	IF	H	H	CH_2OH
6	IF	H	H	CH_3
7	H	S-3	IF	CH_2OH

^{1}H NMR spectra (Table 1) of these compounds suggested that the arabinopyranosyl moieties have an α-configuration and the glucopyranosyl and ribopyranosyl moieties a *β*-configuration. The α-configurations of the rhamnopyranosyl moieties were determined from the broad singlets observed for the anomeric protons. The downfield chemical shift at δ_C 80.9−88.7 (Agly-3) and the upfield chemical shift at δ_C 176.4−176.5 (Agly-28) in the ^{13}C NMR spectra of **1**−**7** (Table 2) indicated these compounds to be bisdesmosidic saponins.

The molecular formula of **1** was determined as $C_{104}H_{162}O_{57}$ by HRESIMS (positive-ion mode) experiments, which revealed a pseudomolecular ion peak $[M + Na]^+$ at *m*/*z* 2345.9679 (calcd 2345.9670). The aglycon of **1** was identified as hederagenin by comparison of the ^{1}H and ^{13}C NMR data obtained in 2D NMR experiments with literature values.[20] The ^{1}H NMR spectrum of **1** exhibited 11 anomeric proton resonances at *δ* 6.26 (1H, brs), 6.20 (1H, d, *J* = 8.0 Hz), 5.83 (1H, brs), 5.79 (1H, d, *J* = 5.0 Hz), 5.43 (1H, brs), 5.31 (1H, d, *J* = 8.0 Hz), 5.29 (1H, d, *J* = 8.0 Hz), 5.04

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Table 1. ^{1}H NMR Data of Compounds **1** and **2** (500 MHz, in C_5D_5N)

position	**1**	**2**	position	**1**	**2**
Glc[1]			5a	4.29 m	4.28 m
1	6.20 d (8.0)	6.21 d (8.0)	5b	4.29 m	4.28 m
2	4.08 dd (9.0, 8.0)	4.08 dd (9.0, 8.0)	Glc[3]		
3	4.20 dd (9.0, 9.0)	4.19 dd (9.0, 9.0)	1	4.90 d (8.0)	4.90 d (8.0)
4	4.28 m	4.28 m	2	3.87 dd (9.0, 8.0)	3.86 dd (9.0, 8.0)
5	4.07 m	4.07 m	3	4.15 m	4.12 m
6a	4.66 m	4.66 m	4	4.27 m	4.26 m
6b	4.31 m	4.31 m	5	3.64 m	3.64 m
Glc[2]			6a	4.23 m	4.23 m
1	4.99 d (8.0)	4.94 d (8.0)	6b	4.14 m	4.14 m
2	3.92 dd (9.0, 8.0)	3.91 dd (9.0, 8.0	Glc[4]		
3	4.14 d (9.0)	4.19 d (9.0)	1	5.29 d (8.0)	5.10 d (8.0)
4	4.40 (9.0)	4.41 (9.0)	2	5.71 dd (9.0, 8.0)	4.06 dd (9.0, 8.0)
5	3.66 m	3.62 m	3	4.13 m	4.01 m
6a	4.19 m	4.16 m	4	4.04 m	4.05 m
6b	4.05 m	4.07 m	5	3.64 brd (10.0)	3.86 m
Rha[1]			6a	4.22 m	4.22 m
1	5.83 brs	5.83 brs	6b	3.94 m	4.17 m
2	4.64 brs	4.64 brs	Glc[5]		
3	4.54 dd (9.0, 3.5)	4.53 dd (9.0, 3.5)	1	4.83 d (8.5)	5.09 d (8.0)
4	4.33 dd (9.0, 9.0)	4.32 dd (9.0, 9.0)	2	3.87 dd (9.0, 8.5)	3.91 dd (9.0, 8.0)
5	4.93 m	4.92 m	3	4.06 dd (9.0, 9.0)	4.07 dd (9.0, 9.0)
6	1.67 d (6.0)	1.67 d (6.0)	4	3.82 dd (9.0, 9.0)	3.89 dd (9.0, 9.0)
Ara[1]			5	4.06 m	4.06 m
1	5.04 d (6.5)	5.05 d (6.5)	6a	4.53 brd (10.0)	4.48 brd
2	4.56 dd (8.0 6.5)	4.54 dd (8.0 6.5)	6b	3.69 m	3.78 brd
3	3.99 dd (8.0 4.0)	4.01 dd (8.0 4.0)	Rha[3]		
4	4.11 m	4.10 m	1	5.43 brs	5.46 brs
5a	4.22 d (9.5)	4.23 d (9.5)	2	4.76 brs	4.83 brs
5b	3.64 d (9.5)	3.67 d (9.5)	3	4.56 m	4.54 m
Rha[2]			4	4.17 dd (9.0, 9.0)	4.16 dd (9.0, 9.0)
1	6.26 brs	6.28 brs	5	4.22 m	4.25 m
2	4.85 brs	4.83 brs	6	1.55 d (6.0)	1.55 d (6.0)
3	4.64 m	4.67 m	Glc[6]		
4	4.39 dd (9.0, 9.0)	4.39 dd (9.0, 9.0)	1	5.31 d (8.0)	5.28 d (8.0)
5	4.62 m	4.64 m	2	4.07 m	4.07 m
6	1.50 d (6.0)	1.50 d (6.0)	3	4.20 m	4.19 m
Rib[1]			4	4.22 m	4.26 m
1	5.79 d (5.0)	5.80 d (5.0)	5	3.95 m	3.93 m
2	4.09 m	4.07 m	6a	4.47 dd (12.0, 5.0)	4.45 dd (12.0, 5.0)
3	4.62 m	4.64 m	6b	4.36 dd (12.0, 5.0)	4.34 dd (12.0, 5.0)
4	4.31 m	4.29 m			

(1H, d, $J = 6.5$ Hz), 4.99 (1H, d, $J = 8.0$ Hz), 4.90 (1H, d, $J = 8.0$ Hz), and 4.83 (1H, d, $J = 8.0$ Hz), respectively. The three-proton doublets observed at δ 1.67 (3H, d, $J = 6.0$ Hz), 1.55 (3H, d, $J = 6.0$ Hz), and 1.50 (3H, d, $J = 6.0$ Hz) suggested the presence of three deoxyhexopyranosyl units in **1**. The spin−spin coupling system of individual monosaccharide units was identified by analysis of 1D TOCSY and 2D NMR spectra. ^{1}H NMR spectra of individual monosaccharide units were obtained by selective irradiation of the anomeric protons or methyl groups of rhamnose units in a series of 1D TOCSY experiments. Analysis of the ^{1}H−^{1}H COSY spectrum resulted in sequential assignment of all proton resonances of the 11 monosaccharide units, including identification of most of their multiple splitting patterns and coupling constants, as shown in Table 1. In HSQC experiments, proton resonances were correlated with those of the corresponding carbons, and associated anomeric protons were correlated with their respective carbon atoms from HSQC-TOCSY data, leading to unambiguous assignments of the carbons in each monosaccharide unit. Comparison of the carbon chemical shifts thus assigned with those of reference methyl glycosides,[21] taking into account the known effects of *O*-glycosylation, indicated that **1** contains one L-arabinopyranosyl unit (Ara), one D-ribopyranosyl unit (Rib), three L-rhamnopyranosyl units (Rha), and six D-glucopyranosyl units (Glc). Sugar−aglycon and sugar−sugar linkages were determined by HMBC and NOESY data analysis (Tables 1−3). Further analysis of the NMR data of **1** revealed the presence of a 3-hydroxy-4-methoxycinnamoyl group (isoferuloyl) (Tables 4 and 5). The occurrence of this substructure in **1** was confirmed on mild alkaline hydrolysis, which afforded isoferulic acid and **2** (see below). The correlation observed between Glc^4-H-2 and the carbonyl carbon (δ 166.7) of the isoferuloyl moiety in the HMBC spectrum indicated that the isoferuloyl moiety is linked to the sugar chain on Glc^4-C-2 (δ 72.7). From the above evidence, the structure of **1** was established as 3-*O*-β-D-glucopyranosyl-(1→2)-α-L-rhamnopyranosyl-(1→6)-β-D-glucopyranosyl-(1→3)-[(2-*O*-isoferuloyl)-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, which has been named clematochinenoside A.

The molecular formula of **2** was determined as $C_{94}H_{154}O_{54}$ by HRESIMS, which showed a pseudomolecular ion peak $[M + Na]^+$ at *m/z* 2169.9244 (calcd 2169.9197). Acid hydrolysis of **2** with 2 N aqueous CF_3COOH gave hederagenin, L-arabinose, D-glucose, L-rhamnose, and D-ribose. Comparison of the NMR data for **2** and **1** revealed that the compounds are similar except that signals due to the isoferuloyl moiety linked to Glc^4-C-2 in **1** were absent for **2**. This observation was supported by a relative upfield shift of Glc^4-H-2 of **2** at δ 4.06 (Glc^4-H-2 of **1**, δ 5.71) in the ^{1}H NMR spectrum and a downfield shift of Glc^4-C-1 of **2** at δ 104.3 (Glc^4-C-1 of **1**, δ 102.1) in the ^{13}C NMR spectrum. Thus, the structure of **2** was determined as 3-*O*-β-D-glucopyranosyl-(1→2)-α-L-rhamnopyranosyl-(1→6)-β-D-glucopyranosyl-(1→3)-β-D-glucopyranosyl-(1→4)-β-D-glucopyranosyl-(1→4)-β-D-ribopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl hederagenin

Table 2. ^{13}C NMR Data for the Aglycon Moieties of **1**−**7** (125 MHz, in C_5D_5N)

position	1	2	3	4	5	6	7
1	39.0	39.0	39.1	39.0	39.0	38.9	39.1
2	26.3	26.3	26.3	26.3	26.3	26.6	26.3
3	80.9	80.9	80.9	81.0	80.9	88.7	80.9
4	43.5	43.5	43.5	43.5	43.5	39.5	43.6
5	47.6	47.6	47.6	47.6	47.6	55.7	47.6
6	18.0	18.1	18.1	18.0	18.0	18.5	18.0
7	32.6	32.6	32.6	32.7	32.6	33.1	32.7
8	39.8	39.8	39.9	39.8	39.8	39.8	39.9
9	48.1	48.1	48.2	48.1	48.1	48.0	48.2
10	36.8	36.8	36.8	36.8	36.8	37.0	36.8
11	24.0	23.8	23.8	23.7	23.7	23.6	23.6
12	123.0	123.0	123.0	123.0	123.0	122.8	122.9
13	144.0	144.0	144.0	144.0	144.0	144.0	144.0
14	42.0	42.0	42.0	42.0	42.0	42.1	42.1
15	28.2	28.2	28.2	28.2	28.2	28.1	28.2
16	23.3	23.3	23.3	23.3	23.2	23.3	23.3
17	46.9	46.9	47.0	46.9	46.9	47.0	47.0
18	41.6	41.6	41.6	41.5	41.5	41.6	41.6
19	46.1	46.1	46.1	46.1	46.1	46.2	46.1
20	30.6	30.7	30.7	30.6	30.7	30.7	30.7
21	33.9	33.9	33.9	33.9	33.9	33.9	33.9
22	32.5	32.5	32.5	32.4	32.5	32.5	32.7
23	63.7	63.8	63.8	63.9	63.8	28.1	63.8
24	14.1	14.0	14.1	14.1	14.0	17.1	14.1
25	16.1	16.1	16.1	16.1	16.1	15.6	16.1
26	17.4	17.5	17.5	17.5	17.4	17.4	17.5
27	26.3	26.3	26.4	26.3	26.3	26.0	26.3
28	176.5	176.5	176.5	176.4	176.4	176.5	176.5
29	33.0	33.0	33.0	33.0	33.0	33.1	33.0
30	23.6	23.6	23.6	23.6	23.6	23.7	23.6

28-*O*-α-L-rhamnopyranosyl-(1→4)-*β*-D-glucopyranosyl-(1→6)-*β*-D-glucopyranoside, (clematochinenoside B).

The molecular formula of **3** was determined as $C_{104}H_{162}O_{57}$ from the pseudomolecular ion peak $[M + Na]^+$ at *m/z* 2345.9673 (calcd 2345.9670) in the HRESIMS. Acid hydrolysis of **3** with 2 N aqueous CF_3COOH gave hederagenin, isoferulic acid, L-arabinose, D-glucose, L-rhamnose, and D-ribose. Comparison of the NMR data for **3** and **1** revealed that the compounds are similar except for the linkage site of the terminal glucopyranosyl group (Glc^6) in the sugar chain, which is connected to Glc^5-C-4 in **3** instead of to Rha^3-C-2 in **1** according to HMBC correlations observed between Glc^6-H-1 at *δ* 4.95 and Glc^5-C-4 at *δ* 82.0. This was further supported by NOE correlations observed between Glc^6-H-1 and Glc^5-H-4 at *δ* 4.12. The proton and carbon signals were assigned unambiguously using ^1H, ^{13}C, $^1H-^1H$ COSY, 1D TOCSY, HSQC, HMBC, and NOESY NMR experiments. Therefore, the structure of **3** (clematochinenoside C) was proposed as 3-*O*-α-L-rhamnopyranosyl-(1→6)-[*β*-D-glucopyranosyl-(1→4)]-*β*-D-glucopyranosyl-(1→3)-[(2-*O*-isoferuloyl)-*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.

The molecular formula of **4** was determined as $C_{86}H_{132}O_{43}$ by HRESIMS, which showed a pseudomolecular ion peak $[M + Na]^+$ at *m/z* 1875.8055 (calcd 1875.8035). Studies of 1D and 2D NMR spectra data led to identification of hederagenin as the aglycon in **4**.[20] Acid hydrolysis of **4** with 2 N aqueous CF_3COOH gave hederagenin, isoferulic acid, L-arabinose, D-glucose, L-rhamnose, and D-ribose. Mild alkaline hydrolysis of **4** with 0.1 N KOH afforded isoferulic acid and 3-*O*-*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.[19] Alkaline hydrolysis of **4** afforded a prosapogenin (**4a**), which was identified as CP_{10} by direct comparison of ^1H and ^{13}C NMR data obtained in 2D NMR experiments with literature values.[4] The sugar sequence of **4a** was confirmed on analysis of its $ESIMS^n$ data in the negative mode, which showed fragments at *m/z* 1043.6, 881.5, 749.4, 603.3, and 471.2, corresponding to $[M - Glc\text{-}H]^-$, $[M - 2Glc\text{-}H]^-$, $[M - 2Glc\text{-}Rib\text{-}H]^-$, $[M - 2Glc\text{-}Rib\text{-}Rha\text{-}H]^-$, and $[M - 2Glc\text{-}Rib\text{-}Rha\text{-}Ara\text{-}H]^-$, respectively. Comparison of the ^1H and ^{13}C NMR data for **4** with those for the known compound clematernoside B revealed that these substances differ only in the aglycon part. Thus, a set of carbon signals at *δ* 80.9 (Agly-3), 47.6 (Agly-5), and 63.9 (Agly-23) was observed in the ^{13}C NMR spectrum of **4** instead of *δ* 88.6 (Agly-3), 55.9 (Agly-5), and 28.0 (Agly-23) for clematernoside B. On the basis of all this evidence, the structure of **4** (clematochinenoside D) was assigned as 3-*O*-[(3-*O*-isoferuloyl)-*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.

The molecular formula of **5** was determined as $C_{86}H_{132}O_{43}$ from the pseudomolecular ion peak $[M + Na]^+$ at *m/z* 1875.7952 (calcd 1875.8035) in the HRESIMS. Analysis of the 1D and 2D NMR spectroscopic data led to identification of the aglycon as hederagenin.[20] Acid hydrolysis of **5** yielded hederagenin, isoferulic acid, L-arabinose, D-glucose, L-rhamnose, and D-ribose. Comparison of the ^1H and ^{13}C NMR spectra of **5** with that of **4** indicated that the two compounds are similar structurally. The only difference found was in the linkage site of the isoferuloyl group, which is at Glc^4-C-6 in **5** instead of Glc^4-C-3 in **4**. This correlation was established by the relative downfield shift at *δ* 5.17 and 4.67 for Glc^4-H-6 in the ^1H NMR spectrum of **4** (*δ* 4.18 and 4.49) and the long-range correlation observed between Glc^4-H-6 and the carbonyl carbon (*δ* 167.2) of the isoferuloyl group in the HMBC spectrum. Thus, the structure of **5** (clematochinenoside E) was established as 3-*O*-[(6-*O*-isoferuloyl)-*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.

The molecular formula of **6** was determined as $C_{86}H_{132}O_{42}$ by HRESIMS, which showed a pseudomolecular ion peak at *m/z* 1859.8105 $[M + Na]^+$ (calcd 1859.8085). The aglycon of **6** was identified as oleanolic acid by comparison of its ^1H and ^{13}C 2D NMR spectroscopic data with reported values.[22] Acid hydrolysis of **6** yielded oleanolic acid, L-arabinose, D-glucose, L-rhamnose, and D-ribose. Mild alkaline hydrolysis of **6** afforded a prosapogenin (**6a**), which was identified as CP_9 by direct comparison of ^1H and ^{13}C NMR data obtained in 2D NMR experiments with literature values.[4] The sugar sequence of **6a** was confirmed on analysis of its $ESIMS^n$ data in the negative mode, which gave fragments at *m/z* 1027.6, 865.5, 733.4, 587.4, and 455.3, corresponding to $[M - Glc\text{-}H]^-$, $[M - 2Glc\text{-}H]^-$, $[M - 2Glc\text{-}Rib\text{-}H]^-$, $[M - 2Glc\text{-}Rib\text{-}Rha\text{-}H]^-$, and $[M - 2Glc\text{-}Rib\text{-}Rha\text{-}Ara\text{-}H]^-$, respectively. Comparison of the ^1H and ^{13}C NMR data for **6** with those for **5** revealed that they differ only in the aglycon part. Thus, a set of carbon signals at *δ* 88.7 (Agly-3), 55.7 (Agly-5), and 28.1 (Agly-23) was observed for **6** compared to *δ* 80.9 (Agly-3), 47.6 (Agly-5), and 63.8 (Agly-23) for **5**. NMR signals due to the sugar moieties were identical for both compounds, except for signals due to an arabinopyranosyl moiety connected to the aglycon (Ara-H-1 at *δ* 4.82, Ara-H-3 at *δ* 4.25, Ara-C-1 at *δ* 105.3, Ara-C-3 at *δ* 74.8). Thus, the structure of **6** (clematochinenoside F) was determined unambiguously as 3-*O*-[(6-*O*-isoferuloyl)-*β*-D-glucopyranosyl]-(1→4)-*β*-D-glucopyranosyl-(1→4)-*β*-D- ribopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl oleanolic acid 28-*O*-α-L-rhamnopyranosyl-(1→4)-*β*-D-glucopyranosyl-(1→6)-*β*-D-glucopyranoside.

The molecular formula of **7** was determined as $C_{92}H_{142}O_{48}$ from the pseudomolecular ion peak at *m/z* 2037.8506 $[M + Na]^+$ (calcd 2037.8563) in the HRESIMS. Acid hydrolysis of **7** afforded hederagenin, L-arabinose, D-glucose, L-rhamnose, and D-ribose.

Table 3. ^{13}C NMR Data for the Sugar Moieties of Compounds **1**−**7** (125 MHz, in C_5D_5N)

position	1	2	3	4	5	6	7	position	1	2	3	4	5	6	7
Glc[1]								Glc[3]							
1	95.6	95.6	95.6	95.5	95.5	95.5	95.6	1	102.8	103.1	102.9	103.1	103.1	103.1	102.9
2	73.8	73.8	73.8	73.8	73.8	73.8	73.8	2	74.3	74.2	74.2	74.2	74.2	74.2	74.2
3	78.6	78.6	78.7	78.6	78.6	78.6	78.6	3	76.4	76.6	76.5	76.4	76.5	76.5	76.5
4	70.7	70.7	70.8	70.7	70.7	70.7	70.8	4	81.0	80.9	81.1	81.0	81.8	81.8	81.1
5	78.0	78.0	78.0	78.0	78.0	78.0	78.0	5	76.3	76.4	76.2	76.4	76.4	76.4	76.4
6	69.1	69.1	69.2	69.1	69.1	69.1	69.2	6	60.6	61.9	60.6	61.2	61.5	61.5	60.7
Glc[2]								Glc[4]							
1	104.7	104.8	104.8	104.7	104.7	104.7	104.8	1	102.1	104.3	102.2	104.3	105.1	105.1	102.3
2	75.3	75.3	75.3	75.3	75.3	75.3	75.3	2	72.7	73.2	72.7	72.5	72.3	72.3	72.9
3	76.3	76.4	76.5	76.4	76.4	76.4	76.3	3	87.3	88.9	86.5	78.9	78.6	78.6	86.4
4	78.3	78.2	78.2	78.1	78.3	78.1	78.2	4	70.1	69.4	70.1	69.1	71.2	71.2	70.2
5	77.1	77.1	77.1	77.1	77.1	77.1	77.1	5	77.7	77.7	77.8	78.1	75.5	75.6	77.9
6	61.2	61.2	61.2	61.2	61.2	61.2	61.2	6	61.9	61.4	62.0	61.9	64.2	64.2	62.2
Rha[1]								Glc[5]							
1	102.6	102.6	102.7	102.7	102.7	102.7	102.7	1	105.8	105.8	105.2				105.5
2	72.5	72.4	72.6	72.5	72.5	72.5	72.5	2	74.5	75.2	73.9				74.6
3	72.7	72.7	72.8	72.7	72.7	72.7	72.7	3	78.3	78.3	76.4				78.3
4	73.9	73.9	74.0	73.9	73.9	73.9	74.0	4	71.8	72.0	82.0				71.5
5	70.2	70.2	70.3	70.2	70.2	70.2	70.3	5	76.3	76.4	74.8				78.7
6	18.5	18.5	18.5	18.4	18.4	18.4	18.5	6	68.9	68.8	68.0				62.6
Ara[1]								Rha[3]							
1	104.6	104.6	104.7	104.6	104.6	105.3	104.7	1	101.3	101.3	102.7				
2	75.3	75.3	75.3	75.3	75.3	75.3	75.3	2	82.0	82.0	71.9				
3	75.2	75.2	75.2	75.1	75.1	74.8	75.2	3	72.6	72.6	72.7				
4	69.7	69.7	69.7	69.6	69.6	69.4	69.7	4	74.5	74.7	73.9				
5	66.3	66.3	66.4	66.2	66.2	65.5	66.4	5	69.6	69.7	69.9				
Rha[2]								6	18.5	18.5	18.6				
1	101.3	101.4	101.4	101.3	101.3	101.4	101.4	Glc[6]							
2	71.8	71.9	71.9	71.8	71.8	71.9	71.9	1	107.2	107.2	105.2				
3	81.9	82.0	82.1	81.2	82.0	82.1	80.9	2	75.9	75.9	74.7				
4	72.7	72.8	72.7	72.6	72.5	72.5	72.7	3	78.3	78.5	78.3				
5	69.6	69.6	69.8	69.7	69.6	69.7	69.7	4	71.2	71.2	71.6				
6	18.4	18.4	18.4	18.4	18.4	18.4	18.4	5	78.6	78.6	78.5				
Rib[1]								6	62.3	62.4	62.5				
1	104.6	104.6	104.7	104.7	104.7	104.7	104.7								
2	72.6	72.5	72.5	72.6	72.6	72.6	72.5								
3	69.6	69.5	69.6	69.6	69.6	69.8	69.7								
4	76.2	76.3	76.5	76.4	76.4	76.4	76.4								
5	61.5	61.6	61.5	61.7	61.6	61.7	61.5								

Table 4. ^1H NMR Data for the Isoferuloyl Moieties of Compounds **1** and **3**−**7** (500 MHz, in C_5D_5N)

position	1	3	4	5	6	7
2	6.89 d (16)	6.90 d (16)	6.56 d (16)	6.64 d (16)	6.64 d (16)	6.89 d (16)
3	8.09 d (16)	8.12 d (16)	7.86 d (16)	8.01 d (16)	8.01 d (16)	8.10 d (16)
5	7.52 d (2)	7.53 d (2)	7.41 d (2)	7.50 d (2)	7.50 d (2)	7.52 d (2)
8	6.87 d (8.5)	6.87 d (8.5)	6.89 d (8.5)	6.83 d (8.5)	6.83 d (8.5)	6.87 d (8.5)
9	7.07 dd (8.5, 2)	7.09 dd (8.5, 2)	6.99 dd (8.5, 2)	7.11 dd (8.5, 2)	7.11 dd (8.5, 2)	7.08dd (8.5, 2)
OCH_3	3.73 s	3.73 s	3.71 s	3.73 s	3.73 s	3.73 s

Table 5. ^{13}C NMR Data for the Isoferuloyl Moieties of Compounds **1** and **3**−**7** (125 MHz, in C_5D_5N)

position	1	3	4	5	6	7
1	166.7	166.6	167.2	167.2	167.3	166.8
2	116.3	116.4	116.5	115.3	115.8	116.4
3	145.6	145.6	145.1	145.8	145.8	145.7
4	128.5	128.6	128.3	128.3	128.3	128.6
5	115.4	115.4	115.2	115.2	115.2	115.4
6	148.4	148.4	148.3	148.3	148.3	148.4
7	150.8	150.9	150.8	150.8	150.8	150.9
8	112.0	112.1	111.9	111.9	111.9	112.1
9	121.4	121.4	121.7	121.7	121.7	121.5
OCH_3	55.8	55.8	55.7	55.7	55.7	55.8

Comparison of the NMR data for **7** with those of **1** revealed that the Rha[3] linked to Glc[5]-C-6 in **1** was absent in **7**. This was supported by a relative upfield shift of the ^{13}C NMR peak at δ 62.6 for Glc[5]-C-6 in **7** compared to δ 68.9 in **1**. Thus, the structure of **7** (clematochinenoside G) was determined as 3-*O*-β-D-glucopyranosyl-(1→3)-[(2-*O*-isoferuloyl)-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.

Compounds **1**−**24** were assayed for their potential anti-inflammatory activity against the COX-1 and COX-2 enzymes. Compounds **1**, **3**−**7**, and **20**−**24** showed inhibitory activities against both enzymes (Table 6). However, no COX-2 selectivity was evident for any of the active compounds.

Experimental Section

General Experimental Procedures. Optical rotations were measured on a Perkin-Elmer 243B digital polarimeter. UV spectra were obtained on a TU-1901 spectrometer. IR spectra were recorded on an Avater-360 spectrometer. NMR spectra were recorded on a Bruker INOVA 500 spectrometer with TMS as internal standard. HRESIMS was measured on a Bruker APEX IV FT-MS (7.0 T) mass spectrometer in the positive-ion mode. ESIMS was obtained on an Agilent XCT 6320 IT mass spectrometer in the negative-ion mode. HPLC was performed on an ODS column (Agilent Technologies 250 × 10 mm i.d., 5 μm) with an Alltech evaporative light scattering detector. GC was employed on an Agilent 6890N gas chromatograph. Column chromatography was performed with silica gel (200−300 mesh, Qingdao Haiyang Chemical

Table 6. Inhibitory Effects of Compounds **1**, **3**−**7**, and **20**−**24** against Cyclooxygenases-(COX-) I and II

	IC_{50} value (μM)			IC_{50} value (μM)	
compound[a]	COX-1	COX-2	compound[a]	COX-1	COX-2
1	7.8	6.7	**21**	7.6	8.8
3	6.3	6.1	**22**	8.8	9.0
4	8.8	7.8	**23**	8.3	8.5
5	8.9	7.9	**24**	5.9	6.7
6	7.8	7.2	SC560[b]	0.01	
7	7.5	8.9	NS398[c]		0.15
20	8.0	7.6			

[a] Compounds **2** and **8**−**19** were inactive (IC_{50} > 50 μM). [b] SC560 is a selective inhibitor of COX-1. [c] NS398 is a selective inhibitor of COX-2.

Co., Ltd.), D101 porous polymer resin (Tianjin Chemical Industry Co., Ltd.), and C_{18} silica gel (150−200 mesh, Merck; performed by applying a N_2 pressure of 0.5 MPa).

Plant Material. The roots and rhizomes of *C. chinensis* were collected in December 2007 in Shaoguan, Guangdong Province, People's Republic of China. The identification of the plant was performed by one of the authors (P.-F.T.). A voucher specimen (CC 200712) is maintained in the herbarium of Peking University Modern Research Center for Traditional Chinese Medicine.

Extraction and Isolation. The dried roots and rhizomes (10 kg) of *C. chinensis* were extracted with 50% EtOH (3 × 30 L). After removing the solvent, the residue (4000 g) was suspended in H_2O and successively extracted with petroleum ether, EtOAc, and *n*-BuOH. The *n*-BuOH extract (230 g) was subjected to D101 porous polymer resin column chromatography and eluted with H_2O and 30% and 80% EtOH, successively. The fraction that eluted with 80% EtOH (120 g) was subjected to silica gel column chromatography (15 cm × 160 cm) and eluted with $CHCl_3$−MeOH (4:1, 2:1, 1:2) to afford fractions 1−3. These fractions were subjected to C_{18} silica gel column chromatography (4 cm × 50 cm) and eluted with MeOH−H_2O in a gradient of MeOH (MeOH−H_2O, 30:70 → 100:0%, N_2 pressure 0.5 MPa) to afford subfractions 1-1-1-4, 2-1-2-3, and 3-1-3-4. Subfraction 1-1 (0.6 g) was isolated by preparative HPLC (MeOH−H_2O, 68:32, 2.0 mL/min) to yield compounds **8** (32 mg) and **10** (12 mg). Subfraction 1-3 (1.1 g) was isolated by preparative HPLC (MeOH−H_2O, 65:35, 2.0 mL/min) to yield compounds **9** (8 mg), **11** (48 mg), and **12** (51 mg). Subfraction 1-4 (2.9 g) was isolated by preparative HPLC (MeOH−H_2O, 62:38, 2.0 mL/min) to yield compounds **14** (27 mg) and **16** (86 mg). Subfraction 2-2 (2.8 g) was isolated by preparative HPLC (MeOH−H_2O, 60:40, 2.0 mL/min) to yield compounds **13** (66 mg) and **18** (72 mg). Subfraction 2-4 (1.6 g) was isolated by preparative HPLC (MeCN−H_2O, 32:68, 2.0 mL/min) to yield compounds **15** (12 mg), **17** (18 mg), and **19** (55 mg). Subfraction 3-1 (3.7 g) was isolated by preparative HPLC (MeCN−H_2O, 29:71, 2.0 mL/min) to yield compounds **4** (19 mg), **5** (37 mg), **6** (52 mg), **20** (17 mg), and **22** (24 mg). Subfraction 3-2 (4.3 g) was isolated by preparative HPLC (MeCN−H_2O, 27:73, 2.0 mL/min) to yield compounds **7** (33 mg), **21** (48 mg), **23** (21 mg), and **24** (32 mg). Subfraction 3-4 (5.8 g) was isolated by preparative HPLC (MeCN−H_2O, 24:76, 2.0 mL/min) to yield compounds **1** (27 mg), **2** (27 mg), and **3** (61 mg).

Clematochinenoside A (1): white, amorphous powder; $[\alpha]_D^{20}$ −62 (*c* 1.0, MeOH); UV (MeOH) λ_{max} (log ε) 243 (2.36), 296 (sh), 324 (4.33) nm; IR (KBr) ν_{max} 3400, 2927, 1724, 1633, 1513, 1062 cm^{-1}; ^1H NMR data, see Tables 1 and 4; ^{13}C NMR data, see Tables 2, 3, and 5; HRESIMS *m/z* 2345.9679 $[M + Na]^+$ (calcd for $C_{104}H_{162}O_{57}Na$, 2345.9670).

Clematochinenoside B (2): white, amorphous powder; $[\alpha]_D^{20}$ −31 (*c* 1.0, MeOH); IR (KBr) ν_{max} 3421, 2927, 1734, 1062 cm^{-1}; ^1H NMR data, see Table 1; ^{13}C NMR data, see Tables 2 and 3; HRESIMS *m/z* 2169.9244 $[M + Na]^+$ (calcd for $C_{94}H_{154}O_{54}Na$, 2169.9197).

Clematochinenoside C (3): white, amorphous powder; $[\alpha]_D^{20}$ −58 (*c* 1.0, MeOH); UV (MeOH) λ_{max} (log ε) 242 (2.33), 296 (sh), 326 (4.27) nm; IR (KBr) ν_{max} 3420, 2927, 1719, 1632, 1513, 1062 cm^{-1}; ^1H NMR data (C_5D_5N, 500 MHz) δ 1.15 (3H, s, Me-27), 1.10 (3H, s, Me-24), 1.07 (3H, s, Me-26), 0.93 (3H, s, Me-25), 0.85 (3H, s, Me-29), 0.84 (3H, s, Me-30), 4.25 (1H, dd, *J* = 12.0, 5.0 Hz, H-3), 4.23 (1H, d, *J* = 12.0 Hz, H-23a), 3.88 (1H, d, *J* = 12.0 Hz, H-23b), 5.37 (1H, brs, H-12), 6.27 (1H, brs, Rha^2-H-1), 6.21 (1H, d, *J* = 8.0 Hz, Glc^1-H-1), 5.83 (1H, brs, Rha^1-H-1), 5.81 (1H, d, *J* = 5.0 Hz, Rib^1-H-1), 5.47 (1H, brs, Rha^3-H-1), 5.29 (1H, d, *J* = 8.0 Hz, Glc^4-H-1), 5.05 (1H, d, *J* = 8.0 Hz, Ara^1-H-1), 4.98 (1H, d, *J* = 8.0 Hz, Glc^2-H-1), 4.95 (1H, d, *J* = 8.0 Hz, Glc^6-H-1), 4.90 (1H, d, *J* = 8.0 Hz, Glc^5-H-1), 4.89 (1H, d, *J* = 8.0 Hz, Glc^3-H-1), 4.56 (1H, dd, *J* = 8.0, 6.5 Hz, Ara^1-H-2), 4.65 (1H, m, Rha^2-H-3), 4.30 (1H, m, Rib^1-H-4), 4.26 (1H, m, Glc^3-H-4), 4.14 (1H, m, Glc^4-H-3), 3.90 (1H, m, Glc^5-H-4), 4.87 (1H, m, Glc^5-H-6a), 4.05 (1H, m, Glc^5-H-6b), 1.69 (3H, d, *J* = 6.0 Hz, Rha^1-H-6), 1.64 (3H, d, *J* = 6.0 Hz, Rha^3-H-6), 1.51 (3H, d, *J* = 6.0 Hz, Rha^2-H-6); ^1H NMR data of the isoferuloyl moiety, see Table 4; ^{13}C NMR data, see Tables 2, 3, and 5; HRESIMS *m/z* 2345.9673 $[M + Na]^+$ (calcd for $C_{104}H_{162}O_{57}Na$, 2345.9670).

Clematochinenoside D (4): white, amorphous powder; $[\alpha]_D^{20}$ −52 (*c* 1.0, MeOH); UV (MeOH) λ_{max} (log ε) 242 (2.36), 298 (sh), 325 (4.11) nm; IR (KBr) ν_{max} 3385, 2922, 1719, 1635, 1058 cm^{-1}; ^1H NMR data (C_5D_5N, 500 MHz) δ 1.15 (3H, s, Me-27), 1.11 (3H, s, Me-24), 1.07 (3H, s, Me-26), 0.93 (3H, s, Me-25), 0.85 (3H, s, Me-29), 0.83 (3H, s, Me-30), 4.24 (1H, dd, *J* = 12.0, 5.0 Hz, H-3), 4.23 (1H, d, *J* = 12.0 Hz, H-23a), 3.87 (1H, d, *J* = 12.0 Hz, H-23b), 5.36 (1H, brs, H-12), 6.33 (1H, brs, Rha^2-H-1), 6.20 (1H, d, *J* = 8.0 Hz, Glc^1-H-1), 5.82 (1H, brs, Rha^1-H-1), 5.78 (1H, d, *J* = 5.0 Hz, Rib^1-H-1), 5.27 (1H, d, *J* = 7.5 Hz, Glc^4-H-1), 5.04 (1H, d, *J* = 6.5 Hz, Ara^1-H-1), 4.97 (1H, d, *J* = 8.0 Hz, Glc^2-H-1), 4.92 (1H, d, *J* = 8.0 Hz, Glc^3-H-1), 4.55 (1H, dd, *J* = 8.0, 6.5 Hz, Ara^1-H-2), 4.65 (1H, m, Rha^2-H-3), 4.31 (1H, m, Rib^1-H-4), 4.27 (1H, m, Glc^3-H-4), 1.68 (3H, d, *J* = 6.0 Hz, Rha^1-H-6), 1.51 (3H, d, *J* = 6.0 Hz, Rha^2-H-6); ^1H NMR data of the isoferuloyl moiety, see Table 4; ^{13}C NMR data, see Tables 2, 3, and 5; HRESIMS *m/z* 1875.8055 $[M + Na]^+$ (calcd for $C_{86}H_{132}O_{43}Na$, 1875.8035).

Clematochinenoside E (5): white, amorphous powder; $[\alpha]_D^{20}$ −44 (*c* 1.0, MeOH); UV (MeOH) λ_{max} (log ε) 238 (2.56), 298 (sh), 321 (4.37) nm; IR (KBr) ν_{max} 3420, 2929, 1712, 1632, 1060 cm^{-1}; ^1H NMR data (C_5D_5N, 500 MHz) δ 1.14 (3H, s, Me-27), 1.08 (3H, s, Me-24), 1.06 (3H, s, Me-26), 0.93 (3H, s, Me-25), 0.84 (3H, s, Me-29), 0.83 (3H, s, Me-30), 4.25 (1H, dd, *J* = 12.0, 5.0 Hz, H-3), 4.23 (1H, d, *J* = 12.0 Hz, H-23a), 3.88 (1H, d, *J* = 12.0 Hz, H-23b), 5.37 (1H, brs, H-12), 6.30 (1H, brs, Rha^2-H-1), 6.20 (1H, d, *J* = 8.0 Hz, Glc^1-H-1), 5.82 (1H, brs, Rha^1-H-1), 5.79 (1H, d, *J* = 5.0 Hz Rib^1-H-1), 5.14 (1H, d, *J* = 7.5 Hz, Glc^4-H-1), 5.04 (1H, d, *J* = 6.5 Hz, Ara^1-H-1), 4.96 (1H, d, *J* = 8.0 Hz, Glc^2-H-1), 4.93 (1H, d, *J* = 8.0 Hz, Glc^3-H-1), 4.56 (1H, dd, *J* = 8.0, 6.5 Hz, Ara^1-H-2), 4.68 (1H, m, Rha^2-H-3), 4.31 (1H, m, Rib^1-H-4), 4.26 (1H, m, Glc^3-H-4), 1.67 (3H, d, *J* = 6.0 Hz, Rha^1-H-6), 1.51 (3H, d, *J* = 6.0 Hz, Rha^2-H-6); ^1H NMR data of the isoferuloyl moiety, see Table 4; ^{13}C NMR data, see Tables 2, 3, and 5; HRESIMS *m/z* 1875.7952 $[M + Na]^+$ (calcd for $C_{86}H_{132}O_{43}Na$, 1875.8035).

Clematochinenoside F (6): white, amorphous powder; $[\alpha]_D^{20}$ −39 (*c* 1.0, MeOH); UV (MeOH) λ_{max} (log ε) 246 (2.06), 292 (sh), 328 (4.43) nm; IR (KBr) ν_{max} 3408, 2938, 1715, 1062 cm^{-1}; ^1H NMR data (C_5D_5N, 500 MHz) δ 1.24 (3H, s, Me-23), 1.22 (3H, s, Me-27), 1.10 (3H, s, Me-24), 1.04 (3H, s, Me-26), 0.87 (3H, s, Me-29), 0.87 (3H, s, Me-30), 0.84 (3H, s, Me-25), 3.25 (1H, dd, *J* = 12.0, 5.0 Hz, H-3), 5.37 (1H, brs, H-12), 6.28 (1H, brs, Rha^2-H-1), 6.21 (1H, d, *J* = 8.0 Hz, Glc^1-H-1), 5.82 (1H, brs, Rha^1-H-1), 5.78 (1H, d, *J* = 5.0 Hz, Rib^1-H-1), 5.14 (1H, d, *J* = 7.5 Hz, Glc^4-H-1), 4.97 (1H, d, *J* = 8.0 Hz, Glc^2-H-1), 4.94 (1H, d, *J* = 8.0 Hz, Glc^3-H-1), 4.82 (1H, d, *J* = 6.5 Hz, Ara^1-H-1), 4.54 (1H, dd, *J* = 8.0, 6.5 Hz, Ara^1-H-2), 4.65 (1H, m, Rha^2-H-3), 4.29 (1H, m, Rib^1-H-4), 4.26 (1H, m, Glc^3-H-4), 1.68 (3H, d, *J* = 6.0 Hz, Rha^1-H-6), 1.51 (3H, d, *J* = 6.0 Hz, Rha^2-H-6); ^1H NMR data of the isoferuloyl moiety, see Table 4; ^{13}C NMR data, see Tables 2, 3, and 5; HRESIMS *m/z* 1859.8105 $[M + Na]^+$ (calcd for $C_{86}H_{132}O_{42}Na$, 1859.8085).

Clematochinenoside G (7): white, amorphous powder; $[\alpha]_D^{20}$ −66 (*c* 1.0, MeOH); UV (MeOH) λ_{max} (log ε) 233 (2.56), 299 (sh), 317 (4.03) nm; IR (KBr) ν_{max} 3421, 2928, 1715, 1636, 1062 cm^{-1}; ^1H NMR data (C_5D_5N, 500 MHz) δ 1.15 (3H, s, Me-27), 1.09 (3H, s, Me-24), 1.07 (3H, s, Me-26), 0.93 (3H, s, Me-25), 0.85 (3H, s, Me-29), 0.84 (3H, s, Me-30), 4.25 (1H, dd, *J* = 12.0, 5.0 Hz, H-3), 4.23 (1H, d, *J* = 12.0 Hz, H-23a), 3.88 (1H, d, *J* = 12.0 Hz, H-23b), 5.37 (1H, brs, H-12), 6.27 (1H, brs, Rha^2-H-1), 6.21 (1H, d, *J* = 8.0 Hz, Glc^1-H-1), 5.84 (1H, brs, Rha^1-H-1), 5.80 (1H, d, *J* = 5.0 Hz, Rib^1-H-1), 5.29 (1H, d, *J* = 7.5 Hz, Glc^4-H-1), 5.03 (1H, d, *J* = 6.5 Hz, Ara^1-H-1), 4.98 (1H, d, *J* = 7.5 Hz, Glc^2-H-1), 4.86 (1H, d, *J* = 8.0 Hz, Glc^3-H-1), 4.85 (1H, d, *J* = 8.0 Hz, Glc^5-H-1), 4.56 (1H, dd, *J* = 8.0, 6.5 Hz, Ara^1-H-2), 4.68 (1H, m, Rha^2-H-3), 4.31 (1H, m, Rib^1-H-4), 4.26 (1H, m, Glc^3-H-4), 4.15 (1H, m, Glc^4-H-3), 1.69 (3H, d, *J* = 6.0 Hz, Rha^1-H-6), 1.51 (3H, d, *J* = 6.0 Hz, Rha^2-H-6); ^1H NMR data of the

isoferuloyl moiety, see Table 4; ^{13}C NMR data, see Tables 2, 3, and 5; HRESIMS *m/z* 2037.8506 $[M + Na]^+$ (calcd for $C_{92}H_{142}O_{48}Na$, 2037.8563).

Acid Hydrolysis of Compounds 1−7. Each compound (5 mg) was hydrolyzed with 2 N aqueous CF_3COOH (10 mL) at 110 °C for 8 h in a sealed tube. The reaction mixture was diluted with H_2O (20 mL) and extracted with EtOAc (3 × 10 mL). The combined EtOAc extract was evaporated under reduced pressure and analyzed by TLC. Oleanolic acid was detected as a product of **6**, hederagenin as a product of **1**−**5** and **7**, and isoferulic acid as a product of **1** and **3**−**7**. The aqueous layer was repeatedly evaporated with MeOH under vacuum until the solvent was completely removed. The residue was dissolved in anhydrous pyridine (100 μL) and then mixed with a pyridine solution of L-cysteine methyl ester hydrochloride (100 μL). After warming at 60 °C for 1 h, hexamethyldisilazane (100 μL) and trimethylsilyl chloride (40 μL) were added, and the mixture was warmed at 60 °C for another 30 min. The mixture was filtered through a 0.45 μm membrane to remove the precipitate and analyzed by GC. Separations were carried out on a HP-5 column (28 m × 0.32 mm). Highly pure He was employed as carrier gas (1.0 mL/min flow rate), and the FID detector operated at 260 °C (column temp 180 °C). The retention times of the monosaccharide derivatives were as follows: L-Rha, 5.41 min; D-Glc, 11.63 min; D-Rib, 5.21 min; L-Ara, 4.97 min.

Mild Alkaline Hydrolysis of Compounds 1 and 4. Each compound (6 mg) was dissolved in 0.1 N KOH (2 mL) and allowed to hydrolyze at room temperature for 1 h. The reaction mixture was neutralized with dilute HCl and analyzed by HPLC. Isoferulic acid and **2** were detected as products of **1** (MeCN−H_2O, 24:76, 1.0 mL/min), and isoferulic acid and 3-*O*-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 were detected as products of **4** (MeCN−H_2O, 29:71, 1.0 mL/min).

Alkaline Hydrolysis of Compounds 4 and 6. Pure compounds **4** and **6** (10−15 mg each) were refluxed in 5% KOH solution (pH 12−13) at 90 °C for 1 h. The reaction mixtures were neutralized with 5% HCl solution and then concentrated to dryness. The residues were extracted with *n*-BuOH, and the organic layers of pure compounds were analyzed by NMR spectroscopy. Hydrolysis of **4** and **6** afforded two known compounds that were identified as CP_{10} and CP_9, respectively.[4]

In Vitro Anti-inflammatory Assay. Potential anti-inflammatory activity was determined according to the literature.[23] In a 78 μL reaction system, Tris-HCl buffer (0.1 M, pH 8.0), 4.4% heme final concentration, and COX-1 or COX-2 (Cayman, Ann Arbor, MI) were added successively. After a 5 min preincubation at 25 °C, *N*,*N*,*N*′,*N*′-tetramethyl-*p*-phenylenediamine and arachidonic acid (Cayman) were added. SC560 and NS398 (Cayman) were used as positive controls. After a second 5 min preincubation at 25 °C, enzyme activity was monitored on a Greiner 384 microplate reader (TECAN Safire2, Switzerland) by following the rate of change in absorbance (*A*) at 590 nm.

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Supporting Information Available: 1D and 2D NMR spectra for compounds **1**−**7** and structures of known compounds **8**−**24**. This material is available free of charge via the Internet at http://pubs.acs.org.

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