Nortriterpene Lactones from the Fruits of *Schisandra arisanensis*

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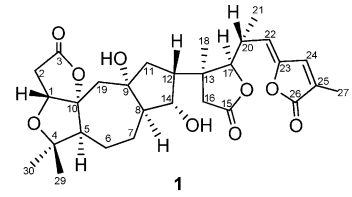
Fractionation of an acetone extract from the fruits of *Schisandra arisanensis* afforded five new nortriterpene lactones, compounds **1**−**5**, together with four known compounds, schindilactones D and E (**6** and **7**) and preschisanartanins A and B (**8** and **9**). Compound **1**, a wuweiziartane-type nortriterpenoid, possesses a new type of fused ring system with a γ-lactone ring between C-15 and C-17. Compounds **2**, **6**, and **7** may be categorized as schisanartane-type nortriterpenoids and compounds **3**−**5**, **8**, and **9** as preschisanartane-type nortriterpenoids. The structures of the new compounds were elucidated by 1D and 2D NMR spectroscopic data interpretation. The structure and relative configuration of **3** were supported by single-crystal X-ray diffraction analysis. The antiviral activity against HSV-1 and inhibitory effects on superoxide anion generation and elastase release by human neutrophils in response to FMLP/CB of compounds **1**−**9** were evaluated.

Schisandra (Schisandraceae) is native to East Asia, and several species of this plant genus have been used in traditional Chinese medicine for the treatment of cough, diabetes, diarrhea, and hepatitis, and as a general tonic agent.[1] From previous work, C_{18}-dibenzocyclooctadiene lignans have been found to be the major components of these medicinal plants. Biological studies have revealed that these natural products possess numerous effects such as hepatoprotective,[2] cancer protective,[3] antioxidative,[4] and antiviral activities.[5] Due to their notable medicinal properties, plants belonging to the family Schisandraceae have been studied extensively, and in recent years, several novel triterpene lactones were isolated and reported.[6−10] These secondary metabolites are highly oxygenated, and some representatives have been found to possess some anti-HIV activity.[11]

Schisandra arisanensis Hayata is an endemic species growing in the mountainous areas of Taiwan. Its stems and fruits have been used in folklore medicine for the treatment of arthritis, diabetes, headache, and hypertension.[12] Previously, 11 lignans have been isolated from the plant and their structures identified.[13] However, triterpenoids from *S. arisanensis* have not been reported before. Herein, we report five new nortriterpenoids, compounds **1**−**5**, and four known substances (**6**−**9**) from this species, as well as their evaluation against HSV-1 in a plaque reduction assay and their inhibitory effects on superoxide anion generation and elastase release.



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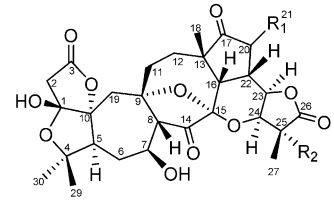
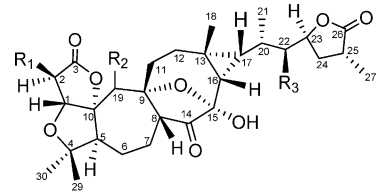


2 R_1 = α-CH_3, R_2 = OH
6 R_1 = β-CH_3, R_2 = H
7 R_1 = α-CH_3, R_2 = H



3 R_1 = H, R_2 = β-OH, R_3 = OH
4 R_1 = OH, R_2 = β-OH, R_3 = OH
5 R_1 = H, R_2 = α-OH, R_3 = OAc
8 R_1 = H, R_2 = β-OH, R_3 = OAc, Δ^{24}
9 R_1 = H, R_2 = β-OH, R_3 = OAc

Results and Discussion

Sephadex LH-20 and HPLC column chromatography afforded five new nortriterpene lactones, **1**−**5**, together with four known nortriterpenes, schindilactones D and E (**6** and **7**)[14,15] and preschisanartanins A and B (**8** and **9**).[8] The structures of the new compounds were established by interpretation of their spectroscopic data, especially 2D NMR. The relative configurations of compounds **3**, **6**, and **7** were determined unambiguously by X-ray crystallographic analysis. Since the ^{1}H and ^{13}C NMR data of compounds **6**−**9** were previously recorded in C_5D_5N, NMR spectroscopic data of **6**−**9** measured in $CDCl_3$ are thus listed in the Experimental Section for comparison.

Arisanlactone A (**1**) was obtained as a white powder and was assigned the molecular formula $C_{29}H_{38}O_9$ (Δ = 11), as deduced from HRESIMS (*m*/*z* 553.2413 $[M + Na]^+$). The IR spectrum showed the presence of hydroxy (3512 cm^{-1}), ester (1768 cm^{-1}), and C−O (1064 cm^{-1}) functionalities. The ^{1}H NMR data of **1** (Table 1) exhibited four methyl singlets (δ 1.21, 1.35, 1.38, and 2.02), one methyl doublet (δ 1.26, J = 6.6 Hz), two olefinic methine singnals (δ 5.11 and 7.03), and three oxygen-bearing methine signals (δ 4.30, 4.44, and 4.47). The ^{13}C NMR (Table 2) and DEPT spectra of **1** showed 29 carbon signals, consisting of three ester

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Table 1. ^{1}H NMR Spectroscopic Data (400 MHz) of Compounds **1**−**5**[a]

position	**1**[b]	**2**[c]	**3**[b]	**4**[b]	**5**[b]
1	4.30 brs		4.71 d (5.5)	4.49 s	4.73 d (5.6)
OH-1			2.53 d (18.3)		
2	2.68 (overlapped)	3.07 d (17.9)	2.97 dd (5.5, 18.3)	4.26 s	2.53 d (18.0)
		3.13 d (17.9)	2.31 m		2.95 dd (5.6, 18.0)
5	2.38 m	2.64 m	1.56 m	2.31 m	2.33 m
6	1.55 m	2.28 m	1.90 m	1.53 m	1.54 m
			1.76 m	2.00 m	1.94 m
7	1.80 m	4.58 m	1.99 m	1.73 m	1.77 m
	1.90 m		2.53 m	2.00 m	1.92 m
8	2.22 m	2.86 d (9.6)	1.23 m	2.58 m	2.52 m
11	1.58 m	1.65 m	2.32 m	1.26 m	1.23 m
	1.95 m	2.25 m		2.28 m	2.35 m
12	2.50 m	1.62 m	1.27 m	1.37 m	1.31 m
		1.80 m	1.92 m	1.83 m	2.08 m
14	4.44 m				
16	2.09 m	2.86 d (9.6)	1.17 d (10.6)	1.11 (overlapped)	1.00 d (8.6)
	2.30 m				
17	4.47 (overlapped)		0.67 t (10.6)	0.71 t (10.0)	0.72 t (9.7)
18	1.38 s	0.96 s	1.05 s	1.06 s	0.94 s
19	1.79 (overlapped)	2.45 d (16.4)	3.70 d (7.8)	4.08 s	3.70 d (7.6)
	2.03 (overlapped)	2.94 d (16.4)			
OH-19			4.39 d (7.8)		4.15 d (7.6)
20	3.36 m	2.80 m	2.97 m	2.66 m	3.04 m
21	1.26 d (6.6)	1.56 d (7.6)	1.24 (overlapped)	1.29 d (6.4)	1.30 (overlapped)
22	5.11 d (10.7)	3.45 dd (8.7, 10.9)	3.33 d (9.3)	3.28 d (9.9)	4.71 (overlapped)
23		5.42 s	4.63 d (8.4)	4.81 d (5.1)	4.77 (overlapped)
24	7.03 s	5.02 s	2.03 m	1.96 m	2.05 m
			2.46 m	2.56 m	2.16 m
25			2.94 m	2.91 m	2.71 m
27	2.02 s	1.63 s	1.23 (overlapped)	1.25 d (7.2)	1.26 d (7.1)
29	1.21 s	1.41 s	1.21 s	1.20 s	1.21 s
30	1.35 s	1.25 s	1.30 s	1.30 s	1.30 s
2′					2.13 s

[a] Chemical shifts are in ppm; *J* values in Hz are in parentheses. [b] Measured in $CDCl_3$. [c] Measured in C_5D_5N.

Table 2. ^{13}C NMR Spectroscopic Data (100 MHz) of Compounds **1**−**5**[a]

position	**1**[b]	**2**[c]	**3**[b]	**4**[b]	**5**[b]
1	80.1 CH	108.8 C	78.7 CH	83.1 CH	78.7 CH
2	35.3 CH_2	43.0 CH_2	34.7 CH_2	71.9 CH	34.7 CH_2
3	173.9 C	173.1 C	175.6 C	174.2 C	175.7 C
4	84.7 C	84.6 C	84.3 C	85.4 C	84.2 C
5	59.3 CH	58.1 CH	61.8 CH	61.2 CH	61.9 CH
6	21.1 CH_2	36.7 CH_2	23.2 CH_2	23.1 CH_2	23.2 CH_2
7	22.9 CH_2	69.0 CH	26.8 CH_2	27.1 CH_2	26.8 CH_2
8	56.3 CH	60.5 CH	55.5 CH	55.0 CH	55.6 CH
9	79.9 C	81.8 C	82.4 C	82.4 C	82.4 C
10	99.4 C	97.3 C	97.9 C	97.7 C	97.8 C
11	42.8 CH_2	41.5 CH_2	37.4 CH_2	37.6 CH_2	37.5 CH_2
12	41.2 CH	30.4 CH_2	24.6 CH_2	25.1 CH_2	24.7 CH_2
13	44.5 C	49.8 C	25.4 C	25.8 C	25.6 C
14	75.5 CH	209.4 C	213.1 C	213.0 C	213.4 C
15	176.6 C	98.4 C	97.9 C	98.3 C	98.2 C
16	42.8 CH_2	45.6 CH	29.6 CH	29.2 CH	29.5 CH
17	93.5 CH	221.5 C	33.6 CH	35.2 CH	34.0 CH
18	23.7 CH_3	25.9 CH_3	28.0 CH_3	28.1 CH_3	28.1 CH_3
19	40.5 CH_2	40.8 CH_2	59.8 CH	71.1 CH	70.1 CH
20	33.5 CH	41.2 CH	30.2 CH	31.0 CH	29.9 CH
21	20.1 CH_3	12.4 CH_3	19.0 CH_3	18.0 CH_3	18.4 CH_3
22	113.6 CH	33.3 CH	79.4 CH	79.3 CH	79.8 CH
23	147.8 C	74.6 CH	78.5 CH	78.0 CH	77.2 CH
24	138.1 CH	75.7 CH	33.1 CH_2	32.4 CH_2	32.7 CH_2
25	130.2 C	76.9 C	34.6 CH	34.8 CH	34.3 CH
26	170.3 C	177.1 C	182.6 C	183.4 C	181.1 C
27	10.7 CH_3	17.6 CH_3	16.4 CH_3	16.2 CH_3	16.6 CH_3
29	21.7 CH_3	25.4 CH_3	21.5 CH_3	21.5 CH_3	21.5 CH_3
30	28.6 CH_3	29.9 CH_3	28.4 CH_3	28.1 CH_3	28.4 CH_3
1′					170.4 C
2′					21.6 CH_3

[a] Assignments were made using HMQC and HMBC techniques. [b] Measured in $CDCl_3$. [c] Measured in C_5D_5N.

carbonyls (δ 170.3, 173.9, and 176.6), two olefinic methines (δ 113.6 and 138.1), two olefinic quaternary carbons (δ 130.2 and 147.8), three oxymethines (δ 75.5, 80.1, and 93.5), three oxygenated quaternary carbons (δ 79.9, 84.7, and 99.4), four aliphatic methines (δ 33.5, 41.2, 56.3, and 59.3), six aliphatic methylenes (δ 21.1, 22.9, 35.3, 40.5, 42.8, and 42.8), and five methyls (δ 10.7, 20.1, 21.7, 23.7, and 28.6). The above findings accounted for five of the 11 degrees of unsaturation, indicating that **1** is a hexacyclic nortriterpenoid with three γ-lactone functionalities. Comparison of the ^{1}H and ^{13}C NMR data of **1** with those of related nortriterpenoids showed partial similarities (rings A−C). However, the remaining data were insufficient to determine its skeleton. Thus, the planar structure of **1** was elucidated by assembling two partial structures, **X** and **Y** (Figure 1).

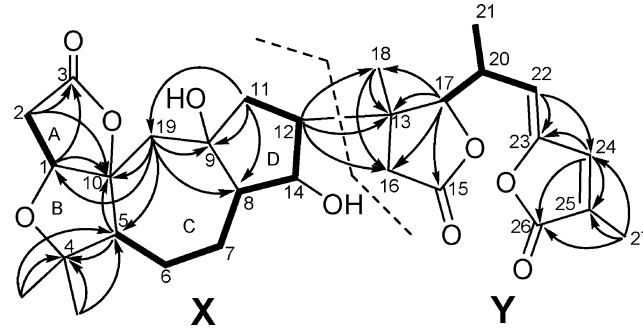

Figure 1. Key COSY (bold bonds) and HMBC correlations (arrows) of **1**.

The COSY spectrum (Figure 1) of **1** exhibited two typical proton spin systems of H-1 (δ 4.30)/H_2-2 (δ 2.68) and H-5 (δ 2.38)/H_2-6 (δ 1.55)/H_2-7 (δ 1.90)/H-8 (δ 2.22). In the HMBC spectrum, the H-1 and H_2-2 resonances also showed correlations to C-3 (δ 173.9) and C-10 (δ 99.4), revealing the presence of a γ-lactone (ring A). In addition, the HMBC correlations of H_2-19 (δ 1.79, 2.03) with C-1 (δ 80.1), C-5 (δ 59.3), C-8 (δ 56.3), C-9 (δ 79.9), and C-10 and of H-5 with C-10 and C-4 (δ 84.7), along with the proton spin system from H-5 to H-8, were used to establish rings B and C. Another proton spin system was found from correlations between H-8/H-14 (δ 4.44)/H-12 (δ 2.50)/H_2-11 (δ 1.58, and 1.95) in the COSY spectrum. Combining this finding with the HMBC correla-

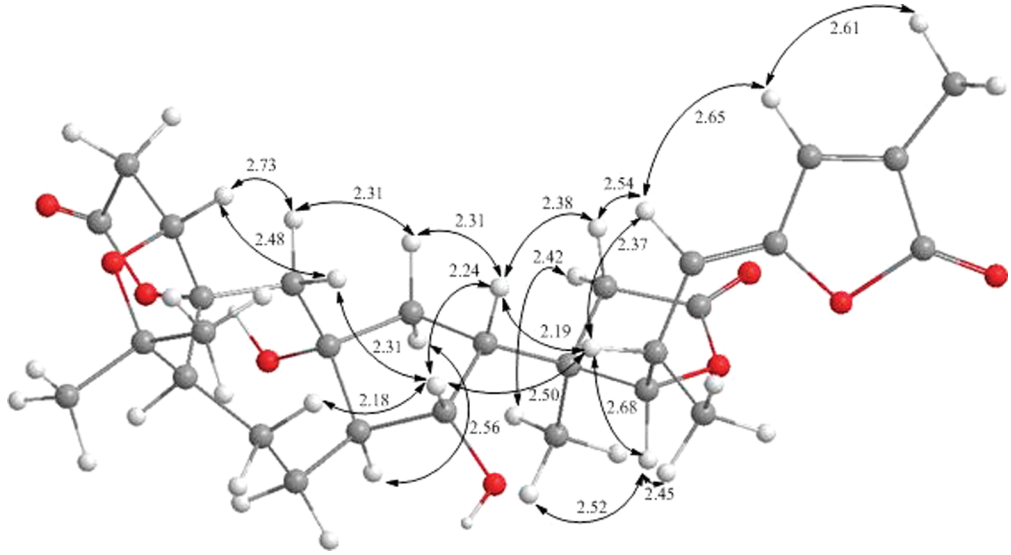

Figure 2. Key NOESY correlations of **1** and their corresponding interatomic distance [Å].

tions from H-11 to C-8, C-9, and C-19 pointed to the presence of a fourth ring (ring D). Moreover, the HMBC correlations of both H_3-29 (δ 1.21) and H_3-30 (δ 1.35) with C-4 and C-5 indicated two geminal methyl groups attached at C-4. Thus, partial structure **X** was established. For the partial structure **Y**, a proton sequence between H-17 (δ 4.47)/H-20 (δ 3.36)/H_3-21 (δ 1.26)/H-22 (δ 5.11) was observed in the COSY spectrum. Moreover, the HMBC correlations of H-24 (δ 7.03) with C-23 (δ 147.8), C-25 (δ 130.2), and C-26 (δ 170.3) and of H_3-27 (δ 2.02) with C-24 (δ 138.1), C-25, and C-26 revealed substructure **Y** to contain an α-methyl,β-unsaturated-γ-lactone ring system. This ring system was connected to the above proton sequence through the HMBC correlations of H-22 with C-23 and C-24 (δ 138.1). The remaining four carbons (C-13, C-15, C-16, and C-18) were connected from the key HMBC correlations of H-17 with C-13 (δ 44.5), C-15 (δ 176.6), C-16 (δ 42.8), and C-18 (δ 23.7) and of H_3-18 (δ 1.38) with C-13 and C-16, which indicated the presence of a β-methyl-γ-lactone ring in partial structure **Y**. Finally, partial structures **X** and **Y** were combined as a result of the HMBC correlations from H-12 to C-13, C-16, and C-18. This 2D NMR spectroscopic analysis was used to identify **1** as a wuweiziartane-type nortriterpenoid with a new type of fused ring system.

The relative configuration of **1** was determined on the basis of NOESY correlations (Figure 2) and confirmed by MM2 minimum energy calculations. On considering previous reports of nortriterpenoids in *Schisandra chinensis*,[6] H-5 is often α-oriented. The NOESY spectrum of **1** showed correlations of H_3-30 and H-5, indicating that they are on the same face of the molecule. Conversely, NOESY cross-peaks were found between H_3-29/H-1/H_2-19/H-11β/H-12/H-14, H-12/H-16β, suggesting that these protons are on the β-face. In turn, the NOESY cross-peaks of H-11α/H-8, H-16α/H_3-18/H-17 indicated the α-orientation of these protons. The NOESY results were quite consistent with the interatomic distances between protons in the molecular model of **1**, as shown in Figure 2. The CD spectrum of **1** exhibited a negative Cotton effect at 310 nm similar to that of schintrilactone A.[13] This result indicated that the absolute configuration at C-20 should be *R*. A biogenetic pathway for compound **1** is proposed and illustrated in Scheme S1, Supporting Information. Nortriterpenes of wuweiziartane type may be derived from schiartane-type precusors.[6,15] Thus, the hydroxy group at C-9 could be assigned with an α-orientation. On the basis of the above discussion, the relative configuration of **1** was established as shown.

Arisanlactone B (**2**) was assigned a molecular formula of $C_{29}H_{36}O_{12}$ and 12 degrees of unsaturation, as deduced from the

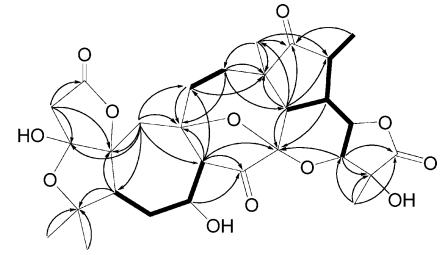

Figure 3. Key COSY (bold bonds) and HMBC correlations (arrows) of **2**.

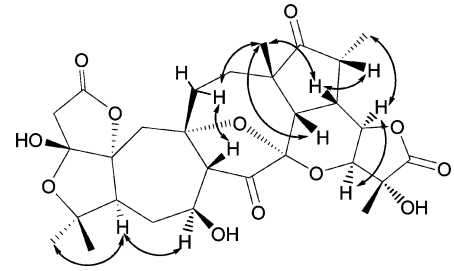

Figure 4. Key NOESY correlations of **2**.

HRESIMS (*m*/*z* 599.2103 $[M + Na]^+$) and ^{13}C NMR spectra. The IR absorption bands revealed the presence of hydroxy (3431 cm^{-1}), ester (1770 cm^{-1}), ketone (1732 cm^{-1}), and C−O (1089 cm^{-1}) functionalities in **2**. The ^1H and ^{13}C NMR data of this compound were similar to those of **6** and **7**, suggesting it to be a schisanartane-type nortriterpene dilactone. On comparing the ^{13}C NMR spectrum of these three compounds, it was noted that the C-21 chemical shifts of **2**, **6**, and **7** were δ 12.4, 15.1, and 12.2, respectively. This suggested that the C-20 configuration of **2** is the same as **7**, and the Me-21 group was assigned as α-oriented. Compound **2** was found to possess an additional hydroxy group, when compared to **7**. This hydroxy group was assigned at C-25, as evidenced from the multiplicity of H_3-27 (singlet in **2**, doublet in **7**) and from the chemical shift of C-25 (δ 76.9 in **2**, δ 42.1 in **7**). This assignment was confirmed by the HMBC correlations (Figure 3) from H-24 (δ 5.02) and H_3-27 (δ 1.63) to C-25. The relative configuration of **2** was determined on the basis of a NOESY experiment (Figure 4) and comparison with the X-ray structures of **6** and **7** (Figure 5). The NOESY correlations of H-5 and H-7 indicated that the hydroxy group attached at C-7 is on the β-face of the molecule. In turn, NOE cross-peaks between H-8/H-11/H_3-18/H-16/H-22/H-20 re-

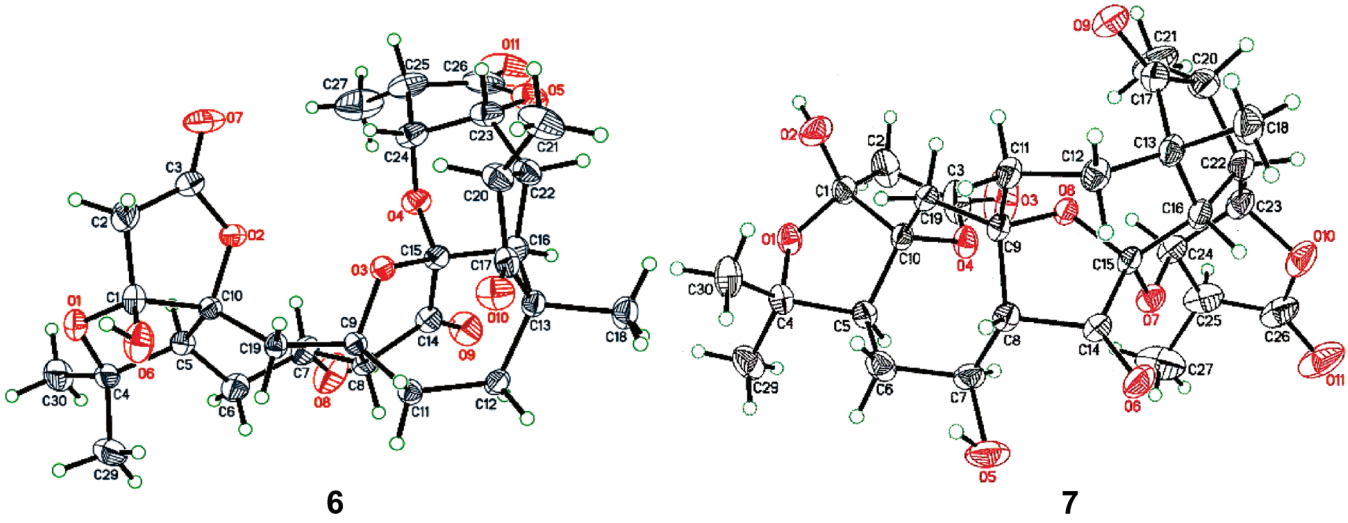

Figure 5. Perspective drawing of the X-ray structures of **6** and **7**.

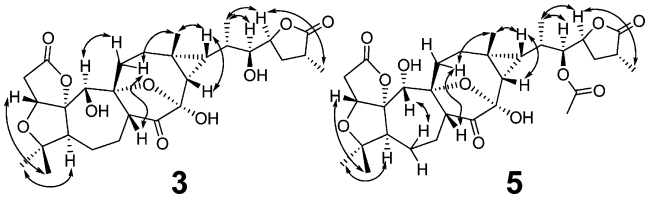

Figure 6. Key NOESY correlations of **3** and **5**.

vealed that these protons are on the β-face and H_3-21 is on the α-face, the same as **7**. In addition, the presence of NOESY correlations between H_3-21/H-23/H-24 and the absence of any NOESY correlation between H-24/H_3-27 proved the α-orientation of the hydroxy group attached at C-25. Therefore, structure **2** was assigned to arisanlactone B.

Arisanlactone C (**3**) exhibited the molecular formula $C_{29}H_{40}O_{10}$ and 10 degrees of unsaturation, as deduced from the HRESIMS (*m*/*z* 571.2520 $[M + Na]^+$). In the ^{1}H NMR spectrum, an upfield-shifted triplet (δ 0.67) was observed, suggesting the presence of a cyclopropane ring. In the ^{13}C NMR spectrum, a ketone (δ 213.1) and two ester carbonyls (δ 175.6 and 182.6) were observed, accounting for three of the 10 degrees of unsaturation, which indicated that **3** possesses a heptacyclic skeleton. From the interpretation of its spectroscopic data and the NMR similarities between **3** and preschisanartanin B (**9**), compound **3** could be categorized as being a preschisanartane-type nortriterpene. On comparing the NMR data of **3** with those of **9**, an acetyl group (δ_H 2.12; δ_C 170.6, 21.6) attached at C-22 in **9** was replaced by a hydroxy group in **3**. This was corroborated from the HMBC correlations from H_3-21 (δ 1.24) and H-23 (δ 4.63) to the oxygen-bearing carbon C-22 (δ 79.4). The NOESY spectrum of **3** (Figure 6) exhibited correlations between H-5/H-30, H-29/H-1, H-19/H-11/H-8, and H_2-11/H_3-18/H-17/H-16, indicating the same relative configuration as **9**. It was noted that the NOESY experiment could not be used to determine the configurations of C-15, C-20, C-22, and C-23, as well as C-25, due to σ-bond rotation. However, a prism crystal of **3** was obtained, and the results of single-crystal X-ray diffraction analysis (Figure 7) were used to determine the relative configuration of **3**. From all of these data, structure **3** was assigned to arisanlactone C.

Compound **4** was obtained as a colorless gum and found to possess the molecular formula $C_{29}H_{40}O_{11}$, as inferred from its HRESIMS (*m*/*z* 587.2465 $[M + Na]^+$). The similarities of the ^{1}H and ^{13}C NMR spectroscopic data of **4** and **3** suggested these compounds to be close analogues. In the ^{1}H NMR spectrum of **4**, the oxymethine signal of H-1 (δ 4.49) appeared as a singlet instead

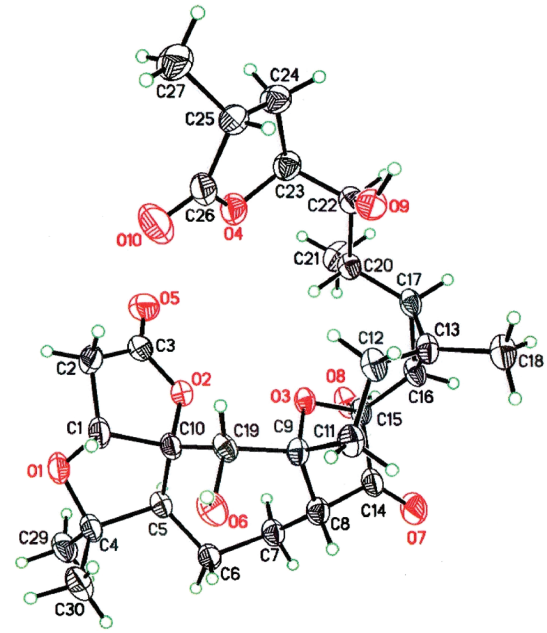

Figure 7. Perspective drawing of the X-ray structure of **3**.

of a doublet, which implied that a substituent is attached at C-2. In the ^{13}C NMR spectrum, **4** was observed as possessing an additional oxymethine and to lack an aliphatic methylene, in comparison to **3**. This finding suggested that there is an additional hydroxy group in **4**. The additional hydroxy group was assigned at C-2 by virtue of the HMBC correlations from H-1 to C-2 (δ 71.9) and from H-2 (δ 4.26) to C-1 (δ 83.1) and C-3 (δ 174.2). The relative configuration of **4** was determined on the basis of a NOESY experiment and the vicinal Karplus correlation. The results of a NOESY experiment were identical to **3**, suggesting the same configuration. The hydroxy group at C-2 was assigned with a β-orientation as a result of the absence of any NOESY correlation between H-1 and H-2 and the coupling constant between H-1 and H-2 ($J = 0$ Hz, $\theta = 90°$). On the basis of above interpretations, the structure of **4** was established as 2β-hydroxyarisanlactone C.

The HRESIMS indicated a molecular formula for arisanlactone D (**5**) identical to that of **9**. The IR and ^{1}H and ^{13}C NMR data were quite similar to those of **9**, suggesting that **5** is an isomer of **9**. In the COSY and HMBC spectra of **5**, the correlations were comparable to those observed from **9**, indicating the same planar structure. To determine the difference between **5** and **9**, a NOESY

experiment (Figure 6) was carried out. The only difference was that the NOESY spectrum of **5** exhibited cross-peaks between H-19 and H-6, but in the case of **9** cross-peaks were found between H-19 and H-12. Accordingly, H-19 was assigned as being β-oriented and opposite that of **9**. Thus, the structure of compound **5** (arisanlactone D) was determined as shown.

The isolated nortriterpenoids **1**−**9** were tested for their in vitro inhibitory activity against the HSV-1 virus. Acyclovir was used as a positive control. Only compounds **2** and **7** showed weak activity (25% and 30% inhibition, respectivly) at 100 μM. Also, inhibitory effects of compounds **1**−**9** were evaluated on superoxide anion generation and elastase release by human neutrophils in response to FMLP/CB at a concentration of 10 μg/mL. As a result, compounds **2** and **4** showed mild anti-inflammatory effects (22.24 ± 4.91% and 18.47 ± 2.20%, respectively) on elastase release. Genistein was used as a standard compound (51.60 ± 5.89%).

Experimental Section

General Experimental Procedures. Meting points were recorded on a Büchi B540 melting point apparatus. Optical rotations were recorded on a JASCO DIP-1000 polarimeter. IR spectra were taken on a HORIBA FT-720 spectrophotometer. The ^{1}H and ^{13}C NMR spectra as well as 2D NMR spectra (COSY, HMQC, HMBC, and NOESY) were recorded in $CDCl_3$ on a Bruker AVX NMR spectrometer operating at 400 MHz for ^{1}H and 100 MHz for ^{13}C using the $CDCl_3$ solvent peak as internal standard (δ_H 7.265, δ_C 77.0 ppm). Low-resolution EIMS were recorded on a VG Quattro 5022 mass spectrometer. HRESIMS were measured on a JEOL HX 110 mass spectrometer. LiChrospher Si 60 (5 μm, 250−10, Merck) and LiChrospher 100 RP-18e (5 μm, 250-10, Merck) were used for NP-HPLC and RP-HPLC (Hitachi, L-6250; flow rate 2 mL/min, UV detection at 254 nm), respectively.

Plant Material. The fruits of *Schisandra arisanensis* were collected in Nan-Tou County, Taiwan, in March 2008. The plant material was identified by one of the authors (C.T.C.). A voucher specimen (code no. TP 98-1) has been deposited at the School of Pharmacy, National Taiwan University, Taipei, Taiwan.

Extraction and Isolation. The freeze-dried fruits (1.38 kg) of *S. arisanensis* were ground and extracted three times with acetone at room temperature and then concentrated under reduced pressure to afford a crude extract (300 g). This crude extract was partitioned between EtOAc and H_2O (1:1) to obtain an EtOAc-soluble layer. After evaporating the organic solvent, the EtOAc residue (61 g) was partitioned between *n*-hexane−MeOH−H_2O (4:3:1) to afford a MeOH−H_2O extract. The MeOH−H_2O extract (35 g) was subjected to passage over a Si gel flash column (*n*-hexane−EtOAc, 40:1 to 0:1) to furnish fractions A−H. Fraction H (2.9 g) was separated on a Sephadex LH-20 column eluted with MeOH to afford fraction H1 (1.4 g), which was further chromatographed on Si gel (*n*-hexane−CH_2Cl_2−MeOH, 100:20:1 to 0:10:1) to furnish subfractions H1D−H1G. Subfraction H1D was subjected to RP-HPLC (MeOH−H_2O, 5:3) and further purified by NP-HPLC (*n*-hexane−EtOAc, 21:19) to give **5** (10.3 mg) and **9** (29.3 mg).[8] Subfraction H1E was subjected to RP-HPLC (MeOH−H_2O, 11:9) to afford **8** (22.6 mg)[8] and another subfraction, H1E3. Subfraction H1E3 was separated by NP-HPLC (*n*-hexane−CH_2Cl_2−MeOH, 75:425:7) to yield **1** (21.3 mg) and **3** (16.4 mg). Separation of subfraction H1F by RP-HPLC (MeOH−H_2O, 1:1) yielded **7** (65.4 mg)[15] and two subfractions, H1F1 and H1F2. Subfraction H1F1 was purified by normal-phase HPLC (*n*-hexane−CH_2Cl_2−MeOH, 12:68:1) to obtain **6** (15.6 mg).[14] Fraction H1F2 was separated by NP-HPLC (*n*-hexane−CH_2Cl_2−MeOH, 48:32:1) and further purified by RP-HPLC (MeCN−H_2O, 2:3) to produce **4** (7.9 mg). Subfraction H1G was purified by RP-HPLC (MeOH−H_2O, 17:23) and again purified by RP-HPLC (MeCN−H_2O, 2:3) to furnish **2** (9.9 mg).

Arisanlactone A (1): off-white, amorphous powder; $[\alpha]_D^{25}$ 62 (*c* 0.1, MeOH); CD IR (neat) ν_{max} 3512, 2971, 2929, 1768, 1176, 1064 cm^{-1}; ^{1}H NMR ($CDCl_3$) and ^{13}C NMR ($CDCl_3$) spectroscopic data, see Tables 1 and 2, respectively; HRESIMS *m*/*z* 553.2413 [M + Na]$^+$ (calcd for $C_{29}H_{38}O_9Na$, 553.2413).

Arisanlactone B (2): off-white, amorphous powder; $[\alpha]_D^{25}$ 24 (*c* 0.1, MeOH); IR (neat) ν_{max} 3431, 2974, 2933, 1770, 1732, 1458, 1375, 1089, 1018 cm^{-1}; ^{1}H NMR (C_5D_5N) and ^{13}C NMR (C_5D_5N) spectroscopic data, see Tables 1 and 2, respectively; HRESIMS *m*/*z* 599.2103 [M + Na]$^+$ (calcd for $C_{29}H_{36}O_{12}Na$, 599.2099).

Arisanlactone C (3): colorless, prism; mp: > 200 °C (dec); $[\alpha]_D^{25}$ 2 (*c* 0.1, MeOH); IR (neat) ν_{max} 3465, 2954, 2924, 2854, 1755, 1458, 1375, 1068 cm^{-1}; ^{1}H NMR ($CDCl_3$) and ^{13}C NMR ($CDCl_3$) spectroscopic data, see Tables 1 and 2, respectively; HRESIMS *m*/*z* 571.2520 [M + Na]$^+$ (calcd for $C_{29}H_{40}O_{10}Na$, 571.2519).

Crystal Data: orthorhombic system, space group $P2(1)2(1)2(1)$, a = 9.1827(5) Å, b = 18.1165(13) Å, c = 18.6612(8) Å, V = 3104.4(3) Å^3, Z = 4, d = 1.319 Mg/m^3. A crystal of dimensions 0.20 × 0.15 × 0.10 mm was used for measurement on a Siemens SMART CCD XRD. The total number of independent reflections measured was 8558, of which 5161 were observed [R(int) = 0.0340]. Completeness to θ = 67.99°: 99.8%, absorption correction: semiempirical from equivalents. Max. and min. transmission: 1.00000 and 0.79414. The structure was solved by direct methods and refined by full-matrix least-squares on F^2. Final R indices [$I > 2\sigma(I)$]: R1 = 0.0634, wR2 = 0.1578. The final X-ray diagram is shown in Figure 7.

2β-Hydroxyarisanlactone C (4): colorless, gum; $[\alpha]_D^{25}$ 32 (*c* 0.1, MeOH); IR (neat) ν_{max} 3406, 2974, 2931, 2871, 1761, 1209, 1072 cm^{-1}; ^{1}H NMR ($CDCl_3$) and ^{13}C NMR ($CDCl_3$) spectroscopic data, see Tables 1 and 2, respectively; HRESIMS *m*/*z* 587.2465 [M + Na]$^+$ (calcd for $C_{29}H_{40}O_{11}Na$, 587.2463).

Arisanlactone D (5): off-white, amorphous powder; $[\alpha]_D^{25}$ 44 (*c* 0.1, MeOH); IR (neat) ν_{max} 3452, 2964, 2927, 1778, 1749, 1456, 1373, 1074, 924 cm^{-1}; ^{1}H NMR ($CDCl_3$) and ^{13}C NMR ($CDCl_3$) spectroscopic data, see Tables 1 and 2, respectively; HRESIMS *m*/*z* 613.2623 [M + Na]$^+$ (calcd for $C_{31}H_{42}O_{11}Na$, 613.2619).

Schindilactone D (6): colorless prisms; mp: > 200 °C (dec); $[\alpha]_D^{25}$ 47 (*c* 0.1, MeOH); IR (neat) ν_{max} 3430, 2973, 2933, 1770, 1731, 1458, 1375, 1090 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) 3.24 (1H, brs, OH-1), 2.75 (1H, d, J = 18.5 Hz, H-2), 2.98 (1H, d, J = 18.5 Hz, H-2), 2.33 (1H, m, H-5), 1.72 (1H, m, H-6), 1.88 (1H, m, H-6), 3.91 (1H, t, J = 9.3 Hz, H-7), 2.32 (1H, overlapped, H-8), 1.56 (1H, m, H-11), 1.87 (1H, m, H-11), 1.44 (1H, m, H-12), 1.92 (1H, m, H-12), 2.56 (1H, d, J = 7.0 Hz, H-16), 1.03 (3H, s, H-18), 2.19 (1H, d, J = 16.4 Hz, H-19), 2.29 (1H, d, J = 16.4 Hz, H-19), 2.43 (1H, m, H-20), 1.21 (3H, overlapped, H-21), 2.67 (1H, m, H-22), 4.33 (1H, brs, H-23), 4.78 (1H, brs, H-24), 2.85 (1H, m, H-25), 1.21 (3H, overlapped, H-27), 1.23 (3H, s, H-29), 1.33 (3H, s, H-30); ^{13}C NMR ($CDCl_3$, 100 MHz) 108.3 (s, C-1), 43.2 (t, C-2), 173.1 (s, C-3), 85.1 (s, C-4), 57.9 (d, C-5), 33.0 (t, C-6), 68.1 (d, C-7), 59.1 (d, C-8), 80.7 (s, C-9), 95.8 (s, C-10), 42.0 (t, C-11), 30.4 (t, C-12), 50.1 (s, C-13), 212.0 (s, C-14), 98.5 (s, C-15), 44.5 (d, C-16), 220.7 (s, C-17), 25.9 (q, C-18), 39.3 (t, C-19), 44.4 (d, C-20), 15.1 (q, C-21), 39.8 (d, C-22), 74.9 (d, C-23), 68.6 (d, C-24), 41.8 (d, C-25), 177.6 (s, C-26), 7.7 (q, C-27), 24.5 (q, C-29), 29.0 (q, C-30); FABMS *m*/*z* 583.2 [M + Na]$^+$.

Crystal Data: orthorhombic system, space group $P2(1)$, a = 8.9185(2) Å, b = 9.8009(2) Å, c = 15.7643(3) Å, V = 1366.89(5) Å^3, Z = 2, d = 1.401 Mg/m^3. A crystal of dimensions 0.20 × 0.15 × 0.10 mm was used for measurement on a Siemens SMART CCD XRD. The total number of independent reflections measured was 4686, of which 3522 were observed [R(int) = 0.0184]. Completeness to θ = 67.97°: 100% absorption correction: semiempirical from equivalents. Max. and min. transmission: 1.00000 and 0.90319. The structure was solved by direct methods and refined by full-matrix least-squares on F^2. Final R indices [$I > 2\sigma(I)$]: R1 = 0.0543, wR2 = 0.1548. The final X-ray diagram is shown in Figure 5.

Schindilactone E (7): colorless prisms; mp: > 200 °C (dec); $[\alpha]_D^{25}$ 146 (*c* 0.1, MeOH); IR (neat) ν_{max} 3444, 2973, 2925, 1768, 1732, 1456, 1379, 1081 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) 3.22 (1H, brs, OH-1), 2.70 (1H, d, J = 18.2 Hz, H-2), 2.96 (1H, d, J = 18.2 Hz, H-2), 2.31 (1H, m, H-5), 1.74 (1H, m, H-6), 1.88 (1H, m, H-6), 3.91 (1H, t, J = 9.2 Hz, H-7), 2.29 (1H, d, J = 9.2 Hz, H-8), 1.53 (1H, m, H-11), 2.03 (1H, m, H-11), 1.45 (1H, m, H-12), 1.90 (1H, m, H-12), 2.61 (1H, d, J = 8.0 Hz, H-16), 1.09 (3H, s, H-18), 2.14 (1H, d, J = 16.4 Hz, H-19), 2.34 (1H, d, J = 16.4 Hz, H-19), 2.75 (1H, m, H-20), 1.21 (3H, overlapped, H-21), 3.26 (1H, m, H-22), 4.46 (1H, brs, H-23), 4.67 (1H, brs, H-24), 2.90 (1H, m, H-25), 1.20 (3H, d, J = 6.6 Hz, H-27), 1.25 (3H, s, H-29), 1.33 (3H, s, H-30); ^{13}C NMR ($CDCl_3$, 100 MHz) 108.3 (s, C-1), 42.7 (t, C-2), 171.9 (s, C-3), 85.4 (s, C-4), 57.7 (d, C-5), 33.2 (t, C-6), 68.4 (d, C-7), 59.5 (d, C-8), 81.0 (s, C-9), 96.0 (s, C-10), 41.6 (t, C-11), 29.7 (t, C-12), 49.7 (s, C-13), 211.7 (s, C-14), 97.8 (s, C-15), 45.2 (d, C-16), 221.7 (s, C-17), 26.0 (q, C-18), 39.4 (t, C-19), 40.6 (d, C-20), 12.2 (q, C-21), 32.8 (d, C-22), 74.3 (d, C-23),

70.9 (d, C-24), 42.1 (d, C-25), 177.4 (s, C-26), 7.7 (q, C-27), 24.8 (q, C-29), 29.2 (q, C-30); FABMS *m/z* 583.2 $[M + Na]^+$.

Crystal Data: orthorhombic system, space group *P*2(1), a = 8.79700(10) Å, b = 9.90100(10) Å, c = 15.9022(2) Å, V = 1364.34(3) Å^3, Z = 2, d = 1.365 Mg/m^3. A crystal of dimensions 0.20 × 0.15 × 0.10 mm was used for measurement on a Siemens SMART CCD XRD. The total number of independent reflections measured was 11 445, of which 4838 were observed [R(int) = 0.0175]. Completeness to θ = 68.00°: 100% absorption correction: semiempirical from equivalents. Max. and min. transmission: 1.00000 and 0.94555. The structure was solved by direct methods and refined by full-matrix least-squares on F^2. Final R indices [$I > 2\sigma(I)$]: R1 = 0.0358, wR2 = 0.1077. The final X-ray diagram is shown in Figure 5.

Preschisanartanin A (8): off-white, amorphous powder; $[\alpha]_D^{25}$ 3 (*c* 0.1, MeOH); IR (neat) ν_{max} 3444, 2971, 2931, 2871, 1753, 1635, 1456, 1373, 1070 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) 4.73 (1H, d, J = 5.2 Hz, H-1), 2.51 (1H, d, J = 18.3 Hz, H-2), 2.93 (1H, dd, J = 5.2, 18.3 Hz, H-2), 2.33 (1H, m, H-5), 1.30 (1H, m, H-6), 2.03 (1H, m, H-6), 1.84 (1H, m, H-7), 1.94 (1H, m, H-7), 2.56 (1H, overlapped, H-8), 1.21 (1H, m, H-11), 2.32 (1H, m, H-11), 1.53 (1H, m, H-12), 1.94 (1H, m, H-12), 1.03 (1H, d, J = 8.6 Hz, H-16), 0.68 (1H, t, J = 9.8 Hz, H-17), 0.93 (3H, s, H-18), 3.71 (1H, d, J = 6.9 Hz, H-19), 4.63 (1H, d, J = 6.9 Hz, OH-19), 3.16 (1H, m, H-20), 1.35 (3H, d, J = 6.3 Hz, H-21), 4.82 (1H, d, J = 9.0 Hz, H-22), 5.12 (1H, s, H-23), 6.94 (1H, s, H-24), 1.87 (1H, s, H-27), 1.22 (3H, s, H-29), 1.30 (3H, s, H-30), 1.97 (3H, s, H-2′); ^{13}C NMR ($CDCl_3$, 100 MHz) 78.6 (d, C-1), 34.7 (t, C-2), 175.8 (s, C-3), 84.3 (s, C-4), 61.8 (d, C-5), 25.0 (t, C-6), 26.7 (t, C-7), 55.5 (d, C-8), 82.5 (s, C-9), 98.2 (s, C-10), 37.5 (t, C-11), 23.2 (t, C-12), 26.0 (s, C-13), 213.7 (s, C-14), 98.0 (s, C-15), 29.7 (d, C-16), 34.1 (d, C-17), 28.1 (q, C-18), 69.9 (d, C-19), 30.2 (d, C-20), 18.5 (q, C-21), 75.3 (d, C-22), 80.9 (d, C-23), 146.5 (d, C-24), 130.8 (s, C-25), 174.8 (s, C-26), 10.4 (q, C-27), 21.5 (q, C-29), 28.2 (q, C-30), 170.5 (s, C-1′), 21.0 (q, C-2′); FABMS *m/z* 611.2 $[M + Na]^+$.

Preschisanartanin B (9): off-white, amorphous powder; $[\alpha]_D^{25}$ 77 (*c* 0.1, MeOH); IR (neat) ν_{max} 3448, 2971, 2931, 2871, 1755, 1074, 924 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) 4.76 (1H, d, J = 5.3 Hz, H-1), 2.50 (1H, d, J = 18.0 Hz, H-2), 2.94 (1H, dd, J = 5.3, 18.0 Hz, H-2), 2.28 (1H, m, H-5), 1.52 (1H, m, H-6), 1.94 (1H, m, H-6), 1.77 (1H, m, H-7), 1.92 (1H, m, H-7), 2.54 (1H, overlapped, H-8), 1.20 (1H, m, H-11), 2.31 (1H, m, H-11), 1.27 (1H, m, H-12), 1.96 (1H, m, H-12), 0.97 (1H, d, J = 8.7 Hz, H-16), 0.70 (1H, t, J = 10.4 Hz, H-17), 0.93 (3H, s, H-18), 3.66 (1H, d, J = 7.5 Hz, H-19), 4.52 (1H, d, J = 7.5 Hz, OH-19), 3.00 (1H, m, H-20), 1.29 (3H, overlapped, H-21), 4.80 (1H, overlapped, H-22), 4.82 (1H, overlapped, H-23), 2.03 (1H, m, H-24), 2.16 (1H, m, H-24), 2.69 (1H, m, H-25), 1.24 (1H, d, J = 7.2 Hz, H-27), 1.21 (3H, s, H-29), 1.29 (3H, s, H-30), 2.12 (3H, s, H-2′); ^{13}C NMR ($CDCl_3$, 100 MHz) 78.6 (d, C-1), 34.6 (t, C-2), 175.8 (s, C-3), 84.2 (s, C-4), 61.9 (d, C-5), 23.2 (t, C-6), 26.8 (t, C-7), 55.6 (d, C-8), 82.5 (s, C-9), 97.9 (s, C-10), 37.4 (t, C-11), 24.6 (t, C-12), 25.6 (s, C-13), 213.8 (s, C-14), 98.2 (s, C-15), 29.4 (d, C-16), 34.0 (d, C-17), 28.0 (q, C-18), 69.9 (d, C-19), 29.8 (d, C-20), 18.2 (q, C-21), 79.7 (d, C-22), 77.2 (d, C-23), 32.7 (t, C-24), 34.2 (d, C-25), 181.1 (s, C-26), 16.6 (q, C-27), 21.5 (q, C-29), 28.3 (q, C-30), 170.6 (s, C-1′), 21.6 (q, C-2′); FABMS *m/z* 613.2 $[M + Na]^+$.

Anti HSV-1 Assay, Cell Culture, and Virus. Vero cells were cultured in minimal essential medium (MEM; Gibco, Grand Island, NY) supplemented with 10% fetal calf, serum (FCS; Hyclone, Logan, UT), 100 U/mL penicillin, and 100 μg/mL streptomycin and incubated at 37 °C in a 5% CO_2 incubator. To prepare a stock of HSV-1 (KOS strain, VR-1493, ATCC), Vero cells were infected by HSV-1 at a multiplicity of infection of three plaque-forming units (PFU)/cell, harvested at 24 h post-infection, and centrifuged at 1500*g* (centrifuge 5810 R, Eppendrof) at 4 °C for 20 min. The supernatant was collected and stored at −70 °C for use.

Plaque Reduction Assay. The assay used followed a procedure described previously.[16] Vero cells (3.5 × 10^5/dish) were incubated with 100 PFU of HSV-1, and various compounds (100 μM) or acyclovir (2.5 μM) was added to the cells. The viruses were adsorbed for 1 h at 37 °C, and 1% methylcellulose was added to each well. After 5 days, the virus plaques formed in HeLa cells were counted by crystal violet staining. The activities of various compounds and acyclovir for inhibition of plaque formation were calculated.

Anti-inflammatory Assays, Inhibitory Effect on Superoxide Anion Generation and Elastase Release by Human Neutrophils. Neutrophils were obtained by means of dextran sedimentation and Ficoll centrifugation. Superoxide generation and elastase release were carried out according to a procedure described previously.[17] Superoxide anion production was assayed by monitoring the superoxide dismutase-inhibitable reduction of ferricytochrome *c*. Elastase release experiments were performed using MeO-Suc-Ala-Ala-Pro-Valp- nitroanilide as the elastase substrate.

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Supporting Information Available: Biogenetic pathway for compound **1** and NMR spectra of compounds **1**−**5**. This information is available free of charge via the Internet at http://pubs.acs.org.

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