Triterpenes and Lignans from *Artemisia caruifolia* and Their Cytotoxic Effects on Meth-A and LLC Tumor Cell Lines

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Received August 29, 2000; accepted October 31, 2000

One new triterpene, 3β-hydroxy-29-norcycloart-24-one (1), and four new lignans, caruilignans (2—5), together with six known compounds were isolated from the aerial part of *Artemisia caruifolia* BUCH.-HAM. ex TOXB. Their structures were determined by various spectroscopic means. Most of the isolated lignans were moderately cytotoxic to Meth-A cells with ED_{50} values of 5—10 μg/ml, but not to Lowis lung carcinoma (LLC) cells. An oxime derivative of 1 showed more potent cytotoxic activity against Meth-A and LLC cells than the original triterpene 1.

Key words *Artemisia caruifolia*; caruilignan; cytotoxicity; lignan; 3β-hydroxy-29-norcycloart-24-one

The aerial part of *Artemisia caruifolia* is a traditional Chinese medicine used for the treatment of infectious diseases.[1] In the course of our search for biologically active compounds from natural sources, we have reported some guaiane dimers and a new germacranolide from this plant.[2] Further investigation of this plant has led to the isolation and identification of a new triterpene (**1**) and four new lignans (**2**—**5**).

Results and Discussion

Repeated chromatography of a $CHCl_3$ soluble part of the MeOH extract of *Artemisia caruifolia* has led to the isolation of compounds **1**—**5**, together with 3β-hydroxycycloart-24-one,[3] syringaresinol,[4] sesamin,[5] diayangambin,[6] sesartemin[7] and artemitin.[8] The identification of the known compounds was performed by comparing their physical and spectral data with those reported. Compounds **1**—**5** are new natural products, and their structures were determined as follows.

The molecular formula of **1** was determined to be $C_{29}H_{48}O_2$ based on the high resolution electron impact (HR-EI)MS. The ^{1}H-NMR spectrum of **1** was similar to that of 3β-hydroxycycloart-24-one, except that a methyl signal disappeared and the H-3 signal became multiplet. The multiplet H-3 suggested that C-4 was not a quaternary carbon. Considering the molecular formula, it was deduced that compound **1** should possess a norcycloartanol skeleton, in which only one methyl group is attached at C-4. This was confirmed by the analysis of its 2D NMR spectra. The ^{13}C-NMR spectrum showed the presence of a carbonyl group (δ 215.5). Since the carbonyl signal showed a long-range correlation with two methyl protons (H_3-26 and H_3-27) in the heteronuclear multiple bond coherence (HMBC) spectrum, the oxo group was placed at C-24. In the nuclear Overhauser effect spectroscopy (NOESY) spectrum, one of the β oriented cyclopropane protons at δ 0.38 (Ha-19) was correlated with one of the methylene protons at δ 0.58 (Ha-6), indicating that Ha-6 is oriented at the β-face. The remaining germinal proton (Hb-6) at δ 1.68 was correlated significantly with a methyl proton (H_3-28) at C-4, indicating that the methyl group is oriented at the α-face. In the double-resonance experiment, on irradiation of a signal at δ 1.19, which was assigned as H-4 by ^{1}H–^{1}H shift correlation spectroscopy (COSY), the multiplet signal of H-3 at δ 3.22 became a double-doublet signal with coupling constants of 4.8 and 10.2 Hz. Therefore, H-3 was determined to be an axial proton (α-face) and the orientation of a hydroxy group at C-3 was consequently assigned to be β. The stereochemistry of **1** was further confirmed by comparing its ^{13}C-NMR data with cycloeucalenol,[9] whose structure differed from **1** only by a 24-methylene group in the side chain. From the above findings, the structure of **1** was determined as 3β-hydroxy-29-norcycloart-24-one.

Compound **2** (named caruilignan A) was assigned the molecular formula $C_{25}H_{32}O_9$ by HR-EI-MS. The ^{1}H-NMR spectrum showed two aromatic proton signals at δ 6.71 and 6.56 (each two protons). In addition, there were seven methoxy signals at δ 3.00—4.00, one of which appeared at δ 3.00, more obviously up-field than the other methoxy signals at δ >3.5, suggesting the presence of an aliphatic methoxyl in its structure. The 2D NMR spectra including ^{1}H–^{1}H COSY and ^{13}C–^{1}H COSY revealed that **2** possessed a furofuranoid lig-

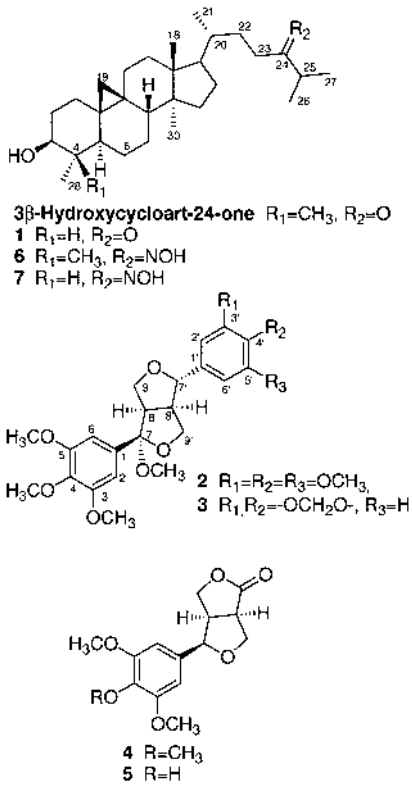

Chart 1

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nan skeleton related to diayangambin, with a methoxyl group substituted at the furan ring. In the HMBC spectrum, the methoxyl proton signal at δ 3.00 had a long-range correlation with an acetal carbon at δ 110.3, which was assigned to be C-7, due to the presence of a long-range correlation with the aromatic proton signal at δ 6.71 (H-2 and H-6). Therefore, the aliphatic methoxyl should be attached at C-7. The NOESY spectrum revealed cross-peaks between H-7′ and one of the H-9 signals at δ 3.09 (H-9β), and between H-7′ and one of the H-9′ signals at δ 4.07 (H-9′β). On the other hand, appreciable cross-peaks were observed between H-9′α and the methoxy protons at C-7, as well as H-9′α and H-8′. The relative stereochemistry of C-7, C-8, C-8′ and C-7′ was

Table 1. ^{13}C-NMR Data of Triterpenes Isolated from *A. caruifolia* and Their Derivatives in $CDCl_3$

Position	3β-Hydroxycycloart-24-one (75 MHz)	**1** (75 MHz)	**6** (100 MHz)	**7** (75 MHz)
1	32.2	31.1	32.1	31.1
2	30.6	35.1	30.3	35.1
3	79.0	77.3	78.3	77.3
4	40.7	44.8	40.4	44.8
5	47.3	43.6	47.1	43.6
6	21.4	24.9	21.1	24.9
7	26.7	25.5	26.4	25.4
8	48.2	47.1	47.9	47.1
9	20.2	23.8	20.1	23.8
10	26.3	29.8	26.1	29.8
11	26.3	27.2	26.0	27.2
12	33.1	33.1	33.6	33.9
13	45.5	45.6	45.3	45.6
14	49.0	49.1	48.8	49.1
15	35.8	35.6	35.5	35.6
16	28.3	28.3	28.0	28.3
17	52.5	52.4	51.9	52.1
18	18.4	18.1	18.0	18.1
19	30.2	27.5	29.8	27.5
20	36.0	36.0	36.7	37.0
21	18.6	18.4	18.0	18.4
22	30.4	30.4	31.9	32.4
23	37.8	37.8	23.6	23.8
24	215.6	215.5	166.8	166.7
25	41.1	41.1	32.8	33.1
26	18.4*	18.6*	19.3*	20.3*
27	18.7*	18.7*	19.9*	20.4*
28	14.3	14.7	13.9	14.7
29	25.7		25.4	
30	19.6	19.4	20.0	19.4

* Assignments in the same column may be exchangeable.

Table 3. ^{13}C-NMR Data of Compounds **2**—**5** in $CDCl_3$

Position	**2** (75 MHz)	**3** (125 MHz)	**4** (100 MHz)	**5** (75 MHz)
1	137.6	137.6	132.1	127.6
2	103.9	103.9	102.8	102.6
3	153.1	153.1	153.6	147.3
4	133.2	133.2	137.5	134.2
5	153.1	153.1	153.6	147.3
6	103.9	103.9	102.8	102.6
7	110.3	110.2	84.2	84.4
8	57.0	56.8	45.9	46.2
9	70.6	70.3	71.0	71.1
1′	137.3	134.9		
2′	103.0	106.7		
3′	153.4	147.9		
4′	136.6	147.2		
5′	153.4	108.1		
6′	103.0	119.6		
7′	88.0	87.8	178.6	178.8
8′	53.1	52.9	43.6	43.8
9′	69.8	69.5	68.4	68.6
3-OCH_3	56.3[a)]	56.2	56.2	56.6
4-OCH_3	61.0[b)]	60.8	60.9	
5-OCH_3	56.3[a)]	56.2	56.2	56.6
7-OCH_3	49.0	48.8		
3′-OCH_3	56.4[a)]			
4′-OCH_3	61.1[b)]			
5′-OCH_3	56.4[a)]			
3′,4′-OCH_2O–		101.0		

a, *b*) Assignments with the same superscripts may be exchangeable.

Table 2. ^{1}H-NMR Spectral Data of Compounds **2**—**5** in $CDCl_3$

Position	**2** (300 MHz)	**3** (500 MHz)	**4** (400 MHz)	**5** (300 MHz)
2	6.71 (s)	6.71 (br s)	6.50 (s)	6.50 (s)
6	6.71 (s)	6.71 (br s)	6.50 (s)	6.50 (s)
7			4.94 (d, 5.6)	4.96 (d, 5.5)
8	3.31 (dt, 8.5, 8.5)	3.29 (dt, 9.0, 9.0)	3.38 (m)	3.38 (m)
9α	3.86 (overlapped)	3.82 (dd, 9.0, 9.0)	3.98 (dd, 6.8, 9.5)	3.97 (dd, 6.6, 9.5)
β	3.09 (dd, 9.0, 9.0)	3.07 (dd, 9.0, 9.0)	4.52 (d, 9.5)	4.52 (d, 9.5)
2′	6.56 (s)	6.85 (d, 1.5)		
5′		6.77 (d, 8.0)		
6′	6.56 (s)	6.81 (dd, 1.5, 8.0)		
7′	4.48 (d, 6.6)	4.46 (d, 6.0)		
8′	3.04 (m)	3.00 (m)	3.40 (m)	3.38 (m)
9′α	4.12 (dd, 9.0, 6.6)	4.09 (dd, 7.0, 9.0)	3.83 (overlapped)	3.86 (dd, 3.9, 9.5)
β	4.07 (dd, 2.2, 9.0)	4.03 (dd, 2.0, 9.0)	4.10 (dd, 8.3, 10.0)	4.09 (dd, 9.5, 8.2)
3-OCH_3	3.89[a)]	3.89	3.86	3.89
4-OCH_3	3.84[b)]	3.87	3.85	
5-OCH_3	3.89[a)]	3.89	3.86	3.89
7-OCH_3	3.00	2.98		
3′-OCH_3	3.88[a)]			
4′-OCH_3	3.88[b)]			
5′-OCH_3	3.88[a)]			
3′,4′-OCH_2O–		5.95		

a, *b*) Assignments with the same superscripts may be exchangeable.

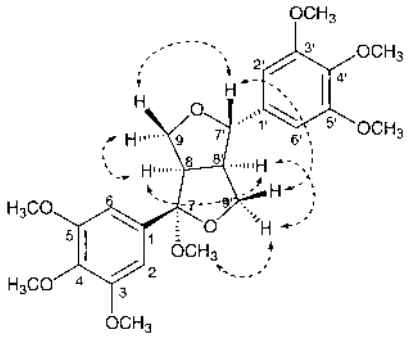

Fig. 1. NOE Correlations in NOESY Spectrum of Compound **2**

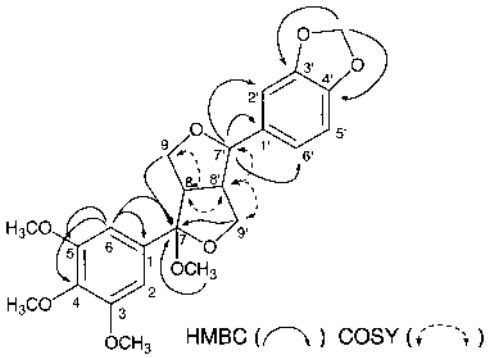

Fig. 2. Significant HMBC and COSY Correlations Observed for Compound **3**

determined as shown in Fig. 1.

Compound **3** was assigned the molecular formula $C_{23}H_{26}O_8$ by HR-EI-MS. In the ^{1}H-NMR spectrum, **3** showed four methoxyl signals at δ 2.98, 3.87, 3.89 and 3.89, as well as one methylenedioxy signal at δ 5.95. The signal for the equivalent aromatic protons at δ 6.71 (H-2 and H-6) was exactly the same as that of **2**, suggesting that one phenyl ring in the structure of **3** possessed the same substitution pattern as that of **2**. Other signals in the low field range were a doublet signal at δ 6.77 (J=8.0 Hz), a double-doublet signal at δ 6.81 (J=1.5, 8.0 Hz), and a doublet signal at δ 6.85 (J=1.5 Hz), each being integrated for one proton, suggesting the presence of a 1,3,4-trisubstituted phenyl group. The signals in the high field closely resembled those of **2**, indicating that the substitution pattern of the furofuran part of compounds **3** and **2** are the same. The planar structure of **3** was finally confirmed by ^{1}H–^{1}H COSY and HMBC experiments (Fig 2). The stereostructure was established by a NOESY experiment in the same manner as in the case of **2**.

The molecular formula of **4** was determined to be $C_{15}H_{18}O_6$ based on the HR-EI-MS spectrum. The ^{1}H-NMR spectrum showed a singlet aromatic proton signal integrated for two protons at δ 6.50 and three methoxyl signals at δ 3.85 and 3.86 (2×OCH_3) and the ^{13}C-NMR spectrum indicated the presence of one carbonyl group. The ^{1}H-detected multiple quantum coherence (HMQC) spectrum revealed the presence of two methylenes and five methines in the structure of **4**. The ^{1}H–^{1}H COSY spectrum indicated the connection of the methylenes and methines as shown in Fig. 3. HMBC correlations were observed between a proton signal at δ 4.52 (H-9) and a carbonyl carbon signal at δ 178.6, between the signals of H-9 and a methine carbon at 43.6 (C-8′), as well as

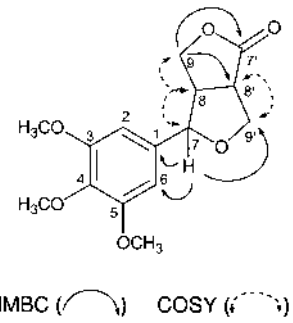

Fig. 3. Significant HMBC and COSY Correlations Observed for Compound **4**

between the signals of H-7 and C-9′, which led to the partial structure in the aliphatic part of **4**. The HMBC correlations observed between the signals of H-7 and C-1, H-7 and C-6, enabled the connection of C-7 to C-1 and thus the planar structure of **4** was established as shown in Fig. 3. The stereochemistry of **4** was determined by a NOESY experiment. One of the methylene protons at δ 3.98 (Ha-9) was correlated with H-8 (α-oriented) more significantly than the other geminal proton (Hb-9), and was consequently determined to be α-oriented. Since H-7 was appreciably correlated with Ha-9 (α) but not with Hb-9 (β), the orientation of H-7 should be α.

Compound **5** had the molecular formula of $C_{14}H_{16}O_6$ as established by HR-EI-MS. Its ^{1}H-NMR spectrum was superimposable over that of **4** except for the signals of methoxyl groups. Compound **5** showed only one signal integrated for two methoxyl groups at δ 3.89. Because the molecular weight of **5** was 14 mass units less than that of **4**, it is most probable that **5** possesses a hydroxy group in the structure, instead of a methoxy group in **4**. Since aromatic protons were observed as only one singlet signal in the ^{1}H-NMR spectrum, **5** should possess a symmetrically substituted phenyl ring and thus the hydroxy group was located at C-4. The 2D NMR spectra further confirmed the planar structure of **5** and its stereochemistry was found to be the same as **4** by a NOESY experiment. It is worth noting that a stereoisomer of **5**, zhebeiresinol, has been isolated from *Fritillaria thunbergii*.[10] The stereochemistry of zhebeiresinol had been established by X-ray analysis as C1–C7 α-bond. In the present experiment, we compared the ^{1}H- and ^{13}C-NMR spectra of **5** with those of zhebeiresinol, measured in the same solvent, dimethyl sulfoxide (DMSO)-d_6. It was found that the NMR data of the two compounds were quite different from each other, and thus the stereochemistry of **5** was proposed as shown in Chart 1.

The cytotoxic activity of the isolated compounds, including the sesquiterpenes reported previously,[2] were tested using Meth-A (sarcoma) and LLC (Lowis lung carcinoma) cell lines. None of the sesquiterpenes showed cytotoxic activity on either cell lines. 3β-hydroxylcycloart-24-one was found to be cytotoxic to both cell lines, while five lignans, **2**, **3**, **4**, sesamin and sesartemin, were found to be cytotoxic only against the Meth-A cell line. A flavonoid compound, artemitin, was also cytotoxic to the Meth-A cell line. As many nitrogen-containing compounds were more biologically active in general, we prepared two oximes of 3β-hydroxylcycloart-24-one and **1**. These oximes showed cytotoxic

Table 4. 50% Growth Inhibition (ED_{50}) of Triterpenes and Lignans from *A. caruifolia* and Related Triterpene Derivatives against Meth-A and LLC Cell Lines

Compound	*vs*. Meth-A (μg/ml)	*vs*. LLC (μg/ml)
3β-hydroxycycloart-24-one	9.0	9.0
1	>10	>10
2	6.5	>10
3	4.9	>10
4	10	>10
5	>10	>10
6	9.5	7.4
7	5.5	6.4
Syringaresinol	>10	>10
Sesamin	6.0	>10
Diayangambin	>10	>10
Sesartemin	9.7	>10
Artemitin	4.3	>10

effects on both cell lines tested, with their EC_{50} values being lower than those of the corresponding original compounds.

Experimental

General Optical rotations were measured with a Jasco DIP-360 automatic polarimeter. UV spectra were measured with a Shimadzu UV-VIS recording spectrophotometer. IR spectra were measured with a Jasco FT/IR-230 infrared spectrometer. ^{1}H- and ^{13}C-NMR spectra were measured with a Varian Gemini 300 (1H, 300 MHz; ^{13}C, 75 MHz) or Varian Unity 500 (^{1}H, 500 MHz; ^{13}C, 125 MHz) or Jeol JNA-LA 400WB-FT (^{1}H, 400 MHz; ^{13}C, 100 MHz) spectrometer, the chemical shifts being represented as ppm with TMS as an internal standard. Electron impact (EI) MS were measured with a Jeol JMS-AX 505 HAD mass spectrometer at an ionization voltage of 70 eV.

Cytotoxicity Assays The *in vitro* Meth-A tumor cell assay was carried out according to the procedure by Geran *et al*.,[11] and LLC cells, by a sulforhodamin B (SRB) method.[12] For the SRB assay, a cell suspension in the culture medium (100000 cells/ml) was inoculated to each well of 96-well microtiter plate. One day after plating, a control plate at time zero was made. In the presence or absence of compounds, the cells were incubated for a further 48 h in a CO_2 incubator. Cells were fixed with 50 μl of 20% trichloroacetic acid (TCA) solution for 1 h at 4 °C and plates were washed 5 times with tap water and air-dried. A 50 μl of SRB solution (0.4% in 1% acetic acid) was added and the staining was done at room temperature for 30 min. The residual dye was washed out with 1% acetic acid and air-dried. To each well, Tris buffer solution (10 mM, pH 10.5) was added. Optical density (OD) was measured with a microtiter plate reader at 540 nm. Growth inhibition was calculated according to the previous method. Briefly, the value ($T_s - T_0$), in which the OD at time-zero (T_0) is subtracted from that of a treated well (T_s), was divided by calculated value of untreated control (T_c). The 50% growth inhibition (ED_{50}) was calculated by Probit method.[13]

Plant Material The aerial part of *Artemisia caruifolia* Buch.-Ham. ex Roxb. was purchased from Yaocaigongyingzhan of Huhhot, Inner Mongolia of the People's Republic of China in September of 1998. The plant material was identified by Dr. Katsuko Komatsu of the Analytical Research Center for Ethnomedicines, Institute of Natural Medicine, Toyama Medical and Pharmaceutical University. A voucher specimen (TMPW No. 19154) is stored at the Museum of Materia Medica, Toyama Medical and Pharmaceutical University, Japan.

Extraction and Isolation The plant (3.0 kg) was extracted with MeOH under reflux (20 l×3, each 2 h). After the solvent was evaporated under vacuum, the MeOH extract (190 g) was partitioned with $CHCl_3$ and H_2O. The $CHCl_3$-soluble part (96 g) was chromatographed on silica gel eluted with *n*-hexane–AcOEt 7 : 3—0 : 1 (fr. 1—4) and then AcOEt–EtOH–H_2O 6 : 2 : 1 (fr. 5) to give the respective fractions in yields of 56.3 g, 9.6 g, 3.1 g, 5.5 g and 18 g.

A portion of fr. 1 (30 g) was chromatographed on an ODS column with 60—100% MeOH to give 3β-hydroxylcycloart-24-one (15 mg, 0.00094%), **1** (10 mg, 0.00063%) and three other sub-fractions. A portion of each of the sub-fractions were purified by preparative-TLC (SiO_2, benzene–acetone 9 : 1) to give sesamin (5 mg, 0.00093%), sesartemin (200 mg, 0.065%), **3** (4 mg, 0.00025%) and artemitin (50 mg, 0.016%). Fraction 2 was chromatographed on an ODS column with 40—60% MeOH; the 40% MeOH eluted sub-fraction was then purified by HPLC to give **4** (4 mg, 0.00025%), the 60% MeOH eluted sub-fraction was re-chromatographed on SiO_2 gel eluted with benzene–acetone 95 : 5 to obtain **2** (18 mg, 0.0011%) and diayangambin (50 mg, 0.0031%). Fraction 3 was chromatographed on an ODS column with 50—60% MeOH and further purified by HPLC to give syringaresinol (10 mg, 0.00063%) and **5** (3 mg, 0.00019%).

3β-Hydroxy-29-norcycloart-24-one (**1**): Amorphous powder; $[\alpha]_D^{24}$ +42.6° (c=0.19, $CHCl_3$); IR (KBr) ν_{max} 3440 (OH), 2965, 2940, 2920, 2868, 1718 (C=O), 1470, 1458, 1130, 1103, 1042, 1002 cm^{-1}; ^{1}H-NMR ($CDCl_3$, 300 MHz) δ: 0.14 (1H, d, J=4.5 Hz, Hb-19), 0.38 (1H, d, J=4.5 Hz, Ha-19), 0.58 (1H, m, Ha-6), 0.86 (3H, d, J=6.5 Hz, H-21), 0.86 (3H, s, H-30), 0.96 (3H, s, H-18), 0.98 (3H, d, J=6.0 Hz, H-28), 1.09 (6H, d, J=6.5 Hz, H-26, 27), 1.19 (overlapped, H-4), 1.68 (1H, m, Hb-6), 2.38 (1H, m, Ha-23), 2.49 (1H, m, Hb-23), 2.62 (1H, septet, J=6.5 Hz, H-25), 3.22 (1H, m, H-3); ^{13}C-NMR: see Table 1; EI-MS m/z 428 $[M]^+$ (60), 413 (100), 410 (80), 395 (90), 302 (30); HR-EI-MS m/z 428.3651 (Cacld for $C_{29}H_{48}O_2$ $[M]^+$, 428.3656).

Caruilignan A (**2**): Amorphous powder; $[\alpha]_D^{24}$ +61.6° (c=0.83, $CHCl_3$); UV ($CHCl_3$) λ_{max} (ε): 243 (7300), 270 (1600); IR (KBr) ν_{max}: 2940, 2880, 2840, 1590, 1510, 1470, 1408, 1340, 1230, 1115 cm^{-1}; ^{1}H-NMR: see Table 2; ^{13}C-NMR: see Table 3; EI-MS m/z 476 $[M]^+$ (100), 279 (25), 248 (100), 217 (90), 195 (100), 181 (100); HR-EI-MS m/z 476.2069 (Calcd for $C_{25}H_{32}O_9$ $[M]^+$, 476.2047).

Caruilignan B (**3**): Amorphous powder; $[\alpha]_D^{24}$ +78.8° (c=0.51, $CHCl_3$); UV ($CHCl_3$) λ_{max} (ε): 242 (9900), 283 (5300); IR (KBr) ν_{max}: 2960, 2940, 2880, 1590, 1505, 1445, 1408, 1340, 1250, 1235, 1130, 1040 cm^{-1}; ^{1}H-NMR: see Table 2; ^{13}C-NMR: see Table 3; EI-MS m/z 430 $[M]^+$ (6), 399 (40), 248 (28), 226 (45), 195 (100), 135 (100); HR-EI-MS m/z 430.1606 (Calcd for $C_{23}H_{26}O_8$ $[M]^+$, 430.1628).

Caruilignan C (**4**): Amorphous powder; $[\alpha]_D^{24}$ +144.1° (c=0.18, $CHCl_3$); UV ($CHCl_3$) λ_{max} (ε): 241 (5300), 272 (1100); IR (KBr) ν_{max}: 2940, 2880, 2840, 1770 (C=O), 1590, 1510, 1460, 1420, 1370, 1330, 1235, 1185, 1130, 1090, 1110 cm^{-1}; ^{1}H-NMR: see Table 2; ^{13}C-NMR: see Table 3; EI-MS m/z 294 $[M]^+$ (100), 279 (10), 196 (20), 181 (60), 263 (10), 169 (20); HR-EI-MS m/z 294.1129 (Calcd for $C_{15}H_{18}O_6$ $[M]^+$, 294.1103).

Caruilignan D (**5**): Amorphous powder; $[\alpha]_D^{24}$ +126.0° (c=0.13, $CHCl_3$); UV ($CHCl_3$) λ_{max} (ε): 241 (3300), 272 (800); IR (KBr) ν_{max}: 3440 (OH), 2920, 2860, 1770 (C=O), 1615, 1520, 1460, 1375, 1220, 1115, 1090, 995 cm^{-1}; ^{1}H-NMR: see Table 2; ^{13}C-NMR: see Table 3; EI-MS m/z 280 $[M]^+$ (100), 181 (100), 167 (90); HR-EI-MS m/z 280.0915 (Calcd for $C_{14}H_{16}O_6$ $[M]^+$, 280.0947).

Preparation of Oximes of 3β-Hydroxycycloart-24-one and 1 A triterpene (7 mg) and hydroxyamine hydrochloride (5 mg) in pyridine (0.5 ml) was heated for 2 h at 50 °C. After cooling to room temperature, the reaction mixture was concentrated under vacuum and then purified with RP-18 (MeOH–H_2O, 80—100%) to yield **6** (3 mg) or **7** (3 mg).

24-Hydroxyiminocycloart-3-ol (**6**): Amorphous powder; ^{1}H-NMR ($CDCl_3$, 400 MHz) δ: 0.33 (1H, d, J=4.2 Hz, Hb-19), 0.56 (1H, d, J=4.2 Hz, Ha-19), 0.81 (3H, s, H-29), 0.89 (3H, s, H-30), 0.94 (3H, d, J=6.4 Hz, H-21), 0.97 (3H, s, H-18), 0.97 (3H, s, H-28), 1.10 (6H, d, J=6.6 Hz, H-26, 27), 2.19 (1H, m, Ha-23), 2.38 (1H, m, Hb-23), 2.49 (1H, septet, J=6.5 Hz, H-25), 3.28 (1H, dd, J=4.4, 11.2 Hz, H-3); ^{13}C-NMR: see Table 1; EI-MS m/z 457 $[M]^+$ (40), 439 (100), 317 (80).

24-Hydroxyimino-29-norcycloart-3-ol (**7**): Amorphous powder; $[\alpha]_D^{24}$ +35.4° (c=0.35, $CHCl_3$); ^{1}H-NMR ($CDCl_3$, 300 MHz) δ: 0.14 (1H, d, J=4.0 Hz, Hb-19), 0.38 (1H, d, J=4.0 Hz, Ha-19), 0.89 (3H, s, H-30), 0.94 (3H, d, J=6.6 Hz, H-21), 0.98 (3H, s, H-18), 0.98 (3H, d, J=6.0 Hz, H-28), 1.10 (6H, d, J=6.6 Hz, H-26, 27), 2.17 (1H, m, Ha-23), 2.35 (1H, m, Hb-23), 2.48 (1H, septet, J=6.5 Hz, H-25), 3.21 (1H, m, H-3); ^{13}C-NMR: see Table 2; EI-MS m/z 443 $[M]^+$ (55), 425 (100), 410 (100), 317 (60).

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