Three New *E*-Secoursane Triterpenoid Saponins from the Leaves of *Ilex dunniana*

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Three new triterpenoid saponins with an 18,19-secours-13(18)-ene skeleton, dunnianaolactones A – C (**1** – **3**, resp.), together with nine known compounds *i.e.*, the ursane-type triterpene saponin **4**, the two benzofuran lignans **5** and **6**, five flavonoid glycosides, and 4-hydroxybenzoic acid, were isolated from the leaves of *Ilex dunniana* LEVL. (Aquifoliaceae). Their structures were elucidated by means of spectroscopic and chemical methods. The configuration of dunnianaolactone A (**1**) was further confirmed by X-ray crystal-structure analysis.

Introduction. – Species of the genus *Ilex*, from the family Aquifoliaceae, have a wide range of pharmacological activities and are generally used to treat cardiovascular disease, fever, cough, and inflammation [1]. *Ilex dunniana* LEVL., an evergreen tree, is distributed in the west of Hubei Province, southwest of Sichuan Province, east of Yunnan Province, and northeast of Guizhou Province in P. R. China [1]. It is locally used as a beverage for the treatment of sore throats and arthritis, and the prevention of wound infections through external application in the northwest regions of Hubei Province. However, there are no reports on the chemical composition of this plant.

In the present study, three new 18,19-secoursane glycosides, namely dunnianaolactones A – C (**1** – **3**), were isolated from the leaves of *I. dunniana*, together with nine known compounds, including one ursane-type triterpenoid saponin (see **4**), two benzofuran lignans (see **5** and **6**), five flavonoid glycosides, and 4-hydroxybenzoic acid (*Fig. 1*). The structures of the new compounds were elucidated by means of spectroscopic and chemical methods. To confirm the configuration of the new 18,19-secourane glycosides, an X-ray diffraction analysis of **1** was also carried out.

Results and Discussion. – The leaves of *I. dunniana* were extracted with 90% EtOH. The EtOH extract was partitioned with petroleum ether, AcOEt, and BuOH. Three new compounds **1** – **3** and nine known compounds were isolated from the BuOH-soluble fraction.

Compound **1** was obtained as colorless, acicular crystals. The HR-ESI-MS (negative-ion mode) suggested that the molecular formula was $C_{47}H_{76}O_{18}$. The ^{13}C-NMR (including DEPT) spectrum (*Table 1*) showed 47 C-atom signals, with 30 attributed to the aglycone part, including one C=O (δ(C) 181.5), one trisubstituted C=C (δ(C) 150.2 and 117.2), and seven Me groups (δ(C) 8.1, 17.2, 17.4, 18.2, 18.9, 22.4, and 28.6). The ^{1}H-NMR spectrum of **1** (*Table 1*) displayed signals corresponding to five

Table 1. *^{1}H- and ^{13}C-NMR Data* (400 and 100 MHz, resp., CD_3OD) *of* **1**–**3**[a]). δ in ppm, J in Hz.

	1		**2**		**3**	
	δ(H)	δ(C)	δ(H)	δ(C)	δ(H)	δ(C)
CH_2(1)	1.78–1.80 (*m*), 1.05–1.13 (overlap)	40.5	1.82–1.85 (*m*), 1.10–1.15 (overlap)	40.2	1.74–1.78 (*m*), 1.07–1.17 (overlap)	40.5
CH_2(2)	1.80–1.82 (*m*), 1.63–1.64 (overlap)	29.8	1.79–1.82 (*m*), 1.63–1.64 (overlap)	29.8	1.78–1.84 (*m*), 1.58–1.63 (overlap)	29.8
H–C(3)	3.14–3.21 (*m*)	90.1	3.17–3.24 (*m*)	90.6	3.19–3.25 (*m*)	90.2
C(4)		40.5		40.5		40.5
H–C(5)	0.84 (*d*, *J* = 11.4)	57.5	0.82 (*d*, *J* = 11.4)	57.3	0.83 (*d*, *J* = 11.2)	57.6
CH_2(6)	1.41–1.54 (*m*)	19.3	1.47–1.64 (*m*)	19.3	1.41–1.61 (*m*)	19.3
CH_2(7)	1.51–1.54 (overlap)	35.6	1.53–1.57 (overlap)	35.8	1.48–1.55 (overlap)	35.8
C(8)		42.4		42.4		42.4
H–C(9)	1.52–1.58 (overlap)	50.6	1.55–1.59 (overlap)	50.6	1.53–1.57 (overlap)	50.7
C(10)		38.4		38.4		38.4
CH_2(11)	1.90 (*dd*, *J* = 12.2, 11.2), 1.30–1.40 (*m*)	32.9	1.93 (*dd*, *J* = 12.2, 11.5), 1.34–1.39 (*m*)	33.2	1.90 (*dd*, *J* = 12.2, 11.5), 1.29–1.39 (*m*)	33.2
H–C(12)	4.18–4.28 (*m*)	70.3	4.25–4.30 (*m*)	70.4	4.18–4.27 (*m*)	70.4
C(13)		150.2		150.2		150.2
C(14)		44.2		44.2		44.2
CH_2(15)	1.87–1.92 , 1.73–1.80 (2*m*)	27.5	1.87–1.92 (*m*), 1.73–1.80 (*m*)	27.4	1.98–2.05, 1.68–1.79 (2*m*)	27.5
CH_2(16)	2.66 (*td*, *J* = 12.9, 2.5), 1.17–1.21 (overlap)	28.6	2.69 (td, *J* = 13.7, 2.9), 1.21–1.25 (*m*)	28.6	2.66 (*td*, *J* = 13.7, 3.0), 1.16–1.22 (overlap)	28.6
C(17)		45.6		45.7		45.6
H–C(18)	5.86 (*s*)	117.2	5.90 (*s*)	117.2	5.85 (*s*)	117.2
H–C(19)	4.85–4.96 (*m*)	79.8	4.91–4.96 (*m*)	79.8	4.85–4.95 (*m*)	79.8
H–C(20)	1.70–1.80 (*m*)	45.3	1.74–1.79 (*m*)	45.3	1.70–1.77 (*m*)	45.3
H–C(21)	4.22 (*dd*, *J* = 6.4, 2.2)	76.0	4.24 (*dd*, *J* = 6.5, 2.1)	76.0	4.20 (*dd*, *J* = 6.4, 2.2)	76.0
CH_2(22)	2.18 (*dd*, *J* = 12.5, 5.2), 1.85–1.94 (*m*)	43.0	2.21 (*dd*, *J* = 12.5, 5.1), 1.88–1.93 (*m*)	43.0	2.17 (*dd*, *J* = 12.5, 5.2), 1.83–1.92 (*m*)	43.0
Me(23)	1.06 (*s*)	28.6	1.12 (*s*)	28.6	1.08 (*s*)	28.6
Me(24)	0.88 (*s*)	17.2	0.91 (*s*)	17.0	0.88 (*s*)	17.3
Me(25)	0.96 (*s*)	17.4	1.00 (*s*)	17.2	0.96 (*s*)	17.4
Me(26)	0.99 (*s*)	18.9	1.03 (*s*)	18.9	0.99 (*s*)	18.9
Me(27)	1.19 (*s*)	22.4	1.23 (*s*)	22.4	1.18 (*s*)	22.4
C(28)		181.5		181.6		181.5
Me(29)	1.25 (*d*, *J* = 6.4)	18.2	1.29 (*d*, *J* = 6.5)	18.2	1.25 (*d*, *J* = 5.8)	18.2
Me(30)	0.94 (*d*, *J* = 7.0)	8.1	0.98 (*d*, *J* = 7.0)	8.1	0.94 (*d*, *J* = 7.0)	8.1
3-Xyl or 3-Glc						
H–C(1′)	4.41 (*d*, *J* = 6.6)	106.4	4.34 (*d*, *J* = 7.4)	107.6	4.44 (*d*, *J* = 7.4)	105.8
H–C(2′)	3.39–3.47 (*m*)	79.0	3.18–3.26 (overlap)	75.6	3.43–3.50 (*m*)	79.7
H–C(3′)	3.39–3.47 (*m*)	78.8	3.37–3.44 (*m*)	78.2	3.40–3.50 (*m*)	79.1
H–C(4′)	3.37 (*dt*, *J* = 6.5, 2.2)	71.7	3.49–3.55 (*m*)	71.4	3.23–3.32 (*m*)	72.2
CH_2(5′) or H–C(5′)	3.88 (*dd*, *J* = 11.4, 5.0), 3.22 (*dd*, *J* = 12.2, 2.7)	66.6	3.86 (*dd*, *J* = 11.8,2.4), 3.17 (*dd*, *J* = 11.8,5.3)	66.8	3.20–3.27 (*m*)	77.8
CH_2(6′)					3.87 (*dd*, *J* = 11.8, 1.9), 3.70 (*dd*, *J* = 11.9, 5.2)	63.0

Table 1 (cont.)

	1		**2**		**3**	
	δ(H)	δ(C)	δ(H)	δ(C)	δ(H)	δ(C)
2′-Rha						
H–C(1″)	5.35 (*d*, *J* = 1.5)	102.1			5.39 (*d*, *J* = 1.5)	102.0
H–C(2″)	3.76 (*dd*, *J* = 9.6, 3.4)	72.3			3.77 (*dd*, *J* = 9.6, 3.4)	72.3
H–C(3″)	3.97–3.99 (*m*)	72.2			3.97 (*dd*, *J* = 3.3, 1.7)	72.2
H–C(4″)	3.36–3.41 (*m*)	74.1			3.40 (*dd*, *J* = 9.6, 3.4)	74.1
H–C(5″)	3.96–3.98 (*m*)	70.2			4.00 (*d*, *J* = 6.0)	70.1
Me(6″)	1.24 (*d*, *J* = 3.1)	18.3			1.25 (*d*, *J* = 3.1)	18.3
19-Glc						
H–C(1‴)	4.31 (*d*, *J* = 7.8)	103.4	4.35 (*d*, *J* = 7.8)	103.4	4.30 (*d*, *J* = 7.8)	103.4
H–C(2‴)	3.15–3.22 (*m*)	75.2	3.18–3.22 (overlap)	75.3	3.14–3.19 (*m*)	75.2
H–C(3‴)	3.32–3.38 (*m*)	78.4	3.37–3.42 (*m*)	78.4	3.33–3.38 (*m*)	78.4
H–C(4‴)	3.31–3.39 (*m*)	71.8	3.38–3.40 (*m*)	71.8	3.31–3.37 (*m*)	71.8
H–C(5‴)	3.23–3.29 (*m*)	77.8	3.28–3.36 (*m*)	77.9	3.23–3.27 (*m*)	77.8
CH_2(6‴)	3.83 (*dd*, *J* = 11.9, 2.3), 3.71 (*dd*, *J* = 11.9, 5.3)	62.8	3.82 (*dd*, *J* = 11.9, 2.3), 3.75 (*dd*, *J* = 11.9, 5.3)	62.9	3.83 (*dd*, *J* = 11.9, 2.2), 3.70 (*dd*, *J* = 11.9, 5.2)	62.8

[a]) Assignments based on HSQC, HMBC, ^{1}H,^{1}H-COSY, and NOESY experiments.

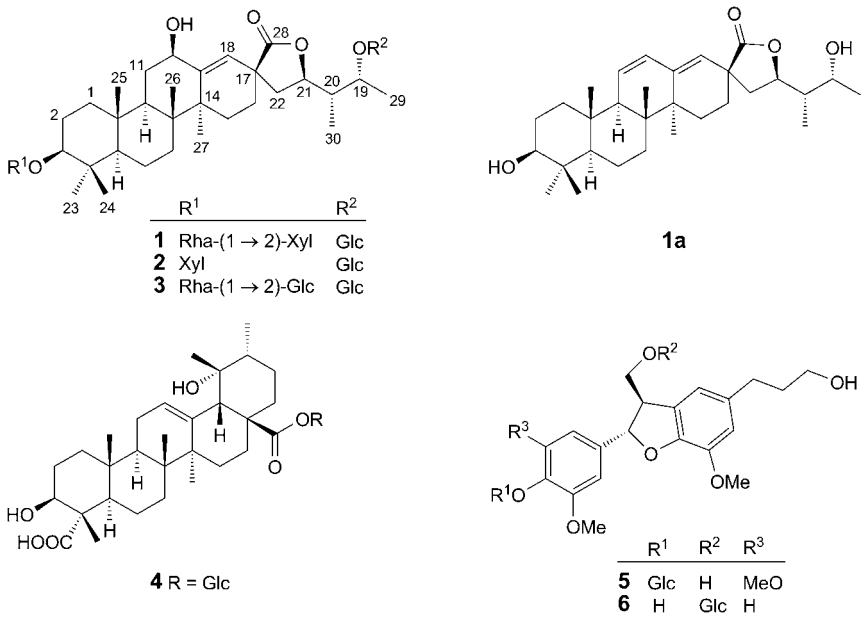

Fig. 1. *Compounds isolated from* I. dunniana

tertiary Me groups (δ(H) 0.88, 0.96, 0.99, 1.06, and 1.19), two secondary Me groups (δ(H) 0.94, 1.25) and one olefinic H-atom signal (δ(H) 5.86). These features suggested that **1** was an ursolic acid type triterpenoid derived from (3β)-3-hydroxyurs-12-en-28-

oic acid. However, the presence of only three non-O-bearing CH (δ(C) 45.3, 50.6, and 57.5) suggested a secoursane structure [2]. Two diagnostic olefinic C-atom signals at δ(C) 117.2 and 150.2, the characteristic resonances of H–C(12) (δ(H) 4.18-4.28 (*m*); δ(C) 70.3) correlated with the olefinic C(13) (δ(C) 150.2) and CH(18) (δ(H) 5.86 (*s*, 1 H); δ(C) 117.2) according to the 1 H,^{13}C-HMBC data, and the Me(30) (δ(H) 0.94 (*d*, $J = 7$ Hz); δ(C) 8.1) vicinal to an OR group, was characteristic of a 18,19-secoursane derivative, which possesses a C(13)=C(18) bond and a β-hydroxy group at C(12) or C(21) [3][4].

The nature of the sugar units of **1** was first determined by the comparison of their ^{13}C-NMR with the corresponding monosaccharides (D-glucose, L-rhamnose, and D-xylose), and was confirmed by HPLC analysis after acid hydrolysis. Acid hydrolysis of compounds **1**–**3** afforded the same triterpene derivative **1a** which lost one H_2O compared with the aglycone of **1**–**3** to form a 11,13(18)-diene moiety (δ(C) 129.3, 129.2, 145.5, and 121.1). The structure of **1a** was established as *rel*-(3β,19*R*,20*S*,21*R*)-3,19-dihydroxy-18,19-secoursane-11,13(18)-diene-28,21-lactone [5] (for NMR data, see *Table 2*).

Table 2. *^{1}H- and ^{13}C-NMR Data* (400 and 100 MHz, resp.; $CDCl_3$) *of Compound* **1a**. δ in ppm, *J* in Hz.

	δ(H)	δ(C)		δ(H)	δ(C)
CH_2(1)	1.03 (*dd*, $J = 13.4, 4.3$), 1.83–1.86 (*m*)	38.3	CH_2(16)	2.44 (*td*, $J = 13.5, 3.5$)	28.6
			C(17)		45.0
CH_2(2)	1.59–1.69 (*m*)	25.1	H–C(18)	5.36 (*s*)	121.1
H–C(3)	3.22 (*dd*, $J = 11.2, 5.0$)	79.2	H–C(19)	4.14 (*qd*, $J = 6.4, 2.2$)	67.6
C(4)		39.1	H–C(20)	1.64–1.67 (overlap)	44.6
H–C(5)	0.75–0.77 (*m*)	55.1	H–C(21)	4.52 (*dt*, $J = 10.1, 5.4$)	78.0
CH_2(6)	1.39–1.44 (*m*), 1.59–1.61 (*m*)	18.3	CH_2(22)	2.19 (*dd*, $J = 13.5, 3.5$), 1.80–1.87 (*m*)	42.5
CH_2(7)	1.30–1.40 (overlap)	32.2	Me(23)	0.97 (*s*)	28.1
C(8)		41.0	Me(24)	0.76 (*s*)	15.3
H–C(9)	1.93 (*d*, $J = 3.0$)	54.6	Me(25)	0.80 (*s*)	16.6
C(10)		41.1	Me(26)	0.88 (*s*)	18.1
H–C(11)	5.89 (*dd*, $J = 10.1, 3.0$)	129.3	Me(27)	0.95 (*s*)	20.3
H–C(12)	5.66 (*d*, $J = 10.1$)	129.2	C(28)		178.8
C(13)		145.5	Me(29)	1.19 (*d*, $J = 6.5$)	20.6
C(14)		37.0	Me(30)	0.84 (*d*, $J = 7.0$)	8.3
CH_2(15)	1.55–1.65 (*m*)	27.3			

In the HMBC spectrum of **1** (*Fig. 2*), the anomeric H-atom of the Glc unit (δ(H) 4.31 (*d*, $J = 7.8$ Hz, H–C(1‴)) was long-range coupled with C(21) (δ(C) 76.0) and C(19) (δ(C) 79.8). The anomeric H-atom of the Rha unit (δ(H) 5.35 (*d*, $J = 1.5$ Hz, H–C(1″))) was long-range coupled with C(2′) of the Xyl unit (δ(C) 79.0), and the anomeric H-atom of the Xyl unit (δ(H) 4.41 (*d*, $J = 6.6$ Hz, H–C(1′))) was long-range coupled with C(3) (δ(C) 90.1). These features established that Xyl was linked at C(3) of the aglycon, Rha was linked at C(2′) of Xyl, and Glc was linked at C(19) of the aglycon. The relative configuration of **1** was determined by NOESY analysis (*Fig. 3*) and comparison with documented data [5][6]. Definitive confirmation of the structure of **1** was obtained from X-ray analysis of a single crystal grown in AcOEt/MeOH/H_2O. Since the absolute

configuration of the sugar residues had been established by HPLC after acid hydrolysis, the absolute configuration of the aglycone of **1** could be deduced from the X-ray data (*Fig. 4*). Thus compound **1** was finally identified as (3*β*,12*β*,19*R*,20*S*,21*R*)-19-(*β*-D-glucopyranosyloxy)-12-hydroxy-3-{[*O*-*α*-L-rhamnopyranosyl-(1 → 2)-*β*-D-xylopyranosyl]oxy}-18,19-secours-13(18)-ene-28,21-lactone, and named dunnianaolactone A.

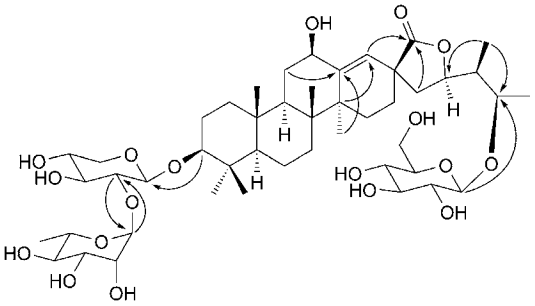

Fig. 2. *Key HMBC features of compound* **1**

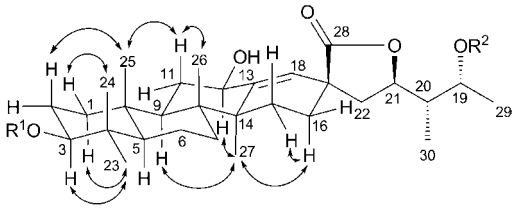

Fig. 3. *Key NOESY correlations of compound* **1**

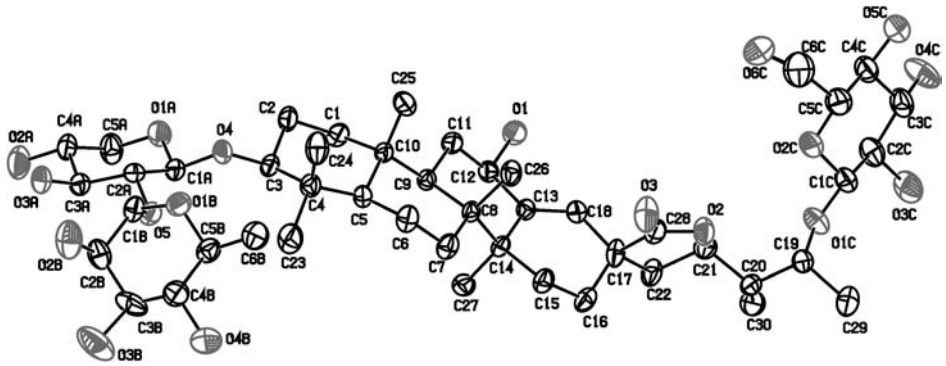

Fig. 4. *ORTEP Drawing of compound* **1**. Partially arbitary atom numbering.

Compound **2** was obtained as white, minute acicular crystals. Comparison of the NMR data of **1** and **2** in CD_3OD (*Table 1*) indicated that **2** had the same aglycone part as **1**. HR-ESI-MS (negative-ion mode) analysis suggested that the molecular formula was $C_{41}H_{68}O_{15}$. Acidic hydrolysis of **2** gave D-xylose and D-glucose.

The position of their attachment was deduced from the HMBC spectrum and further confirmed by comparison with the data reported for alatoside A (same 3-*O*-substituent) and alatosides B and C (similar 19-*O*-substituent) [5]. Thus compound **2** was established as (3*β*,12*β*,19*R*,20*S*,21*R*)-19-(*β*-D-glucopyranosyloxy)-12-hydroxy-3-(*β*-D-xylopyranosyloxy)-18,19-secours-13(18)-ene-28,21-lactone, and named dunnianaolactone B.

Compound **3** was obtained as white, amorphous solid, which had the same aglycone part as **1** and **2**, according to the NMR data (*Table 1*). HR-ESI-MS analysis suggested that the molecular formula of **3** was $C_{48}H_{78}O_{19}$. On acid hydrolysis, **3** furnished D-glucose and L-rhamnose in the ratio 2:1.

The position of attachment and the interglycosidic linkage were established from the HMBC data and were corroborated by comparison with the data reported for alatoside C (same 3-*O*-substituent and 19-*O*-substituent) [5]. Compound **3** was thus established as (3*β*,12*β*,19*R*,20*S*,21*R*)-19-(*β*-D-glucopyranosyloxy)-12-hydroxy-3-{[*O*-*α*-L-rhamnopyranosyl-(1 → 2)-*β*-D-glucopyranosyl]oxy}-18,19-secours-13(18)-ene-28,21-lactone, and named dunnianaolactone C.

Nine known compounds were also isolated from this plant, *i.e.*, an ursane-type triterpenoid saponin, rotundioic acid 28-*β*-D-glucopyranosyl ester (= ilexoside XXX) (**4**) [7] and two benzofuran lignans, 4-[*erythro*-2,3-dihydro-3-(hydroxymethyl)-5-(3-hydroxypropyl)-7-methoxybenzofuran-2-yl]-2,6-dimethoxyphenyl *β*-D-glucopyranoside (**5**) [8] and (7*S*,8*R*)-dihydrodehydrodiconiferyl alcohol 9-(*β*-D-glucopyranoside) (=[(2*S*,3*R*)-2,3-dyhidro-2-(4-hydroxy-3-methoxyphenyl)-5-(3-hydroxyprophyl)-7-methoxybenzofuran-3-yl]methyl *β*-D-glucopyranoside; **6**) [9] (*Fig. 1*) besides five flavonoid glycosides, namely quercetin (=2-(3,4-dihydroxyphenyl)-3,5,7-trihydroxy-4*H*-1-bensopygran-4-one) [10], quercetin 3-(*β*-D-glucopyranoside) [11], quercetin 3-[*α*-L-rhamnopyranosyl-(1 → 6)-*β*-D-glucopyranoside] (rutin) [12], kaempferol 3-(*α*-L-rhamnopyranosyl-(1 → 6)-*β*-D-glucopyranoside [13] and kaempferol 3-[*α*-L-rhamnopyranosyl-(1 → 6)-*β*-D-galactopyranoside] [13] (kaempferol = 3,5,7-trihydroxy-2-(4-hydroxyphenyl)-4*H*-1-benzopyran-4-one), and 4-hydroxybenzoic acid [14].

Discussion. – To date, 19 *E*-secoursane-type triterpenoids have been reported, and twelve of them are from the genus *Ilex*, including *I. crenata*, *I. cornuta*, and *I. aculeolata* [15 – 17]. The most similar isolates are alatosides A – C from *Sesamum alatum* THONN., which possess the same carbon skelekon as dunnianaolactones A – C [5]. Interestingly, alatosides A – C possess an OH–C(18) group and a C(12)=C(13) bond, whereas the situation is reversed in dunnianaolactones A – C, with an OH–C(12) group and a C(13)=(18) bond.

Experimental Part

General. Column chromatography (CC): silica gel *H* (SiO_2; 200– 300 mesh; *Qingdao Marine Chemical Industry*), reversed-phase SiO_2 *CG161* (75 μm; *Rohm and Hass*), *Sephadex LH-20* (20 – 150 μm, *Amersham*), macroreticular resin *HPD-100* (60 – 16 mesh; *Bonchem Chemical Industry*). Anal. HPLC: *Hitachi* HPLC system with UV detector *L-2400* and pump *L-2130*; λ_{max} (log ε) in nm. M.p.: *Tech X-5* microscope melting point apparatus *(Beijing Instrument Co., Ltd.);* uncorrected. ORD: *Jasco-P-1010* spectropolarimeter. IR Spectra: *Bruker-Vertex-70* spectrophotometer; ν in cm^{-1}. 1D- and 2D-NMR Spectra: *Bruker-AM-400* spectrometer; at 400 (^1H) and 100 MHz (^{13}C) in CD_3OD or $CDCl_3$; δ in ppm

rel. to Me_4Si as internal standard, *J* in Hz. MS: *Bruker Microtof-Q-II-10238* mass spectrometer with *Bruker* Compass Datanalysis 4.0 software; in *m*/*z*.

Plant Material. Ilex dunniana LEVL. was collected in October 2007 in Enshi, Hubei Province, China. The plant was identified and authenticated by Prof. *Yingming Wang* at the Wuhan Botanical Garden, Chinese Academy of Sciences. A voucher specimen (ID 20071003) was deposited with the Pharmacy School, Huazhong University of Science and Technology, Wuhan, P. R. China.

Extraction and Isolation. Dried *I. dunniana* leaves were coarsely powdered. Exactly 10 kg of the dried, powdered plant material was extracted 5 × with 95% EtOH (*ca.* 20 l each time), at r.t., for 1 d each time. The EtOH soln. was concentrated resulting in 390 g (3.9%) of extract. The residue was suspended in H_2O (1.5 l) and partitioned successively with petroleum ether (5 × 2.0 l), AcOEt (5 × 2.0 l), and BuOH (5 × 2.0 l). A total of 55.3 g of the BuOH-soluble residue was subjected to CC (*HPD-100,* 20, 50, and 90% EtOH). The 50% EtOH fraction (10.9 g) was then subjected to CC (SiO_2, gradient $CHCl_3$/MeOH/H_2O): *Frs. A – G. Fr. B* was subjected to CC (SiO_2, $CHCl_3$/MeOH 100 : 1 → 1 : 1): quercetin (80 mg) and quercetin 3-(*β*-D-glucopyranoside(44 mg). *Fr. C* was subjected to CC (SiO_2, AcOEt/MeOH/H_2O 80 : 20 : 1): **1** (1.1 g) and **3** (90 mg). *Fr. D* was subjected to CC (SiO_2, AcOEt/MeOH/H_2O 90 : 10 : 1): *Fr. D1*. *Fr. D1* was then subjected to CC (*CG161*, MeOH/H_2O 6 : 1): **2** (6 mg) and **4** (8 mg), which were purified by CC (*Sephadex LH-20,* MeOH). *Fr. E* was subjected to CC (*CG161*, MeOH/H_2O 6 : 1): **5** (11 mg) and **6** (23 mg). *Fr. F* was separated by CC (*Sephadex LH-20,* MeOH): *Frs. F1* and *F2*. *Fr. F1* was subjected to CC (SiO_2, AcOEt/MeOH/H_2O 50 : 50 : 1): to give quercetin 3-[*α*-L-rhamnopyranosyl-(1 → 6)-*β*-D-glucopyranoside] (20 mg), kaempferol 3-[*α*-L-rhamnopyranosyl-(1 → 6)-*β*-D-glucopyranoside] (6 mg), and kaempferol 3-[*α*-L-rhamnopyranosyl-(1 → 6)-*β*-D-glucopyranoside] (9 mg). From *Fr. F2*, 4-hydroxybenzoic acid (6 mg) was separated and purified by CC (*Sephadex LH-20,* MeOH).

Dunnianaolactone A (= *(3β,12β,19*R,*20*S,*21*R*)-3β-{[2-O-(6-Deoxy-α-*L*-mannopyranosyl) β-D-xylopyranosyl]oxy}-19-(β-D-glucopyranosyl)oxy)-12,21-dihydroxy-18,19-secours-13(18)-en-28-oic Acid γ-Lactone*; **1**): Colorless, acicular crystals (AcOEt/MeOH/H_2O). M.p. 285 – 286°. $[\alpha]_D^{28} = -102.1$ (*c* = 1.0, MeOH). UV (MeOH): 238 (1.8). IR (KBr): 3550 – 3250, 2935, 1767, 1649, 1459, 1382. ^{1}H- and ^{13}C-NMR: *Table 1*. HR-ESI-MS: 963.4722 ($[M + Cl]^-$, $C_{47}H_{76}O_{18}Cl^-$; calc. 963.4720).

Dunnianaolactone B (= *(3β,12β,19*R,*20*S,*21*R*)-19-(β-D-Glucopyranosyloxy)-12,21-dihydroxy-3-(β-D-xylopyranosyloxy]-18,19-secours-13(18)-en-28-oic Acid γ-Lactone*; **2**): Colorless, minute acicular crystals (AcOEt/MeOH/H_2O). M.p. 196 – 200°. $[\alpha]_D^{28} = -83.6$ (*c* = 0.17, MeOH). UV (MeOH): 236 (2.4). IR (KBr): 3550 – 3250, 2940, 1751, 1650, 1460, 1383. ^{1}H- and ^{13}C-NMR: *Table 1.* HR-ESI-MS: 817.4153 ($[M + Cl]^-$, $C_{41}H_{66}O_{14}Cl^-$; calc. 817.4141).

Dunnianaolactone C (= *(3β,12β,19*R,*20*S,*21*R*)-3-{[2-O-(6-Deoxy-α-*L*-mannopyranosyl)-β-D-glucopyranosyl]oxy}-19-(β-D-glucopyranosyloxy)-12,21-dihydroxy-18,19-secours-13(18)-en-28-oic Acid γ-Lactone*; **3**): White solid. M.p. 316 – 319°. $[\alpha]_D^{28} = -95.5$ (*c* = 0.99, MeOH). UV (MeOH): 238 (1.9). IR (KBr): 3550 – 3250, 2945, 1769, 1631, 1459, 1377. ^{1}H- and ^{13}C-NMR: *Table 1.* HR-ESI-MS: 957.5037 ($[M - H]^-$, $C_{48}H_{77}O_{19}^-$; calc. 957.5059).

Sugar Identification [18][19]. Each compound **1** – **3** (2.0 mg) was heated seperately at 80° in 2M HCl (2 ml) for 4 h. After cooling, the mixture was extracted with $CHCl_3$ (3 × 2 ml). The $CHCl_3$ extract was washed to give **1a** as an amorphous powder (see *Table 2*). The H_2O-soluble fraction was concentrated and the residue dissolved in pyridine (1 ml) and treated with L-cysteine methyl ester (10 mg) for 1.5 h at 60°. Then, *O*-tolyl isothiocyanate (20 µl) was added and the mixture heated at 60° for 1.5 h. The mixture was analyzed by anal. HPLC (*Thermo ODS Hypersil Dim* (5 µm, 4.6 × 250 mm): at 35°, MeCN/50 mM H_3PO_4 in H_2O 3 : 7 for 40 min and washing of the column with 90% MeCN; detection at 250 ml). D-Glucose (t_R 16.76 min), L-rhamnose (t_R 26.65 min), D-xylose (t_R 18.34 min), and L-arabinose (t_R 17.67 min), identified by comparison with authentic samples.

X-Ray Crystal-Structure Analysis of Dunnianaolactone A (**1**)[1]). Crystal data: $C_{47}H_{77}O_{18} \cdot 4(H_2O)O$; M_r 1018.15; crystal size 0.10 × 0.10 × 0.12 mm, monoclinic, space group *C*2 (No. 5); *a* = 35.23900 Å, *b* = 6.58900 Å, *c* = 24.74800 Å, *α* = 90°, *β* = 109.8600°, *γ* = 90°; *V* = 5404.48 Å^3; *Z* = 4; D_{calc} = 1.251 g cm^{-3}; F_{000} = 2204; *μ*(Mo*K*$_\alpha$) = 0.099 mm^{-1}. Data collection was performed with a *Smart Apex CCD* and

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[1]) CCDC-796161 contains the supplementary crystallographic data for this article. These data can be obtained free of charge *via* http://www.ccdc.cam.ac.uk/data_request/cif.

graphite-monochromated radiation (λ 0.71073 Å), 6565 unique reflections were collected to $\theta_{max} = 25.0°$, in which 4401 reflections were observed ($I > 2.0\ \sigma(I)$). The structure was solved by direct methods (SHELXTL) and refined by full-matrix least squares on I. In the structure refinements, non-H-atoms were refined anisotropically. H-Atoms bonded to C-atoms were placed on the geometrically ideal positions by the 'ride on' method. H-Atoms bound to O-atom were located by the difference *Fourier* method and were included in the calculation of structure factors with isotropic temp. factors. The final indices were $R = 0.0659$, $R_w = 0.1791$, and $S = 0.96$.

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Received October 19, 2010