Triterpene Saponins from *Tupidanthus calyptratus*

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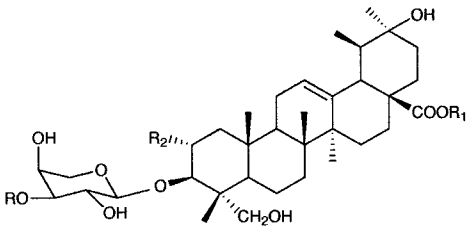
Five new bisdesmosidic saponins (**1**−**5**) were isolated from the aerial parts *of Tupidanthus calyptratus*. Their structures were determined by ^{1}H−^{1}H correlation spectroscopy (COSY, TOCSY, ROESY) and ^{1}H−^{13}C correlation (HSQC, HMBC) NMR experiments, FABMS, and chemical data.

As a part of our studies on the biologically active metabolites from Araliaceae plants from the Botanical Garden of Palermo,[1] we describe herein the separation and structural elucidation of five new bisdesmosidic saponins isolated from the MeOH extract of the aerial parts of *Tupidanthus calyptratus* (Araliaceae).[2]

Saponins isolated and identified from Araliaceae species are reported to have various pharmacological activities including increasing mental efficiency, permitting the recovery of physical balance, stimulation of metabolic function, and other general health-promoting effects.[3,4]

Results and Discussion

The *n*-BuOH extract of aerial parts of *Tupidanthus caliptratus* was subjected to Sephadex LH-20 column cromatography, followed by droplet countercurrent chromatography (DCCC) and then reversed-phase HPLC, to give five pure compounds (**1**−**5**).



	R	R_1	R_2
1	Xyl	Glc-(1−>6)-Glc	OH
2	Xyl	Glc	OH
3	H	Glc	OH
4	Xyl	Rha-(1−>3)-Rha-(1−>6)-Glc	H
5	Xyl	Rha-(1−>6)-Glc	H

Compound **1** had a molecular formula $C_{52}H_{84}O_{24}$, as determined by ^{13}C and ^{13}C DEPT NMR and negative-ion FABMS. The FABMS of **1** showed the $[M - H]^-$ ion at *m/z* 1091 and prominent fragments at *m/z* 929 $[(M - H) - 162]^-$ and *m/z* 945 $[(M - H) - 178]^-$ (cleavage of one hexose unit with or without the glycosidic oxygen), at *m/z* 797 $[(M - H) - 162 + 132]^-$ due to the subsequent loss of one pentose unit, and at *m/z* 635 $[(M - H) - 162 + 132 + 162)]^-$ due to sequential loss of a hexose unit. A peak at *m/z* 503 was attributed to the aglycon moiety.

The ^{13}C NMR spectrum showed 52 signals, of which 30 were assigned to a triterpenoid moiety and 22 to the saccharide portion. The following NMR data suggested the structural features of urs-12-en-28-oic acid for the aglycon of compound **1**: olefinic hydrogen at δ 5.27 (1H, m H-12), a methyl doublet (δ 0.96, d, J = 6.2 Hz, Me-29), signals for C-12 and C-13 at δ 127.0 and 139.8, and the carbonyl carbon resonance at δ 177.5 (C-28). The ^{1}H NMR spectrum of **1** showed also signals at δ 3.69 and 3.40 ascribable respectively to the 2β- and 3α-protons on carbons bearing a hydroxyl function. An AB doublet, δ 3.27 (J = 12 Hz) and 3.70 (J = 12 Hz), indicated the presence of a $-CH_2OH$ function.[5] The chemical shifts of C-4 and Me-24 led to placement of the $-CH_2OH$ at the C-23 position.[5] Full assignments of the proton and carbon signals of the aglycon part of **1** were secured by ^{1}H−^{1}H DQF-COSY and HSQC spectra. The proton and carbon signals due to the A, B, C, and D rings were in agreement with those reported for the asiatic acid (2α,3β,23-trihydroxyurs-12-en-28-oic acid).[6,7]

However the NMR spectra of **1**, compared with those of asiatic acid, lacked a methyl doublet (Me-30) and contained a signal for a methyl singlet at δ 1.09 in the ^{1}H NMR as well as a quaternary hydroxylated carbon (δ 88.0) in the ^{13}C NMR spectrum.[6,7] The COSY spectrum also indicated C-18−C-19−Me-29 connectivities starting from the well-resolved signals at δ 2.22 (H-18); the above-mentioned properties left only position C-20 for the tertiary hydroxyl group.[8,9] This interpretation was unambiguously confirmed by the HMBC spectrum of **1**, which showed significant cross-peaks, due to $^2J_{C-H}$ and $^3J_{C-H}$ correlation between H-22 and C-20; H-18 and C-20; C-30 and H-19, H-22; and Me-29 and H-21.

The stereochemistry at C-18, C-19 and the orientation of the hydroxyl group at C-20 were determined using the ROESY spectrum, which showed key correlation peaks between H-19 and the signals of Me-27 and Me-30 as well as between Me-30 and H-21 and H-22. Therefore the aglycon of compound **1** is 2α,3β,20β-23-tetrahydroxyurs-12-en-28-oic acid. To the best of our knowledge this is the first report of this ursane skeleton.

Glycosidation of the alcoholic function at C-3 and esterification of the 28-COOH group were indicated by the

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Table 1. ^{1}H NMR Data (δ values, J in Hz) for the Oligosaccharide Moieties of Coumpounds **1**−**5** in CD_3OD[a]

proton	1	2	3	4	5
Ara 1	4.37 d (7.6)	4.40	4.34	4.36 d (7.0)	4.34
2	3.70 dd (7.6, 9.0)	3.71	3.82	3.73 dd (7.0, 9.0)	3.75
3	3.88 dd (2.5, 9.0)	3.88	3.55	3.85 dd (9.0, 3.0)	3.87
4	3.85 m	3.85	3.87	3.87, m	3.88
5a	3.91 dd (2.0, 12.0)	3.91	3.63	3.91 dd (12.0, 2.0)	3.92
5b	3.62 dd (3.0, 12.0)	3.62	3.91	3.62 dd (12.0, 3.5)	3.62
Glc 1	5.40 d (7.5)	5.39	5.37	5.36 d (7.5)	5.40
2	3.50 dd (7.5, 9.0)	3.49	3.36	3.36 dd (7.5, 9.0)	3.36
3	3.31 t (9.0)	3.31	3.40	3.40 t (9.0)	3.41
4	3.43 t (9.0)	3.43	3.37	3.39 t (9.0)	3.39
5	3.55 m	3.35	3.43	3.52 m	3.55
6a	3.98 dd (3.0, 12.0)	3.76	3.78	3.99 dd (3.0, 12.0)	3.98
6b	3.76 dd (5.0, 12.0)	3.66	3.64	3.84 dd (5.0, 12.0)	3.84
Xyl 1	4.62 d (7.6)	4.60		4.64 d (7.5)	4.64
2	3.20 dd (7.5, 9.5)	3.18		3.20 dd (7.5, 9.5)	3.18
3	3.35 t (9.5)	3.27		3.32 t (9.5)	3.30
4	3.39 m	3.39		3.36 m	3.35
5a	3.80 dd (10.0, 5.0)	3.82		3.80 dd (10.0, 5.0)	3.82
5b	3.16 (10.0, 12.0)	3.15		3.16 dd (10.0, 12.0)	3.15
GlcI 1	4.57 d 7.5				
2	3.35 dd (7.5, 9.5)				
3	3.46 t (9.5)				
4	3.38 t (9.5)				
5	3.40 m				
6a	3.81 dd (3.0, 12.0)				
6b	3.66 dd (5.0, 12.0)				
Rha 1				4.75 d (1.5)	
2				4.18 dd (1.5, 3.0)	
3				3.95 dd (9.0, 3.0)	
4				3.66 t (9.0)	
5				3.84 dd (9.0,6.2)	
6				1.30 d (6.2)	
RhaI 1				5.15 d (1.6)	5.12
2				3.90 dd (1.6, 3.5)	3.90
3				3.73 dd (9.0, 3.5)	3.74
4				3.50 t (9.0)	3.50
5				3.92 dd (9.0, 6.5)	3.93
6				1.28 d (6.5)	1.28

[a] Assignments were confirmed by COSY-DQF, 1D TOCSY, HSQC, and HMBC experiments.

Table 2. ^{13}C NMR Data for the Oligosaccharide Moieties of Compound **1**−**5** in CD_3OD[a]

	1	2	3	4	5
Ara 1	105.5	105.4	106.04	106.0	105.4
2	71.6	71.8	73.09	72.0	71.8
3	79.4	79.6	74.8	79.8	79.6
4	68.4	68.4	70.3	69.0	69.0
5	66.5	66.8	64.4	66.1	65.5
Xyl 1	105.0	104.9		105.0	105.0
2	74.9	74.9		74.7	74.6
3	77.5	77.5		77.7	77.7
4	72.0	72.0		71.8	72.1
5	66.1	66.1		66.8	66.7
Glc 1	94.0	93.5	95.8	95.0	94.5
2	73.6	73.5	73.8	73.5	73.5
3	77.4	77.0	78.4	77.6	77.6
4	70.5	70.2	71.3	70.4	70.6
5	77.0	77.5	78.7	77.0	76.9
6	68.0	62.3	62.6	68.5	68.8
GlcI 1	104.6				
2	74.0				
3	78.0				
4	71.5				
5	77.9				
6	62.0				
Rha 1				102.5	101.9
2				71.0	71.6
3				80.0	72.8
4				72.0	74.5
5				69.5	69.8
6				18.0	18.0
RhaI 1				101.7	
2				71.8	
3				72.6	
4				74.5	
5				70.3	
6				18.20	

[a] Assignments (δ values) were confirmed by COSY-DQF, 1D TOCSY, HSQC, and HMBC experiments.

downfield shift (+10 ppm) and the highfield shift (−4 ppm) observed, respectively, for these carbon resonances in **1**, relative to the corresponding signals in model compounds.[10]

The sugar portion of **1** contained, in the ^{1}H NMR spectrum (Table 1), four anomeric proton signals (δ 4.37 d, J = 76 Hz; 4.57 d, J = 7.5 Hz; 4.62 d, J = 7.6 Hz; 5.40 d, J = 7.5). The structures of the oligosaccharide moieties were deduced using 1D TOCSY and 2D DQF-COSY experiments (Table 2). In the HSQC experiments glycosidation shifts were observed for C-3_{ara} (79.4 ppm) and C-6_{glc} (68.0 ppm). The absence of any ^{13}C glycosidation shift for the xylopyranosyl and glucopyranosyl moieties suggested that these sugars were terminal units. Chemical shifts of H-1_{glc} (5.40) and C-1_{glc} (94.0) indicated that this sugar was involved in an ester linkage with the C-28 carboxylic group. The positions of the sugar residues were unambiguously defined by the HMBC experiments. A cross-peak due to the long-range correlation between C-3 (δ 87.6) of the aglycon and H-1_{ara} (δ 4.37) indicated that arabinose was the pentose linked to C-3 of the aglycon, and a cross-peak between C-3_{ara} (79.4) and H-1 of the terminal xylose (δ 4.62) indicated that xylose was the second unit of the disaccharide chain at C-3 of the aglycon. Chemical shift, multiplicity, absolute values of the coupling constants, and magnitude in the ^{1}H NMR spectrum as well as ^{13}C NMR data indicated the β-configuration at the anomeric positions for the xylose and glucose units and the α-configuration for arabinose.[11,12] Thus, compound **1** was identified as 3β-*O*-(β-D-xylopyranosyl-(1−3)-α-L-arabinopyranosyl)-2α,20β,23-trihydroxyurs-12-en-28-*O*-[β-D-glucopyranosyl-(1−6)-β-D-glucopyranosyl] ester.

The FABMS of compound **2** ($C_{46}H_{74}O_{19}$) showed $[M - H]^-$ at m/z 929 and prominent fragments at m/z 797 $[(M - H) - 162]^-$ and m/z 635 $[(M - H) - 132 + 162]^-$ (cleavage of one hexose and one pentose unit), and at m/z 503 $[(M - H) - 132 + 162 + 132]^-$ due to the subsequent loss of a pentose unit. The ^{13}C and DEPT ^{13}C NMR spectra showed 46 signals, of which 16 were assigned to the saccharide portion and 30 to a triterpene moiety. Analysis of NMR data of compound **2** and comparison with those of **1** showed **2** to differ from **1** only in the absence of the terminal glucopyranosyl unit (Tables 1 and 2). Therefore, the structure 3β-*O*-(β-D-xylopyranosyl-(1−3)-α-L-arabinopyranosyl)-2α,20β,23-trihydroxyurs-12-en-28-*O*-(β-D-glucopyranosyl) ester was assigned to **2**.

The FABMS of compound **3** ($C_{41}H_{66}O_{15}$) displayed $[M - H]^-$ at m/z 797 and prominent fragments at m/z 635 $[(M - H) - 162]^-$ and m/z 503 $[(M - H) - 162 + 132]^-$ due to the subsequent losses of a hexose and pentose unit. Analysis of NMR data of compound **3** and comparison with those of **2** showed **3** differs from **2** only in the absence of the terminal xylopyranosyl unit (Tables 1 and 2). Therefore, compound **3** was determined to be 3β-*O*-(α-L-arabinopyranosyl)-2α,20β,23-trihydroxyurs-12-en-28-*O*-β-D-glucopyranosyl ester.

The FABMS of compound **4** ($C_{58}H_{94}O_{26}$) showed $[M - H]^-$ at m/z 1205 and prominent fragments at m/z 1059 $[(M - H) - 146]^-$ and m/z 913 $[(M - H) - 146 + 146]^-$ (cleavage of one deoxyhexose and two deoxyhexose, respectively); a peak at m/z 487 was attributed to the aglycon moiety. Comparison of ^{1}H and ^{13}C NMR data of the aglycon of **4**

with those of **1** indicated structural similarity. The main differences were the upfield shifts of C-2 (δ 26.4), C-3 (δ 82.0), and C-1 (δ 39.8) and downfield shift of C-10 (38.1) and C-4 (43.7) in the ^{13}C NMR spectrum and the upfield shift of H-1, H-2 and the downfield shift of H-3 (δ 3.64) in the ^{1}H NMR spectrum, implying an absence of a hydroxyl group at C-2.[8,9] This hypothesis was confirmed unambiguously by the 2D NMR experiments. Thus, the aglycon of **4** was identified as 3β,20β,23-trihydroxyurs-12-en-28-oic acid.

Analysis of NMR data for the sugar portion of compound **4** and comparison with those of **1** showed **4** to differ from **1** in the glycoside at C-28. Also, in this case, the proton coupling network within each sugar residue was determined using a combination of NMR experiments, which led to the identification of two rhamnopyranose units and one glucopyranose, one xylopyranose, and one arabinopyranose unit. Once again, directed evidence for the sugar sequence and the linkage sites was derived from HSQC and HMBC experiments.

These results established that the trisaccharide chain at C-28 of **4** contained a terminal α-L-rhamnopyranose, an inner α-L-rhamnopyranose substituted at C-3, and a β-D-glucopyranose esterified at C-28 and substituted at C-6.[11,12] On the basis of the data the structure of **4** was 3β-*O*-(β-D-xylopyranosyl-(1−3)-α-L-arabinopyranosyl)-20β,23-dihydroxy urs-12-en-28-*O*-[α-L-rhamnopyranosyl-(1−3)-α-L-rhamnopyranosyl-(1−6)-β-D-glucopyranosyl] ester.

The FABMS of compound **5** ($C_{52}H_{84}O_{22}$) showed $[M - H]^-$ at *m/z* 1059 and prominent fragments at *m/z* 913 $[(M - H) - 146]^-$ (cleavage of one deoxyhexose) and *m/z* 781 $[(M - H) - (146 + 132)]^-$ (due to the sequential loss of one pentose). A peak at *m/z* 487 was attributed to the aglycon moiety. The ^{13}C and DEPT ^{13}C NMR spectra showed 52 signals, of which 22 were assigned to the saccharide portion and 30 to the triterpene moiety. Analysis of NMR data of compound **5** and comparison with those of **4** showed **5** to differ from **4** only in the absence of the terminal rhamnopyranosyl unit linked at C-3 of the inner rhamose (Tables 1 and 2). Therefore, the structure 3β-*O*-(β-D-xylopyranosyl-(1−3)-α-L-arabinopyranosyl-20β,23-dihydroxyurs-12-en-28-*O*-[α-L-rhamnoppyranosyl-(1−6)-β-D-glucopyranosyl] ester was assigned to **5**.

All isolated compounds were tested for their antiproliferative activity on three continuous culture cell lines (J774, WEHI-164, and HEK-293), but showed no significant activity at the doses evaluated (>100 mM).

Experimental Section

General Experimental Procedures. Optical rotations were measured on a Perkin-Elmer 141 polarimeter using a sodium lamp operating at 589 nm on 1% w/v solutions in MeOH. A Bruker DRX-600 spectrometer operating at 599.19 MHz for ^{1}H and 150.858 MHz for ^{13}C and the UXNMR software package were used for NMR measurements in CD_3OD solutions. 2D experiments: ^{1}H−^{1}H DQF-COSY, inverse detected ^{1}H−^{13}C HSQC, HMBC, and ROESY were obtained using UX-NMR software. 1D TOCSY spectra were acquired using waveform generator-based GAUSS-shaped pulses, mixing time ranging from 100 to 120 ms. FABMS were recorded in a glycerol matrix in the negative ion mode on a VG ZAB instrument (Xe atoms of energy of 2−6 kV). DCCC was performed on an apparatus manufactured by Buchi, equipped with 300 tubes. HPLC separations were performed on a Waters 590 series pumping system equipped with a Waters R401 refractive index detector and with a Waters μ-Bondapak C_{18} column and a U6K injector. GC were run using a Hewlett-Packard 5890 gas chromatograph equipped with mass-selective detector MSD 5970 MS, a split/splitness injector, and a Hewlett-Packard HP-5 fused-silica column (25 m × 0.2 mm; i.d. 0.33 mm film; Wilmington, DE).

Plant Material. The plant *Tupidanthus calyptratus* was collected in Palermo, Italy, in September 1996; a sample has been deposited in the Herbarium of the Botanical Garden of Palermo.

Extraction and Isolation. The dried leaves (400 g) were defatted with petroleum ether (2 g of residue) and then extracted with EtOH to give 7 g of residue. The ethanolic extract was dissolved in H_2O. The H_2O extract was partitioned between EtOAc and *n*-BuOH to afford an *n*-BuOH-soluble portion (4.1 g) and an EtOAc portion (1.5 g). The *n*-BuOH extract (2 g) was chromatographed on a Sephadex LH-20 column (100 × 5 cm) with MeOH as the eluent. Fractions (8 mL) were collected and checked by TLC [silica gel plates, *n*-BuOH−AcOH−H_2O (12:3:5)]. Fractions 18−24 containing the crude glycosidic mixture were further purified by DCCC using *n*-BuOH−Me_2CO−H_2O (30:9:11) in which the stationary phase consisted of the lower phase (descending mode, flow 14 mL/h). DCCC fractions 140−230 (135 mg) were chromatographed on a C_{18} μ-Bondapak column (30 cm × 7.8 mm i.d., flow rate 3.5 mL/min) eluting with MeOH−H_2O (2:3), to yield pure compounds **2** (15 mg, t_R = 7 min), **3** (18 mg, t_R = 13.5 min), and **5** (12 mg, t_R = 6 min). DCCC fractions 240−270 (70 mg) afforded **1** (25 mg, t_R = 12.5 min) and **4** (19 mg, t_R = 11.0 min) using MeOH−H_2O (1:1) as the eluent (flow rate 3.5 mL/min).

Methanolysis of Compounds 1−5. A solution of each compound (2 mg) in anhydrous 2 N HCl/MeOH (0.5 mL) was heated at 80 °C in a stoppered reaction vial for 12 h. On cooling, the solution was neutralized with Ag_2CO_3 and centrifuged. The supernatant was evaporated to dryness under N_2. The residue was reacted with Trisil-Z (Pierce, Rockford, IL), and the carbohydrates were analyzed by GC.

Compound 1: white amorphous powder; $[\alpha]^{25}_D$ +9° (*c* 1, MeOH); ^{1}H NMR (CD_3OD, 600 MHz) δ 5.27 (1H, m, H-12), 3.69 (1H, ddd, *J* = 11.0, 9.0, 3.0 Hz, H-2), 3.40 (1H, d, *J* = 11.0 Hz, H-3), 3.27 (1H, d, *J* = 12.0 Hz, H-23a), 3.07 (1H, d, *J* = 12.0 Hz, H-23b), 2.22 (1H, d, *J* = 13.0 Hz, H-18), 2.12 (1H, m, H-1a), 2.07 (1H, m, H-15a), 2.00 (1H, m, H-11a), 1.96 (1H, m, H-11b), 1.75 (1H, m, H-22a), 1.67 (1H, m, H-22b), 1.64 (1H, s, H-9), 1.60 (1H, m, H-7a), 1.51 (1H, m, H-21a), 1.50 (1H, m, H-6a), 1.45 (1H, m, H-21b), 1.40 (1H, s, H-19), 1.38 (1H, m, H-6b), 1.35 (1H, m, H-7b), 1.31 (1H, br d, *J* = 11.0 Hz, H-5), 1.20 (1H, s, H-27), 1.13 (1H, m, H-16a), 1.09 (1H, m, H-15b), 1.09 (1H, s, H-30), 1.00 (1H, s, H-26), 0.96 (1H, d, *J* = 6.2 Hz, H-29), 0.95 (1H, m, H-1b), 0.95 (1H, m, H-16b), 0.83 (1H, s, H-25), 0.74 (1H, s, H-24); ^{13}C NMR (CD_3OD, 600 MHz) δ 177.5 (C-28), 139.8 (C-13), 127.0 (C-12), 88.0 (C-20), 87.6 (C-3), 66.9 (C-2), 63.0 (C-23), 54.2 (C-18), 46.5 (C-1), 48.5 (C-17), 47.2 (C-9), 46.9 (C-5), 44.0 (C-14), 43.1 (C-4), 40.9 (C-8), 40.4 (C-19), 37.5 (C-21), 37.1 (C-10), 32.7 (C-7), 31.7 (C-22), 29.3 (C-15), 25.3 (C-27), 24.5 (C-11), 24.0 (C-16), 24.0 (C-30), 21.5 (C-29), 18.0 (C-6), 17.9 (C-26), 17.6 (C-25), 14.4 (C-24); FABMS *m/z* 1091 $[M - H]^-$, 929 $[(M - H) - (162)]^-$, 797 $[(M - H) - (162 + 132)]^-$, 635 $[(M - H) - (162 + 162 + 132)]^-$, 503 $[(M - H) - (162 + 162 + 132 + 132)]^-$.

Compound 2: white amorphous powder: $[\alpha]^{25}_D$ +15° (*c* 1, MeOH); NMR data of aglycon moiety are identical to those of compound **1**; ^{1}H and ^{13}C NMR of sugar moieties, Tables 1 and 2; FABMS *m/z* 929 $[M - H]^-$, 797 $[(M - H) - 162]^-$, 635 $[(M - H) - (162 + 132)]^-$, 503 $[(M - H) - (162 + 132 + 132)]^-$.

Compound 3: white amorphous powder: $[\alpha]^{25}_D$ +18° (*c* 1, MeOH); NMR data of aglycon moiety are identical to those of compound **1**; ^{1}H and ^{13}C NMR of sugar moieties, Tables 1 and 2; FABMS *m/z* 797 $[M - H]^-$, 635 $[(M - H) - 162]^-$, 503 $[(M - H) - (162 + 132)]^-$.

Compound 4: white amorphous powder; $[\alpha]^{25}_D$ +35° (*c* 1, MeOH); ^{1}H NMR (CD_3OD, 600 MHz) δ 5.28 (1H, s, H-12) 3.65 (1H, d, *J* = 11.0 Hz, H-23a), 3.64 (1H, dd, *J* = 11.0, 4.5 Hz, H-3), 3.20 (1H, d, *J* = 11.0 Hz, H-23b), 2.28 (1H, d, *J* = 13.0 Hz, H-18), 2.09 (1H, m, H-11a), 2.03 (1H, m, H-11b), 1.92 (1H, m, H-2a), 1.70 (1H, m, H-7b), 1.70 (1H, s H-22), 1.66 (1H, s,

H-5), 1.63 (1H, m, H-1a), 1.62 (1H, s, H-9), 1.61 (1H, m, H-2b), 1.59 (1H, m, H-21a), 1.48 (1H, m, H-6a), 1.40 (1H, m, H-19), 1.36 (1H, m, H-7b), 1.38 (1H, m, H-6b), 1.35 (1H, m, H-21b), 1.19 (1H, s, H-27), 1.15 (1H, m, H-16a), 1.10 (1H, m, H-15a), 1.09 (1H, s, H-30), 1.05 (1H, m, H-15), 1.00 (1H, s, H-26), 0.98 (1H, m, H-1b), 0.98 (1H, m, H-16b), 0.97 (1H, d, $J = 6.2$ Hz, H-29), 0.84 (1H, s, H-25), 0.72 (1H, s, H-24); ^{13}C NMR (CD_3OD, 600 MHz) δ 178.0 (C-28), 140.0 (C-13), 127.0 (C-12), 88.5 (C-20), 82.0 (C-3), 63.4 (C-23), 53.0 (C-18), 48.5 (C-9), 48.5 (C-17), 48.5 (C-5), 43.7 (C-4), 43.5 (C-14), 39.8 (C-8), 39.8 (C-1), 39.5 (C-19), 38.5 (C-10), 48.5 (C-9), 37.0 (C-22), 31.7 (C-21), 31.1 (C-7), 28.7 (C-15), 26.4 (C-2), 24.6 (C-16), 24.5 (C-27), 23.5 (C-11), 23.0 (C-30), 21.3 (C-29), 18.9 (C-6), 16.2 (C-26), 16.1 (C-25), 13.2 (C-24); FABMS *m*/*z* 1205 $[M - H]^-$, 1059 $[(M - H) - 146]^-$, 913 $[(M - H) - (146 + 146)]^-$, 781 $[(M - H) - (146 + 146 + 132)]^-$, 487 $[(M - H) - (162 + 132 + 132 + 146 + 146)]^-$.

Compound 5: white amorphous powder; $[\alpha]^{25}_D$ +27° (*c* 1, MeOH); NMR data of aglycon moiety are identical to those of compound **1**; ^{1}H and ^{13}C NMR of sugar moieties, Tables 1 and 2; FABMS *m*/*z* 1059 $[M - H]^-$, 913 $[(M - H) - 146]^-$, 781 $[(M - H) - (132 + 146)]^-$, 619 $[(M - H) - (146 + 162 + 132)]^-$, 487 $[(M - H) - (162 + 132 + 132 + 146)]^-$.

Antiproliferative Assays. For experimental data of Antiproliferative assay see De Tommasi et al.[13]

References and Notes

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