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PREGNANE GLYCOSIDES FROM *SANSEVIERIA TRIFASCIATA*

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Key Word Index—*Sansevieria trifasciata*; Agavaceae; pregnane glycosides; 1β,3β-dihydroxypregna-5,16-dien-20-one glycosides.

Abstract—Phytochemical analysis of the whole plant of *Sansevieria trifasciata*, one of the most common Agavaceae plants, has resulted in the isolation of four new pregnane glycosides. Their structures have been determined by spectroscopic analysis and acid- and alkaline-catalysed hydrolysis to be 1β,3β-dihydroxypregna-5,16-dien-20-one glycosides. This is believed to be the first report of the isolation of the pregnane glycosides from a plant of the family Agavaceae. Copyright © 1996 Elsevier Science Ltd

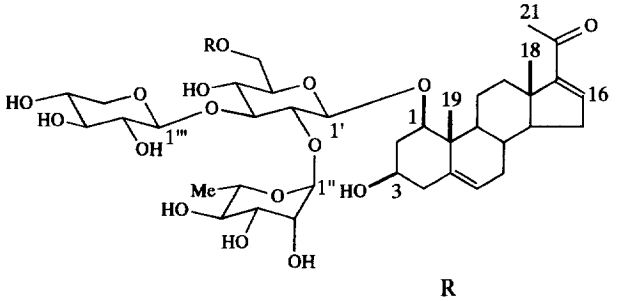
INTRODUCTION

Previously, we have reported the isolation and structural assignment of ten new steroidal saponins from *Sansevieria trifasciata* native to the subtropical regions of Africa [1]. Further phytochemical analysis of *S. trifasciata* has resulted in the isolation of four new pregnane glycosides. This paper deals with the structural determination of the new pregnanes based on spectroscopic analysis and acid- and alkaline-catalysed hydrolysis.

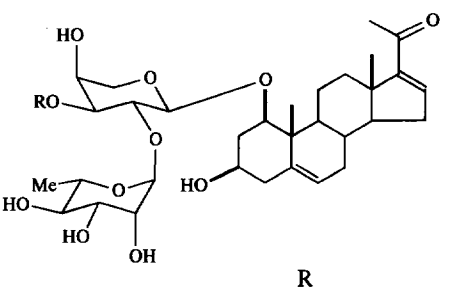
RESULTS AND DISCUSSION

A methanolic extract of the fresh whole plant of *S. trifasciata* was partitioned between 1-butanol and water. Compounds **1–4** were isolated from the 1-butanol-soluble phase after a series of chromatographic separations.

Compound **1**, $C_{38}H_{58}O_{16}$ (positive FAB-mass spectrum m/z 793 $[M+Na]^+$, 771 $[M+H]^+$), $[\alpha]_D$ −26.4° (methanol), was obtained as an amorphous solid. The ^1H NMR spectrum of **1** showed two tertiary methyl proton signals at δ 1.44 and 1.02 (each *s*), an acetylmethyl proton signal at δ 2.21 (*s*), two olefinic proton signals at δ 6.56 (*br s*) and 5.60 (*br d*, J = 5.5 Hz), and three anomeric proton signals at δ 6.39 (*br s*), 4.92 (*d*, J = 7.6 Hz) and 4.80 (*d*, J = 7.7 Hz). The presence of an α,β-unsaturated carbonyl group was indicated by the IR (ν_{max} 1655 cm^{-1}), UV [λ_{max} 241 nm (log ε 3.82)] and ^{13}C NMR [δ 196.2 (C)] spectra. Acid hydrolysis of **1** with 1 M hydrochloric acid in dioxane–water gave an aglycone ($C_{21}H_{30}O_3$), and D-glucose, D-xylose and L-rhamnose in a ratio of 1:1:1 as the carbohydrate compounds. The above data implied that **1** was a triglycoside of a pregnane derived from a spirostanol by Marker's degradation [2]. Comparison of the ^{13}C NMR spectrum of **1** (Table 2) with that of 3β-hydroxypregna-5,16-dien-20-one 3-*O*-glycoside [3], indicated the introduction of one more oxygen



	R
1	H
2	Ac



	R
3	H
4	β-D-Xyl*p*

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atom onto the aglycone moiety of compound **1**. A careful analysis of the ^1H–^1H COSY spectrum in conjunction with the HOHAHA data, which were recorded in a mixed solvent of pyridine-d_5 and methanol-d_4 (11:1) to remove exchangeable protons and minimize signal overlap, verified the spin-networks of the aglycone moiety of compound **1** as shown in Table 1. The multiplet signal centred at δ 3.70 assignable to 3α(ax)-H was coupled to methylene protons at δ 2.58 and 2.27, which in turn were coupled to a deshielded proton at δ 3.83, accounting for the location of the other oxygen atom at C-1. The β(eq)-configuration of the C-1 oxygen atom was shown by the multiplicity of the 1-H proton (doublet of doublets) with the J values, 11.9 and 4.1 Hz. Thus, the structure of the aglycone of compound **1** was assigned as 1β,3β-dihydroxypregna-5,16-dien-20-one. The HMBC correlations optimized for $^nJ_{C,H}$ = 8 Hz allowed assignment of 18-Me, 19-Me and 21-Me and provided further evidence for a 5,16-dien-20-one structure, as shown in Fig. 1.

Table 1. ^1H NMR and ^{13}C NMR chemical shift assignment of compound **1***

Position	^1H	J (Hz)	^{13}C
1	3.83 *dd*	11.9, 4.1	83.8
2 ax	2.27 *q*-like	11.9	37.5
eq	2.58		
3	3.70 *m*		68.1
4 ax	2.62 *dd*	12.7, 12.7	43.6
eq	2.51 *dd*	12.7, 5.0	
5			139.6
6	5.59 *br d*	5.8	124.9
7 a	1.58		31.5
b	1.88 *ddd*	12.4, 5.8, 1.8	
8	1.58		31.8
9	1.62 *m*		50.9
10			42.9
11 ax	1.74 *dddd*	12.7, 12.7, 12.7, 3.8	24.0
eq	2.90 *dddd*	12.7, 3.8, 3.8, 3.8	
12 ax	1.64 *ddd*	12.7, 12.7, 3.8	35.7
eq	2.55 *ddd*	12.7, 3.8, 3.8	
13			46.2
14	1.44 *m*		56.8
15 a	2.13 *ddd*	17.0, 6.4, 3.3	32.6
b	1.93 *ddd*	17.0, 13.5, 1.7	
16	6.62 *dd*	3.3, 1.7	144.8
17			155.9
18	1.00 *s*		16.3
19	1.38 *s*		15.0
20			196.7
21	2.22 *s*		27.0
1′	4.73 *d*	7.4	99.9
2′	4.01 *dd*	8.8, 7.4	76.5
3′	3.97 *dd*	8.8, 8.4	88.4
4′	3.65 *dd*	8.4, 8.4	70.1
5′	3.68 *ddd*	8.4, 5.1, 1.7	77.6
6′	4.33 *dd*	11.4, 1.7	63.1
	4.01 *dd*	11.4, 5.1	
1″	6.22 *d*	1.5	101.6
2″	4.64 *dd*	3.3, 1.5	72.3
3″	4.42 *dd*	9.5, 3.3	72.2
4″	4.16 *dd*	9.5, 9.2	74.0
5″	4.68 *dq*	9.2, 6.2	69.6
6″	1.66 *d*	6.2	19.1
1‴	4.84 *d*	7.7	105.1
2‴	3.85 *dd*	8.8, 7.8	74.6
3‴	3.95 *dd*	8.8, 8.8	78.1
4‴	4.02 *ddd*	10.9, 8.8, 5.3	70.5
5‴	4.19 *dd*	10.9, 5.3	67.1
	3.61 *dd*	10.9, 10.9	

*Spectra were measured in pyridine-d_5–methanol-d_4 (11:1).

Inspection of the ^1H–^1H COSY spectrum, combined with the HOHAHA data, allowed the sequential assignment of the resonances for each monosaccharide of the triglycoside moiety of **1**, starting from the easily distinguished anomeric protons. The HMQC spectrum correlated all the ^1H resonances with those of the corresponding carbons. Comparison of the ^{13}C NMR shifts thus assigned with those of the reference methyl glycosides [4, 5], taking into account the known effects of O-glycosylation and the result of acid hydrolysis, indicated that **1** contained a terminal α-L-rhamnopyranosyl ($^1C_4$: δ 101.6, 72.3, 72.2, 74.0, 69.6 and 19.1), a terminal β-D-xylopyranosyl ($^4C_1$: δ 105.1, 74.6, 78.1, 70.5 and 67.1) and a 2,3-disubstituted β-D-glucopyranosyl ($^4C_1$: δ 99.9, 76.5, 88.4, 70.1, 77.6 and 63.1) moieties. In the HMBC spectrum, the anomeric proton signals at δ 6.22 (*d*, J = 1.5 Hz, rhamnose), 4.84 (*d*, J = 7.7 Hz, xylose) and 4.73 (*d*, J = 7.4 Hz, glucose) exhibited correlations with the ^{13}C signals at δ 76.5, (C-2 of glucose), 88.4 (C-3 of glucose) and 83.8 (C-1 of aglycone), respectively. Accordingly, the structure of **1** was characterized as 1β,3β-dihydroxypregna-5,16-dien-20-one 1-*O*-{*O*-α-L-rhamnopyranosyl-(1→2)-*O*-[β-D-xylopyranosyl-(1→3)]-β-D-glucopyranoside}.

The spectral features of **2**, $C_{40}H_{60}O_{17}$ (positive FAB-mass spectrum m/z 835 $[M+Na]^+$), were quite similar to those of **1**. The presence of an acetyl group in the molecule was shown by the IR (ν_{max} 1745 cm^{-1}), ^1H NMR [δ 2.04 (3H, *s*)] and ^{13}C NMR [δ 170.6 (C) and 20.9 (Me)] spectra. On treatment of **2** with 10% ammonia solution (water–methanol, 1:1), it was hydrolysed to yield **1**. Therefore, compound **2** was confirmed to be a monoacetate of **1**. In the ^{13}C NMR spectrum of **2**, the signal due to the glucose C-6 carbon was shifted to lower field by 0.7 ppm, whereas the signal due to C-5 shifted to upper field by 3.7 ppm, as compared with those of **1**. Furthermore, in the ^1H NMR spectrum of **2** the AB part of the ABX-like signals at δ 4.67 (*dd*, J = 11.7, 2.0 Hz) and 4.60 (*dd*, J = 11.7, 5.0 Hz) were attributable to 6-H_2 of the glucose, which were removed to lower fields by 0.25 and 0.50 ppm, respectively, as compared with those of **1**. Thus, the acetyl moiety was shown to be linked to the glucose C-6 hydroxyl group, and the structure of **2** was assigned as 1β,3β-dihydroxypregna-5,16-dien-20-one 1-*O*-{*O*-α-L-rhamnopyranosyl-(1→2)-*O*-[β-D-xylopyranosyl-(1→3)]-6-*O*-acetyl-β-D-glucopyranoside}.

The NMR data of **3**, $C_{32}H_{48}O_{11}$ (positive FAB-mass

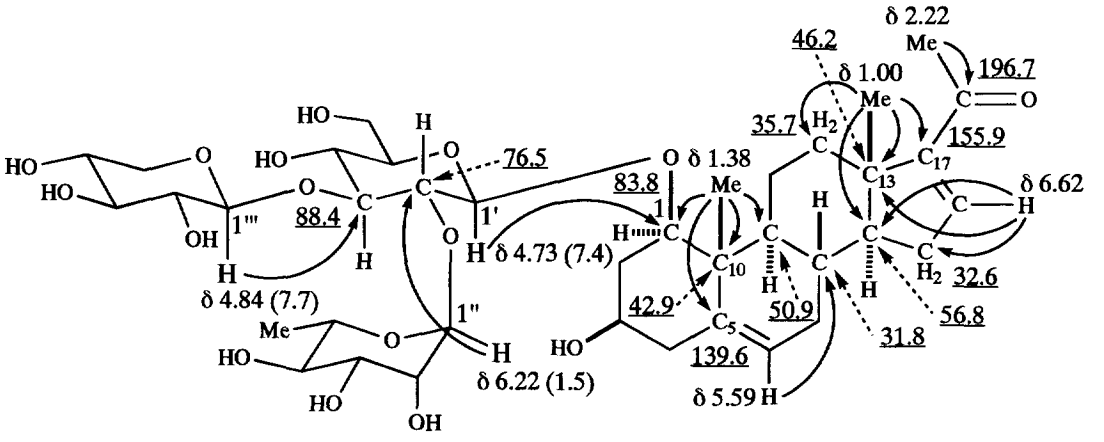

Fig. 1. HMBC correlations of **1** in pyridine-d_5–methanol-d_4 (11:1). *J* values (Hz) in the ^{1}H NMR spectrum are given in parentheses. Underlined values indicate ^{13}C NMR chemical shifts.

spectrum *m/z* 631 $[M+Na]^+$), showed that it possessed an identical aglycone structure to **1** and **2**, but differed from them in terms of the saccharide structure. Acid hydrolysis of **3** gave **1a**, and L-arabinose and L-rhamnose in a ratio of 1:1. The ^{13}C NMR assignment of the saccharide moiety was established by comparison with the chemical shifts of authentic methyl glycosides [4, 5], considering the glycosylation-induced shift. The ^{13}C shifts of a terminal α-L-rhamnopyranosyl unit (δ 101.7, 72.6, 72.7, 74.3, 70.0 and 19.0) and a C-2 glycosylated α-L-arabinopyranosyl unit (δ 100.2, 75.2, 75.9, 70.0 and 67.2) could be attributed [6]. The above data led to the assignment of the structure of **3** as 1β,3β-dihydroxypregna-5,16-dien-20-one 1-*O*-{*O*-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranoside}.

Acid hydrolysis of **4**, $C_{37}H_{56}O_{15}$ (positive FAB-mass spectrum *m/z* 763 $[M+Na]^+$), gave **1a**, and D-xylose, L-arabinose and L-rhamnose in a ratio of 1:1:1. On comparison of the whole ^{13}C NMR spectrum of **4** with that of **3**, a set of additional signals, corresponding to a terminal β-D-xylopyranosyl unit, appeared at δ 106.5, 74.7, 78.2, 71.0 and 67.1, and the signals due to the inner arabinose moiety varied, while all other signals remained almost unaffected. It was observed that the signal of the inner arabinose was markedly displaced downfield at δ 84.5 as compared with that of **3**, indicating the C-3 position of the arabinose was the glycosylated position to which the additional D-xylose was linked. Compound **4** was subjected to partial acid hydrolysis with 0.2 M hydrochloric acid to yield **3** as one of the hydrolysates. Thus, the structure of **4** was established as 1β,3β-dihydroxypregna-5,16-dien-20-one 1-*O*-{*O*-α-L-rhamnopyranosyl-(1→2)-O-[β-D-xylopyranosyl-(1→3)]-α-L-arabinopyranoside}.

Pregn-16-en-20-one derivatives (16-dehydropregnanolones) obtained from spirostanols through Marker's degradation are important starting materials for the synthesis of steroid hormones [7], and have been naturally detected in several plants such as *Solanum vespertilio* (Solanaceae) [8], *Nerium odorum* (Apocynaceae) [9], *Paris polyphylla* (Liliaceae) [10] and *Heloniopsis orientalis* (Liliaceae) [3]. To the best of our knowledge, **1–4** are believed to be the first examples of the pregnane glycosdies from a plant of the Agavaceae.

EXPERIMENTAL

General. NMR (ppm, *J* Hz): Bruker AM-400 (400 MHz for ^{1}H NMR); Bruker AM-500 (500 MHz for ^{1}H NMR). CC: silica-gel (Fuji-Silysia Chemical), octadecylsilanized (ODS) silica-gel (Nacalai Tesque) and Diaion HP-20 (Mitsubishi-Kasei). TLC: precoated Kieselgel 60 F_{254} (0.25 mm thick or 0.5 mm thick, Merck) and RP-18 $F_{254}S$ (0.25 mm thick, Merck). HPLC: a Tosoh HPLC system (pump, CCPM; controller, CCP controller PX-8010; detector, UV-8000) equipped with a TSK-gel ODS-Prep column (Tosoh, 4.6 mm i.d. × 250 mm, ODS, 5 μm).

Plant material. Sansevieria trifasciata was purchased from Exotic Plants, Japan, and the plant specimen is on file in our laboratory.

Extraction and isolation. The plant material (fresh weight 5.4 kg) was extracted with hot MeOH. The MeOH extract was concd under red. pres., and the viscous concentrate was partitioned between H_2O and *n*-BuOH. CC of the *n*-BuOH-soluble phase on silica-gel and elution with a gradient mixt. of $CHCl_3$–MeOH system, and finally with MeOH, gave six frs (I–VI). Fr. V was subjected to ODS silica-gel CC eluting with MeOH–H_2O (4:1) and silica-gel with $CHCl_3$–MeOH–H_2O (40:10:1) to give **2** with a few impurities, and **3** (29.2 mg). Final purification of **2** was established by prep. TLC developing with $CHCl_3$–Et_2O–MeOH–H_2O (5:5:4:1) to furnish **2** (13.1 mg) as a pure compound. After removal of saccharides from fr. VI by CC on Diaion HP-20 eluting with H_2O with a increasing amount of MeOH in H_2O, it was chromatographed on silica-gel eluting with $CHCl_3$–MeOH–H_2O (30:10:1) to yield **1** (21.8 mg) and **4** (57.3 mg).

Compound **1**. Amorphous solid, $[\alpha]_D^{26}$ −26.4° (MeOH; *c* 0.25). Positive FAB-MS *m/z* 793 [M+

Table 2. ^{13}C NMR spectral data for compounds **1**, **1a** and **2–4***

C	1	1a	2	3	4
1	83.8	77.9	85.0	83.1	83.5
2	37.7	43.6	38.0	37.3	37.4
3	68.0	68.1	68.2	68.2	68.3
4	43.8	44.0	43.7	43.9	43.9
5	139.8	140.8	139.7	140.1	140.0
6	124.6	124.1	124.6	124.5	124.5
7	31.5	31.9	31.5	31.6	31.5
8	31.8	31.6	31.8	31.8	31.8
9	50.9	51.9	51.0	51.0	50.9
10	42.9	43.7	42.7	43.0	43.0
11	23.9	24.1	24.1	24.0	24.0
12	35.7	35.9	35.5	35.7	35.6
13	46.1	46.1	46.1	46.0	46.0
14	56.7	56.8	57.2	56.8	56.8
15	32.5	32.6	32.5	32.5	32.5
16	144.4	144.5	144.4	144.2	144.2
17	155.8	155.7	156.0	155.9	155.9
18	16.4	16.2	16.4	16.2	16.2
19	15.0	13.8	15.0	15.0	15.0
20	196.2	196.3	196.4	196.1	196.2
21	27.1	27.1	27.1	27.1	27.2
1′	99.9		100.4	100.2	100.5
2′	76.4		75.8	75.2	74.3
3′	88.4		88.1	75.9	84.5
4′	70.2		69.3	70.0	69.5
5′	77.8		74.1	67.2	67.0
6′	63.3		64.0		
1″	101.7		101.7	101.7	101.8
2″	72.5		72.4[a]	72.6[a]	72.5
3″	72.5		72.5[a]	72.7[a]	72.5
4″	74.2		74.2	74.3	74.2
5″	69.6		69.5	70.0	69.6
6″	19.3		19.2	19.0	19.1
1‴	105.2		105.3		106.5
2‴	74.8		74.7		74.7
3‴	78.3		78.4		78.2
4‴	70.6		70.6		71.0
5‴	67.2		67.2		67.1
Ac			170.6		
			20.9		

*Spectra were measured in pyridine-d_5.
[a]Assignments may be interchanged.

Na]$^+$, 771 [M + H]$^+$; UV λ_{max}^{MeOH} nm (log ε): 241 (3.82); IR ν_{max}^{KBr} cm^{-1}: 3400 (OH), 2930 (CH), 1655 (C=O), 1580 (C=C), 1435, 1375, 1320, 1235, 1150, 1040, 980, 905, 890, 835, 805, 695; ^1H NMR (pyridine-d_5): δ 6.56 (1H, *br s*, 16-H), 6.39 (1H, *br s*, 1″-H), 5.60 (1H, *br d*, J = 5.5 Hz, 6-H), 4.92 (1H, *d*, J = 7.6 Hz, 1‴-H), 4.80 (1H, *d*, J = 7.7 Hz, 1′-H), 4.42 (1H, *br d*, J = 11.1 Hz, 6′a-H), 4.10 (overlapping, 6′b-H), 3.89 (1H, *dd*, J = 12.0, 3.9 Hz, 1-H), 3.77 (1H, *m*, 3-H), 2.21 (3H, *s*, 21-Me), 1.75 (3H, *d*, J = 6.1 Hz, 6″-Me), 1.44 (3H, *s*, 19-Me), 1.02 (3H, *s*, 18-Me).

Acid hydrolysis of **1**. A soln of **1** (5.0 mg) in 1 M HCl (dioxane–H_2O, 1:1, 1 ml) was heated at 100° for 2 hr under an Ar atmosphere. After cooling, the reaction mixt. was neutralized by passing it through an Amberlite IRA-93ZU (Organo) column and chromatographed on silica-gel eluting with a gradient mixt. of $CHCl_3$–MeOH (19:1; 1:1) to give an aglycone (**1a**) (2.1 mg) and a mixt. of monosaccharides (1.6 mg). Compound **1a**: amorphous solid, $[\alpha]_D^{26}$ −10.2° (MeOH; *c* 0.10). CI-MS *m/z* 331 [M + H]$^+$, 313 [M − OH]$^+$; EI-MS *m/z* 312.2089 [M − H_2O]$^+$ (calcd. for $C_{21}H_{28}O_2$: 312.2090); UV λ_{max}^{MeOH} nm (log ε): 240 (3.86); IR ν_{max}^{KBr} cm^{-1}: 3420 (OH), 2930 and 2860 (CH), 1650 (C=O), 1585 (C=C), 1455, 1430, 1375, 1325, 1260, 1240, 1065, 1040, 1005, 990, 970, 950, 910, 860, 800, 720; ^1H NMR (pyridine-d_5): δ 6.61 (1H, *dd*, J = 3.2, 1.8 Hz, 16-H), 5.65 (1H, *br d*, J = 5.7 Hz, 6-H), 3.96 (1H, *m*, 3-H), 3.80 (1H, *dd*, J = 11.6, 3.6 Hz, 1-H), 2.23 (3H, *s*, 21-Me), 1.36 (3H, *s*, 21-Me), 1.03 (3H, *s*, 18-Me). The monosaccharide mixt. (1.6 mg) was diluted with H_2O (1 ml) and treated with (−)-α-methylbenzylamine (5 mg) and Na[BH_3CN] (8 mg) in EtOH (1 ml) at 40° for 4 hr, followed by acetylation with Ac_2O (0.3 ml) in pyridine (0.3 ml). The reaction mixt. was passed through a Sep-Pak C_{18} cartridge (Waters) with H_2O–MeCN (4:1; 1:1; 1:9, each 10 ml). The H_2O–MeOH (1:9) eluate fr. was further passed through a Toyopak IC-SP M cartridge (Tosoh) with EtOH (10 ml) to give a mixt. of 1-[(S)-*N*-acetyl-α-methylbenzylamino]-1-deoxyalditol acetate derivatives of the monosaccharides, which was then analysed by HPLC under the following conditions: solvent, MeCN–H_2O (2:3); flow rate, 0.8 ml min^{-1}; detection, UV 230 nm. The derivatives of D-xylose, D-glucose and L-rhamnose were detected. R_t (min): D-xylose, 17.34; D-glucose, 22.47; L-rhamnose, 25.31.

Compound **2**. Amorphous solid, $[\alpha]_D^{26}$ −12.0° (MeOH; *c* 0.25). Positive FAB-MS *m/z* 835 [M + Na]$^+$; UV λ_{max}^{MeOH} nm (log ε): 241 (3.82); IR ν_{max}^{KBr} cm^{-1}: 3430 (OH), 2940 (CH), 1745 and 1665 (C=O), 1585 (C=C), 1440, 1375, 1320, 1240, 1040, 985, 910, 840, 810, 700; ^1H NMR (pyridine-d_5): δ 6.60 (1H, *br s*, 16-H), 6.39 (1H, *br s*, 1″-H), 5.64 (1H, *br d*, J = 5.7 Hz, 6-H), 4.90 (1H, *d*, J = 7.6 Hz, 1‴-H), 4.70 (1H, *d*, J = 7.8 Hz, 1′-H), 4.67 (1H, *dd*, J = 11.7, 2.0 Hz, 6′a-H), 4.60 (1H, *dd*, J = 11.7, 5.0 Hz, 6′b-H), 3.84 (1H, *m*, 3-H), 3.73 (1H, *dd*, J = 12.0, 3.2 Hz, 1-H), 2.23 (3H, *s*, 21-Me), 2.04 (3H, *s*, Ac), 1.74 (3H, *d*, J = 6.1 Hz, 6″-Me), 1.42 (3H, *s*, 19-Me), 1.04 (3H, *s*, 18-Me).

Alkaline hydrolysis of **2**. Compound **2** (3.0 mg) was subjected to alkaline hydrolysis with 10% ammonia soln in H_2O–MeOH (1:1, 2 ml) at room temp. for 8 hr. After evapn of the soln, the reaction mixt. was chromatographed on silica-gel using $CHCl_3$–MeOH–H_2O (20:10:1) to provide **1** (1.3 mg).

Compound **3**. Amorphous solid, $[\alpha]_D^{26}$ −22.5° (MeOH; *c* 0.25). Positive FAB-MS *m/z* 631 [M + Na]$^+$; UV λ_{max}^{MeOH} nm (log ε): 242 (3.90); IR ν_{max}^{KBr} cm^{-1}: 3420 (OH), 2930 (CH), 1660 (C=O), 1585 (C=C), 1435, 1375, 1360, 1235, 1175, 1135, 1045, 980, 940, 910, 870, 840, 810, 780, 700; ^1H NMR (pyridine-d_5): δ 6.56 (1H, *br s*, 16-H), 6.30 (1H, *br s*, 1″-H),

5.62 (1H, *br d*, $J = 5.6$ Hz, 6-H), 4.81 (1H, *dq*, $J = 9.3$, 6.2 Hz, 5″-H), 4.72 (1H, *d*, $J = 7.3$ Hz, 1′-H), 4.71 (1H, *br d*, $J = 3.4$ Hz, 2″-H), 4.62 (1H, *dd*, $J = 9.3$, 3.4 Hz, 3″-H), 4.57 (1H, *dd*, $J = 8.4$, 7.3 Hz, 2′-H), 4.28 (1H, *dd*, $J = 9.3$, 9.3 Hz, 4″-H), 4.23 (1H, *dd*, $J = 12.0$, 2.2 Hz, 5′a-H), 4.13 (2H, overlapping, 3′-, 4′-H), 3.87 (1H, *m*, 3-H), 3.84 (1H, *dd*, $J = 11.9$, 3.7 Hz, 1-H), 3.64 (1H, *br d*, $J = 12.0$ Hz, 5′b-H), 2.22 (3H, *s*, 21-Me), 1.69 (3H, *d*, $J = 6.2$ Hz, 6″-Me), 1.45 (3H, *s*, 19-Me), 0.98 (3H, *s*, 18-Me).

Compound **4**. Amorphous solid, $[\alpha]_D^{26}$ −23.2° (MeOH; *c* 0.25). Positive FAB-MS *m/z* 763 $[M + Na]^+$; UV λ_{max}^{MeOH} nm (log ε): 241 (3.86); IR ν_{max}^{KBr} cm^{-1}: 3410 (OH), 2930 (CH), 1655 (C=O), 1580 (C=C), 1430, 1370, 1235, 1130, 1035, 975, 905, 835, 805, 780, 695; ^1H NMR (pyridine-d_5): δ 6.56 (1H, *br s*, 16-H), 6.29 (1H, *br s*, 1″-H), 5.62 (1H, *br d*, $J = 5.5$ Hz, 6-H), 4.97 (1H, *d*, $J = 7.5$ Hz, 1‴-H), 4.76 (1H, *dq*, $J = 9.5$, 6.1 Hz, 5″-H), 4.75 (1H, *br d*, $J = 3.3$ Hz, 2″-H), 4.71 (1H, *d*, $J = 7.3$ Hz, 1′-H), 4.61 (1H, *dd*, $J = 8.9$, 7.3 Hz, 2′-H), 4.59 (1H, *dd*, $J = 9.5$, 3.3 Hz, 3″-H), 4.39 (1H, *br s*, 4′-H), 4.29 (1H, *dd*, $J = 11.0$, 5.0 Hz, 5‴a-H), 4.26 (1H, *dd*, $J = 9.5$, 9.5 Hz, 4″-H), 4.20 (1H, *dd*, $J = 12.3$, 2.3 Hz, 5′a-H), 4.17-4.05 (3H, overlapping, 3′-, 3‴-, 4‴-H), 3.93 (1H, *dd*, $J = 7.9$, 7.5 Hz, 2‴-H), 3.88 (1H, *m*, 3-H), 3.82 (1H, *dd*, $J = 12.0$, 3.9 Hz, 1-H), 3.66 (1H, *dd*, $J = 11.0$, 11.0 Hz, 5‴b-H), 3.65 (1H, *br d*, $J = 12.3$ Hz, 5′b-H), 2.22 (3H, *s*, 21-Me), 1.69 (3H, *d*, $J = 6.1$ Hz, 6″-Me), 1.43 (3H, *s*, 19-Me), 0.97 (3H, *s*, 18-Me).

Acid hydrolysis of **3** *and* **4**. Each soln of **3** (5.0 mg) and **4** (25.0 mg) in 1 M HCl was treated identically to that of **1** to give **1a** (1.6 mg) and saccharide fr. (2.0 mg), and **1a** (8.5 mg) and saccharide fr. (11.4 mg), respectively. L-Arabinose and L-rhamnose in a ratio of 1:1 in the saccharide fr. from **3**, and L-arabinose, L-rhamnose and D-xylose in a ratio of 1:1:1 in the saccharide fr. from **4**, were identified by converting them into 1-[(*S*)-*N*-acetyl-α-methylbenzylamino]-1-deoxyalditol acetate derivatives, followed by HPLC analysis. R_t (min): L-arabinose, 16.50; D-xylose, 17.59; D-glucose, 21.49; L-rhamnose, 25.52.

Partial hydrolysis of **4**. A soln of **4** (9.7 mg) in 0.2 M HCl (dioxane–H_2O, 1:1, 2 ml) was heated at 95° for 30 min under an Ar atmosphere. The reaction mixt. was neutralized by passing it through an Amberlite IRA-93ZU column and partitioned by Sep-Pak C_{18} cartridge eluting initially with H_2O–MeOH (4:1) and then with MeOH into the fr. with several hydrolysates (5.3 mg) and a mixt. of monosaccharides (1.6 mg). The hydrolysates were purified by prep. TLC developing with $CHCl_3$–MeOH–H_2O (30:10:1) to give **3** (0.7 mg), **1a** (0.8 mg) as well as the starting material, **4** (1.9 mg). In the monosaccharide mixt., rhamnose, xylose and arabinose were detected by TLC. R_f: rhamnose, 0.69; xylose, 0.61; arabinose, 0.52 (*n*-BuOH–Me_2CO–H_2O, 4:5:1).

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