Journal of Natural Products
Vol. 58, No. 11, pp. 1739-1744, November 1995

NEW TRITERPENES FROM *TRICHOCEREUS PACHANOI*

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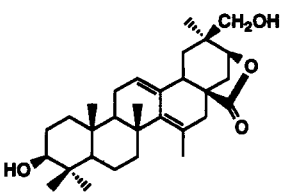
ABSTRACT.—Two novel triterpene aglycones, pachanols A [**1**] and B [**2**] were obtained from *Trichocereus pachanoi* along with known triterpenes bridgesigenins A [**3**] and B [**4**], by acid hydrolysis of a methanol extract. Pachanols A [**1**] and C [**5**] and bridgesigenin C [**6**], together with bridgesigenin A [**3**], were also obtained, by enzymatic degradation of the extract.

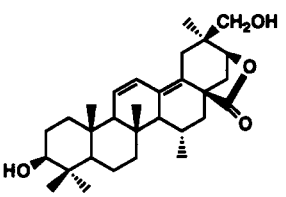
The triterpene sapogenins of plants of the Cactaceae were first studied by Djerassi *et al.* (1). Only a few reports (2–4) were published, however, before our initial studies on the same compounds (5–7). This paper reports new triterpenes from *Trichocereus pachanoi* Britton & Rose, named pachanols A [**1**], B [**2**], and C [**5**] (whose skeleton has been named pachanan), and bridgesigenin C [**6**].

A crude mixture of sapogenins from $CHCl_3$-soluble fractions of the acid-hydrolyzed MeOH extract of *T. pachanoi* was separated, and four triterpenes were obtained by cc over Si gel using a $CHCl_3$/MeOH solvent system as eluent. The structure of pachanol A [**1**] was determined as pachanan-14(15)-ene-3β,30-diol-28(21) lactone by single-crystal X-ray diffraction and nmr spectral analysis (6). Additional single-crystal X-ray diffraction data are reported herein for **1** (Figure 1).

The structure of pachanol B [**2**] was determined by comparison of the nmr data with those of **1**. ^1H-^1H and ^1H-^{13}C correlations in the nmr spectra which permitted the assignment of the partial structure of **2** are shown in Figure 2. The 15*R* configuration was assigned by the results of an nOe nmr experiment (Figure 3). Two other sapogenins were identified as bridgesigenins A [**3**] and B [**4**], respectively, by direct comparison with pub-



1



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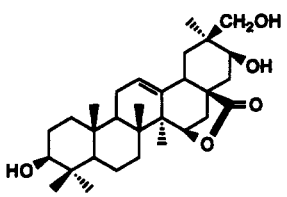

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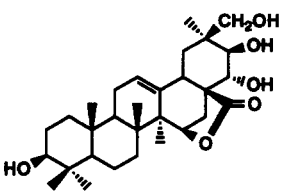

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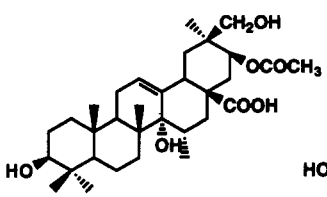

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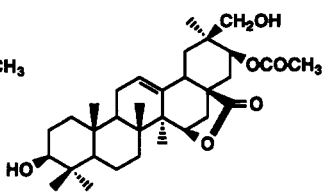

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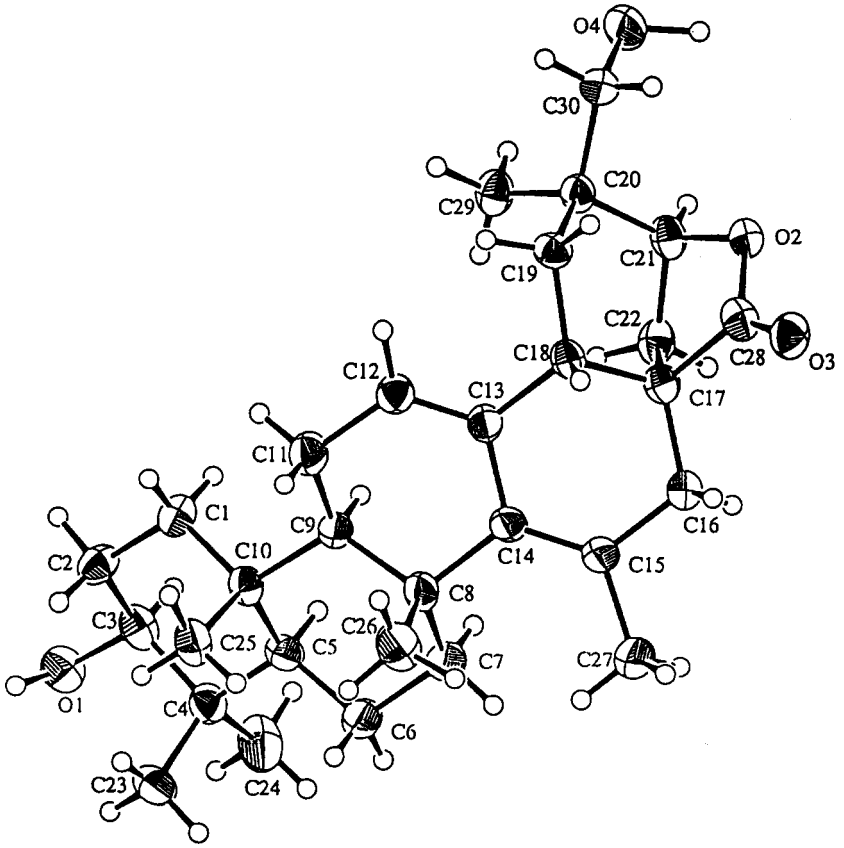

FIGURE 1. ORTEP drawing of **1**.

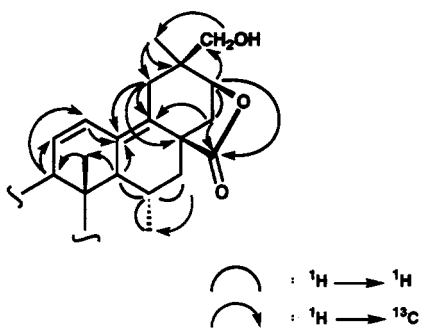

FIGURE 2. Correlations for the partial structure of **2**.

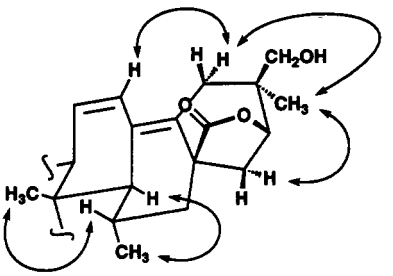

FIGURE 3. Observed positive nOes for **2** from nOe-difference spectra (indicated by arrows).

lished nmr data and physical characteristics (5).

Four triterpenes [**1**, **3**, **5**, **6**] were obtained through enzymatic degradation of the MeOH extract from *T. pachanoi*. Pachanol C [**5**] is a new triterpene with the pachanan skeleton, and its structure was determined by ^{1}H-^{1}H and ^{1}H-^{13}C correlations (Figure 4) and nOe observations (Figure 5). Bridgesigenin C [**6**] was also a new compound, whose structure was determined in the same way (Figures 6 and 7).

It is to be noted that the acid and

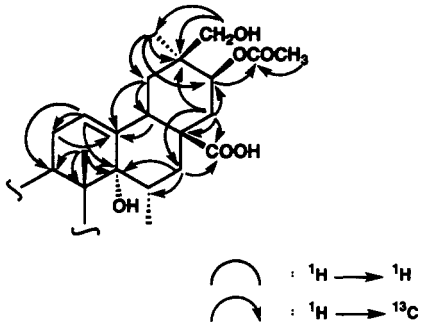

FIGURE 4. Correlations for the partial structure of **5**.

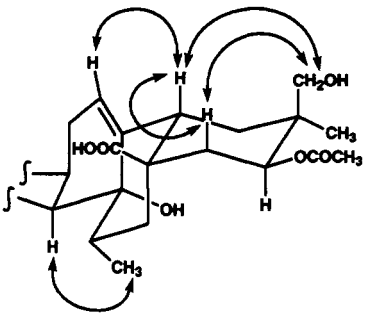

FIGURE 5. Observed positive nOes for **5** from nOe-difference spectra (indicated by arrows).

enzymatic hydrolysis of the MeOH extracts of *T. pachanoi* gave different results. Pachanol C [**5**] and bridgesigenin C [**6**], obtained by enzymatic degradation, were treated with acid, affording pachanol B [**2**] and bridgesigenin A [**3**], respectively. Pachanol B [**2**], an acid hydrolysis product, may be produced by the following reaction steps: hydrolysis of the acetate, with dehydration of the C-14 hydroxyl, producing a double bond, and formation of lactone. The precise mechanism, however, has not been confirmed as yet. The above results suggest that pachanol B [**2**] is formed during the course of acid hydrolysis. The assignments of ^{1}H- and ^{13}C-nmr signals of triterpenes **2**, **5**, and **6** are shown in Tables 1–3, respectively.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Mps were determined with a Yanagimoto MP micro-melting point apparatus. The $[\alpha]_D$ values were determined with a Jasco DIP-140 digital polarimeter. The ir spectra were obtained with a Jasco A-102 ir spectrometer. ^{1}H-, ^{13}C-, DEPT, ^{1}H-^{1}H, and ^{1}H-^{13}C COSY, COLOC, and differential nOe nmr spectra were recorded using a JEOL GSX-400 (^{1}H 400 MHz; ^{13}C 100 MHz) spectrometer, with C_5D_5N as solvent and TMS as the internal standard. The chemical shifts are expressed in ppm (δ) (Tables 1–3). Cc was carried out on 70–230 mesh Si gel (Merck). Hplc was performed using a SSC-3100-J pump with an Oyo-Bunko Uvilog 7 uv detector at 220 nm. Hrms and eims spectra were obtained using a JEOL JMS-DX 302 mass spectrometer. The X-ray crystallographic measurement was made on a Rigaku AFC-7R automatic single-crystal X-ray structure analysis system.

PLANT MATERIAL.—*Tricocereus pachanoi* was cultivated at the Research Institute of Evolutionary Biology, Setagaya-ku, Tokyo, Japan, and Izu National History Park, Itoh, Shizuoka, Japan. This cactus was collected in July 1993, and identified by D. Hirosi Yuasa. A voucher specimen is deposited at the Research Institute of Evolutionary Biology, Setagaya-ku, Tokyo, Japan.

EXTRACTION AND ISOLATION.—Extraction of

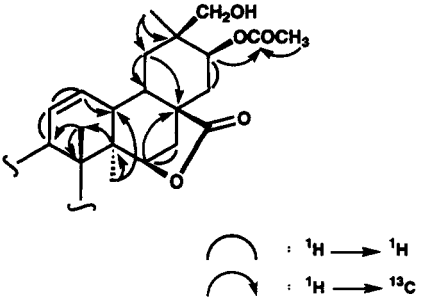

FIGURE 6. Correlations for the partial structure of **6**.

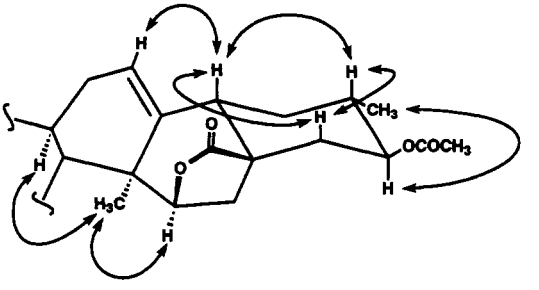

FIGURE 7. Observed positive nOes for **6** from nOe-difference spectra (indicated by arrows).

TABLE 1. Nmr Assignments for Pachanol B [**2**].

Position	δ ^{13}C (C_5D_5N)	δ ^{1}H (C_5D_5N)	Multiplicity (*J* in Hz)	DEPT
1	38.0	1.00	m	CH_2
		1.80	m	
2	27.9	1.89	m	CH_2
3	78.0	3.48	dd (7.3, 16.1)	CH
4	39.4			C
5	55.0	0.85	m	CH
6	19.1	1.62	m	CH_2
7	39.9	1.29	m	CH_2
		1.81	m	
8	40.7			C
9	61.3	1.96	br s	CH
10	37.3			C
11	126.2	6.54	dd (3.1, 10.4)	CH
12	129.0	5.83	d (10.4)	CH
13	132.7			C
14	56.5	1.89	m	CH
15	23.6	2.38	m	CH
16	37.0	1.31	m	CH_2
		2.09	m	
17	46.2			C
18	126.9			C
19	33.0	2.13	m	CH_2
		2.71	d (16.6)	
20	40.3			C
21	79.6	4.85	br s	CH
22	39.5	2.15	d (2.2)	CH_2
23	28.4	1.24	s	CH_3
24	16.0	1.02	s	CH_3
25	15.5	0.84	s	CH_3
26	17.1	0.83	s	CH_3
27	25.6	1.09	d (6.7)	CH_3
28	179.1			C
29	21.5	1.21	s	CH_3
30	68.0	3.62	d (10.4)	CH_2
		3.97	d (10.4)	

the entire plant of *T. pachanoi* with MeOH was performed as described previously (6). The MeOH extract (12.9 g) was hydrolyzed with 3.5% HCl at 110° for 2.5 h. The precipitate (2.98 g) produced was subjected to cc on Si gel ($CHCl_3$/MeOH) and purified by hplc over Si gel (Nucleosil 60-5, 1×25 cm), eluted with $CHCl_3$-MeOH (50:1), resulting in the isolation of pachanols A [**1**] (22.7 mg) and B [**2**] (37.0 mg). Elution with $CHCl_3$-MeOH (20:1) afforded bridgesigenins A [**3**] (34.3 mg) and B [**4**] (24.2 mg). Pachanol A [**1**] was recrystallized with MeOH for X-ray crystallographic analysis.

In an established procedure, the MeOH extract (5.0 g) of *T. pachanoi* was hydrolyzed with β-glucosidase (1.5 g) using 25 ml of HOAc/NaOAc buffer (pH 4.5) at 37° for 4 days. The reaction mixture was extracted with EtOAc and the extract (444.1 mg) subjected to cc on Si gel ($CHCl_3$/MeOH) followed by hplc over a Si gel column (Develosil 60-5, 1×25 cm). Elution with $CHCl_3$-MeOH (50:1) resulted in the purification of pachanols A [**1**] (1.8 mg) and C [**5**] (41.8 mg), and bridgesigenin C [**6**] (35.2 mg); and elution with $CHCl_3$-MeOH (20:1) yielded bridgesigenin B [**4**] (5.5 mg).

Pachanol B [**2**].—Colorless needles ($CHCl_3$/MeOH); mp >300° (dec); $[\alpha]^{20}D$ −95.3° (*c*=0.146, $CHCl_3$); ir ν max 3400, 3450, 2950, 2900, 1740, 1220, 1030 cm^{-1}; uv ($CHCl_3$) λ max (log ε) 255 (3.94) nm; ^{13}C- and ^{1}H-nmr data, see Table 1; eims *m/z* $[M]^+$ 468, 450, 424, 393, 351, 207 (100), 189, 145, 119; hreims *m/z* $[M]^+$ 468.3241 (calcd for $C_{30}H_{44}O_4$, 468.3241).

Pachanol C [**5**].—Colorless needles ($CHCl_3$/MeOH); mp 280°–289°; $[\alpha]^{20}D$ +11.8° (*c*=0.205, $CHCl_3$); ir ν max 3450, 2900, 1720, 1380, 1240, 1140, 1020 cm^{-1}; ^{13}C- and ^{1}H-nmr data, see Table 2; eims *m/z* $[M]^+$ 542, 528, 468, 438, 424, 423,

TABLE 2. Nmr Assignments for Pachanol C [**5**].

Position	δ ^{13}C (C_5D_5N)	δ ^{1}H (C_5D_5N)	Multiplicity (J in Hz)	DEPT
1	39.3	0.90	m	CH_2
		1.55	m	
2	28.0	1.82	m	CH_2
3	77.9	3.44	t-like (7.2)	CH
4	39.4			C
5	55.9	0.88	m	CH
6	18.3	1.43	m	CH_2
		1.64	m	
7	33.6	1.58	m	CH_2
		1.81	m	
8	40.1			C
9	47.9	1.66	m	CH
10	37.6			C
11	22.9	1.85	m	CH_2
12	124.5	5.59	t-like (2.4)	CH
13	133.2			C
14	90.8			C
15	37.2	2.28	m	CH
16	34.3	1.62	m	CH_2
		1.73	m	
17	42.5			C
18	37.1	2.86	bs	CH
19	33.5	1.39	m	CH_2
		2.58	dd (4.8, 14.3)	
20	39.4			C
21	75.8	5.20	t-like (2.4)	CH
22	31.7	1.98	m	CH_2
		2.51	t-like (13.1)	
23	28.8	1.24	s	CH_3
24	16.5	1.03	s	CH_3
25	16.4	0.92	s	CH_3
26	17.6	1.10	s	CH_3
27	20.1	1.02	d (6.8)	CH_3
28	176.7			C
29	24.0	1.28	s	CH_3
30	61.7	3.87	d (10.7)	CH_2
		4.23	d (10.7)	
CH_3CO	170.5			C
CH_3CO	20.1	2.03	s	CH_3

368, 207, 185, 135, 105, 55, 43; hreims *m/z* $[M]^+$ 542.3243 (calcd for $C_{32}H_{46}O_7$, 542.3245).

Bridgesigenin C [**6**].—Colorless needles ($CHCl_3$); mp 278–285°, $[\alpha]^{20}D$ +2.5° (*c*=0.114, $CHCl_3$); ir ν max 3500, 3000, 1780, 1390, 1240, 1130, 1040 cm^{-1}; ^{13}C- and ^{1}H-nmr data, see Table 3; eims *m/z* $[M]^+$ 528, 510, 468, 450, 438, 424, 405, 368, 353, 207, 197, 185, 135, 43; hreims *m/z* $[M]^+$ 528.3460 (calcd for $C_{32}H_{48}O_6$, 528.3452).

CRYSTALLOGRAPHIC ANALYSIS OF **1**.[1]—$C_{30}H_{44}O_4$, *M*, 468.68, orthorhombic, space group $P2_12_12_1$ (No. 19), *a*=13.608(4), *b*=28.388(4), *c*=6.482(5) Å, V=2504(1) Å^3, Dc=1.243 gcm^{-3}; 2204 unique diffractometer data measured at ca. 295° K [2θ max 120°; 2θ/w scan mode, monochromatic CuKα radiation λ 1.5418 A)], 1632 with $I>3\sigma(I)$ considered observed and used in the full-matrix least-squares refinement (μ=6.3 cm^{-1}; specimen 0.30×0.20×0.15 mm). Anisotropic thermal parameter refinement for C and O; isotropic thermal parameters for H; R=0.064 (8). An ORTEP drawing of **1** is shown in Figure 1.

[1]Hydrogen coordinates, thermal parameters, bond distances and angles, and observed and calculated structure factors have been deposited with the Cambridge Crystallographic Data Centre and can be obtained upon request from Dr. Olga Kennard, University Chemical Laboratory, 12 Union Road, Cambridge CB2 1EZ, UK.

TABLE 3. Nmr Assignments for Bridgesigenin C [**6**].

Position	δ ^{13}C (C_5D_5N)	δ ^{1}H (C_5D_5N)	Multiplicity (*J* in Hz)	DEPT
1	39.4	0.97	m	CH_2
		1.54	m	
2	28.1	1.81	m	CH_2
3	78.0	3.46	m	CH
4	39.4			C
5	55.9	0.85	m	CH
6	18.5	1.43	m	CH_2
		1.56	m	
7	33.8	1.55	m	CH_2
8	40.5			C
9	48.6	1.55	m	CH
10	37.2			C
11	23.5	1.79	m	CH_2
12	128.2	5.55	bs	CH
13	137.9			C
14	45.9			C
15	80.2	4.59	d (6.0)	CH
16	34.4	1.96	m	CH_2
		2.77	d (12.3)	
17	46.3			C
18	41.4	2.96	d-like (10.7)	CH
19	41.1	1.84	m	CH_2
		2.43	dd (4.0, 14.0)	
20	40.4			C
21	76.4	5.20	dd (4.8, 12.7)	CH
22	32.4	1.79	m	CH_2
		2.55	t-like (12.9)	
23	28.7	1.24	s	CH_3
24	16.5	1.08	s	CH_3
25	16.3	0.90	s	CH_3
26	19.8	1.19	s	CH_3
27	25.4	1.20	s	CH_3
28	179.0			C
29	24.1	1.25	s	CH_3
30	61.3	3.93	d (10.5)	CH_2
		4.42	d (10.5)	
CH_3CO	170.5			C
CH_3CO	20.9	2.05	s	CH_3

ACKNOWLEDGMENTS

The authors would like to thank Dr. Shoji Shibata of the Laboratory of Natural Medicinal Materials, Shimjuku-ka, Tokyo, for helpful suggestions.

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Received 2 March 1995