New Polyhydroxylated Pregnadienes from the South African Soft Coral *Pieterfaurea unilobata*

Denzil R. Beukes,[†] Michael T. Davies-Coleman,*[,†] Drake S. Eggleston,[‡] R. Curtis Haltiwanger,[‡] and Brian Tomkowicz[‡]

Department of Chemistry, Rhodes University, Grahamstown, 6140 South Africa, and Department of Physical and Structural Chemistry, SmithKline Beecham Pharmaceuticals, King of Prussia, Pennsylvania 19406

Received October 14, 1996[⊗]

Seven pregnadiene metabolites, pregna-5,20-diene-3α,7α-diol 3α-acetate (**1**), pregna-5,20-diene-3*β*,7α-diol 7α-acetate (**2**), pregna-5,20-dien-3*β*-ol (**3**), pregna-5,20-diene-3α,7α,11α-triol 3α-acetate (**4**), pregna-5,20-diene-3α,7α,11α-triol 3α,7α-diacetate (**5**), pregna-5,20-diene-3α,7α,19-triol 3α,19-diacetate (**6**), and pregna-5,20-diene-3α,7α,11α,19-tetrol 3α,7α,19-triacetate (**7**) were isolated from an EtOAc extract of the endemic South African soft coral *Pieterfaurea unilobata*. Standard spectroscopic techniques including X-ray crystallography were used to determine the structures of these compounds, which, apart from compound **3**, are all new Δ^5,Δ^{20} pregnadiene steroids.

South African soft corals have yielded several metabolites that exhibit interesting antiinflammatory,[1,2] anti-HIV,[3] and cytotoxic activity.[4] A high proportion of South African octocorals are endemic (60−70%),[5] and in continuation of our search for new pharmaceuticals from South African soft corals of the order Alcyonacea we have examined the endemic species *Pieterfaurea unilobata* Thomson 1921[5] (family Nidaliidae). Specimens of *P. unilobata*, a species confined to the southeast coast of southern Africa,[5] were collected by scuba (−15 m) from Riet Point near Port Alfred in September 1994.

Results and Discussion

An EtOAc extract of freeze-dried *P. unilobata* (144 g) was concentrated to give a dark-orange oil (2.3 g). Flash chromatography of this oil on Si gel using a hexane−EtOAc solvent gradient gave several crude fractions that were examined by ^{1}H-NMR spectroscopy. Selected fractions were further purified by HPLC on a normal-phase column to obtain seven polyhydroxylated pregnadiene sterols: pregna-5,20-diene-3α,7α-diol 3α-acetate (**1**) (0.09% dry wt), pregna-5,20-diene-3*β*,7α-diol 7α-acetate (**2**) (0.013% dry wt), pregna-5,20-dien-3*β*-ol (**3**) (0.04% dry wt), pregna-5,20-diene-3α,7α,11α-triol 3α-acetate (**4**) (0.003% dry wt), pregna-5,20-diene-3α,7α,11α-triol 3α,7α-diacetate (**5**) (0.004% dry wt), pregna-5,20-diene-3α,7α,19-triol 3α,19 diacetate (**6**) (0.007% dry wt) and pregna-5,20-diene-3α,7α,11α,19-tetrol 3α,7α,19-triacetate (**7**) (0.008% dry wt). Of these seven pregnadiene sterols only **3** is a known compound.

The spectroscopic data of compounds **1**−**7** suggested that these compounds possessed identical skeletal structures, varying only in the degree of hydroxylation and/or acetylation. Accordingly, it was apparent that we had only to establish the structure, substitution pattern, and stereochemistry of the most abundant metabolite, compound **1**, to gain insight into the structures of the rest of this series.

Table 1. ^{13}C-NMR Data (100 MHz, $CDCl_3$) for Compounds **1**−**7**

	compound						
carbon	**1**	**2**	**3**	**4**	**5**	**6**	**7**
1	33.5	31.3	31.7	35.5	35.8	30.9	33.5
2	25.9	36.0	32.0	26.1	27.3	26.2	26.5
3	70.3	68.6	71.8	70.3	70.4	70.1	70.3
4	36.2	41.9	42.3	36.8	36.7	36.4	36.7
5	144.3	147.8	140.9	144.6	146.1	139.6	141.6
6	124.6	119.9	121.6	124.4	120.5	128.5	124.1
7	64.4	71.2	32.1	65.3	68.9	65.0	68.0
8	37.7	36.8	37.3	37.5	35.5	38.8	36.8
9	42.6	43.3	50.5	48.5	48.4	43.0	49.4
10	38.0	37.3	36.7	39.6	39.4	41.0	42.4
11	20.1	20.4	20.7	69.1	68.4	20.7	69.3
12	36.9	36.8	37.4	48.6	48.4	37.1	48.5
13	43.2	43.5	43.4	43.8	43.4	43.4	44.1
14	48.7	48.5	56.0	49.6	50.4	49.1	50.2
15	24.8	24.7	24.9	24.5	24.3	24.7	24.1
16	27.3	27.2	27.2	27.4	26.1	27.3	27.3
17	55.2	55.2	55.3	54.9	55.0	55.1	54.8
18	12.5	12.3	12.7	13.5	13.4	12.6	13.4
19	17.8	18.2	19.4	17.5	17.5	64.4	64.7
20	139.7	139.5	139.8	139.1	138.8	139.1	138.8
21	114.6	114.7	114.5	115.2	115.3	114.8	115.4
OAc	170.7	170.8		170.7	170.7	170.6	170.6
					170.7	170.5	170.5
							170.2
OAc	21.4	21.3		21.4	21.2	21.3	21.1
					21.2	21.0	21.0
							21.9

The molecular formula of **1**, a colorless crystalline solid (mp 156−159 °C; $[\alpha]_D$ −84°, $CHCl_3$), was established as $C_{23}H_{34}O_3$ (*m*/*z* 358.2522 Δmmu + 14) from HREIMS data. The ^{13}C-NMR spectrum of **1** revealed 23 well-resolved signals of which four were vinylic (*δ* 114.6, 124.6, 139.7, and 144.3) and one was consistent with an ester carbonyl (*δ* 170.7). This accounted for three of the seven degrees of unsaturation implied by the molecular formula and required the basic skeleton of **1** to be tetracyclic. The ^{13}C-NMR data of **1** (see Table 1), supported by a DEPT NMR experiment, were consistent with the reported values for a tetracyclic, pregnadiene skeleton.[2,6]

The ^{1}H-NMR spectrum of **1** showed signals indicative of a single acetate methyl group (*δ* 2.0) and the two angular methyl groups (*δ* 0.60 and 1.0) of a pregn-20-ene sterol.[2] Further evidence for the exocyclic Δ^{20} olefin

* To whom correspondence should be addressed. Phone: 27-461-318264. FAX: 27-461-25109. E-mail: chdc@warthog.ru.ac.za.
[†] Rhodes University.
[‡] SmithKline Beecham.
[⊗] Abstract published in *Advance ACS Abstracts,* May 15, 1997.

S0163-3864(96)00684-2 CCC: $14.00

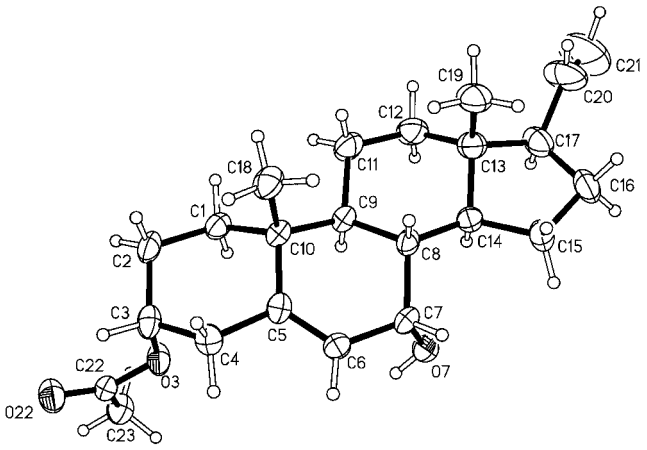


Figure 1. X-ray structure of **1**.

in **1** followed from the characteristic triplet of doublets at δ 5.77 (H-20) and the methylene multiplet at δ 5.0 (H-21) in the ^{1}H-NMR spectrum of this compound.[2] The ^{13}C chemical shifts of the two exocyclic vinylic carbons C-20 and C-21 (δ 139.7 and 114.6) were also consistent with published values[2,6] for pregn-20-enes. The two remaining vinylic ^{13}C signals, a methine and a quaternary carbon (δ 124.6 and 144.3), required the endocyclic olefin in **1** to be at a ring junction, thus limiting the position of this functionality to either Δ^4, Δ^5, Δ^6, $\Delta^{9,11}$, or Δ^{14}. This is at variance with the pregna-1,20-diene compounds normally isolated from soft corals.[2,6]

A combination of HMQC, HMBC, and COSY NMR experiments placed the quarternary vinylic carbon at C-5. Although extensive overlap of the ^{1}H-NMR signals in the methylene envelope between δ 1.1 and 1.9 hampered an unequivocal assignment of either a Δ^4 or Δ^5 olefin, the partial structure on either side of the endocyclic olefin could be delineated by 2D NMR as follows. A prominent COSY correlation was observed between the olefinic proton (δ 5.55) and an allylic oxymethine proton (δ 3.84), while an HMQC-NMR experiment coupled this latter proton to the ^{13}C-NMR signal at δ 64.4 . The second oxymethine carbon (δ 70.3) was conversely coupled to a methine proton (δ 5.05), which in turn was placed in a homoallylic position from, first, a three-bond HMBC correlation between this proton and C-5 and, second, from a COSY coupling to two deshielded, allylic methylene protons (δ 2.25 and 2.55). The downfield chemical shift of the methine proton at δ_H 5.05 placed the acetate group in the homoallylic position and thus confined the single hydroxyl moiety to the allylic oxymethine carbon.

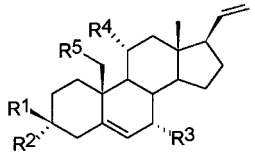
In order to settle the uncertainty in the position of the endocyclic olefin and at the same time determine the stereochemistry of **1**, an X-ray analysis was carried out. The stereoscopic drawing of **1**, shown in Figure 1, established a pregna-5,20-diene-3α,7α-diol 3α-acetate structure for this compound. The atomic coordinates are presented in Table 2. The C5−C6 bond distance of 1.325(5) Å is consistent with a double bond at this position, as are the relevant bond angles. The unusually short exocyclic C20−C21 bond length (1.18 Å) and large C17−C20−C21 bond angle (133°) are attributable to positional disorder for C21. Attempts to refine a disorder model, however, were unsuccessful.

LAH reduction of **1** gave the diol **8** as a colorless oil ($[\alpha]_D$ −122°, $CHCl_3$). The ^{1}H-NMR spectrum of **8** contained a broad singlet (δ 4.1, $W_{1/2}$ = 10 Hz)[2] assigned to the 3β proton (a 3α proton requires $W_{1/2}$ = 25 Hz),[7] thus confirming the α configuration of the oxygen functional group at C-3. Compound **2** was shown by HREIMS data ($C_{23}H_{34}O_3$ 358.2502 Δmmu −6) to be isomeric with **1**. An HMBC correlation between the allylic oxymethine proton and the acetate carbonyl signal at δ 170.8 placed the acetate functionality at C-7 in this compound. The α orientation of the acetate followed from the allylic coupling constant ($J_{6,7}$ = 4.9 Hz), which was in accordance with that observed for **1** ($J_{6,7}$ = 5.5 Hz), while the $W_{1/2}$ value (21 Hz) of the H-3 proton multiplet in the ^{1}H-NMR spectrum of **2** confirmed the 3β configuration of the hydroxyl moiety. Interestingly, the resonances attributed to the axial and equatorial allylic protons at C-4 in **8** (δ 2.60 and 2.20) and **1** (δ 2.55 and δ 2.25) were clearly separated, while in **2** the two signals had coalesced to form a complex multiplet centered at δ 2.30. This ^{1}H-NMR spectral characteristic of the C-4 methylene protons proved a particularly useful method for quickly assigning an α or β configuration to the oxygen moiety at C-3 in this series of pregna-5,20-diene compounds.

Table 2. Atomic Coordinates [× 10^4] and Equivalent Isotropic Displacement Parameter [Å × 10^3] for **1** (esd's in parentheses)

atom	*x*/*a*	*y*/*b*	*z*/*c*	*U*(eq)[a]
O3	6096(1)	4376(5)	364(1)	36(1)
O7	3278(1)	4659(5)	1733(1)	35(1)
O22	6999(2)	4987(6)	−383(1)	41(1)
C1	6477(2)	3452(7)	1690(2)	31(1)
C2	7110(2)	4561(8)	1280(2)	35(1)
C3	6600(2)	5885(7)	779(2)	33(1)
C4	5947(2)	7433(7)	1044(2)	31(1)
C5	5390(2)	6470(6)	1525(2)	27(1)
C6	4526(2)	6936(7)	1516(2)	30(1)
C7	3917(2)	6222(7)	1986(2)	30(1)
C8	4446(2)	5271(7)	2559(2)	26(1)
C9	5191(2)	3765(7)	2363(2)	27(1)
C10	5879(2)	5039(7)	2019(2)	27(1)
C11	3635(2)	2409(8)	2908(2)	41(1)
C12	4962(3)	1377(9)	3302(2)	44(1)
C13	4341(2)	3054(8)	3538(2)	37(1)
C14	3844(2)	4117(7)	2973(2)	30(1)
C15	3091(2)	5390(8)	3235(2)	38(1)
C16	2844(3)	4024(10)	3790(2)	45(1)
C17	3536(3)	2168(9)	3859(2)	43(1)
C18	6483(2)	6479(8)	2463(2)	38(1)
C19	4869(3)	4682(10)	3968(2)	48(1)
C20	3723(3)	1369(13)	4515(2)	75(2)
C21	3516(5)	−210(22)	4766(4)	129(4)
C22	6408(2)	3976(7)	−179(2)	30(1)
C23	5947(2)	2105(8)	−503(2)	37(1)

[a] *U*(eq) is defined as one third of the trace of the orthogonalized U_{ij} tensor.



1: $R^1 = R^4 = R^5 = H$, $R^2 = OAc$, $R^3 = OH$
2: $R^1 = OH$, $R^2 = R^4 = R^5 = H$, $R^3 = OAc$
3: $R^1 = OH$, $R^2 = R^3 = R^4 = R^5 = H$
4: $R^1 = R^5 = H$, $R^2 = OAc$, $R^3 = R^4 = OH$
5: $R^1 = R^5 = H$, $R^2 = R^3 = OAc$, $R^4 = OH$
6: $R^1 = R^4 = H$, $R^2 = R^5 = OAc$, $R^3 = OH$
7: $R^1 = H$, $R^2 = R^3 = R^5 = OAc$, $R^4 = OH$
8: $R^1 = R^4 = R^5 = H$, $R^2 = R^3 = OH$

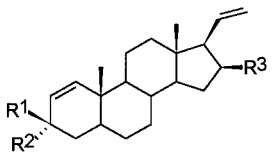
The molecular ion at *m*/*z* 300.2470 (Δmmu +17) in the HREIMS of **3** ($[\alpha]_D$ −58°) is compatible with the

molecular formula $C_{21}H_{32}O$, which corresponds to the loss of an acetoxy moiety and associated secondary alcohol from **2**. The ^{13}C-NMR data of **3** accordingly only differed from that of **2** in the replacement of oxymethine signal at δ 71.7 with a methylene resonance (δ 32.05). The ^{1}H-NMR data of **3** also revealed the loss of the acetate methyl singlet, while both the coalesced allylic methylene signal (δ 2.30) and the broad oxymethine singlet (δ 3.55 $W_{1/2}$ = 22 Hz) supported a 3β hydroxyl group in this compound. The NMR data of **3** from *P. faurea* are consistent with those reported for pregna-5,20-dien-3β-ol ($[\alpha]_D$ −62°) isolated from the Alcyonacean octocoral *Gersemia rubiformis*[8] and the aglycon ($[\alpha]_D$ −44°) of muricin-1 from *Muricea fruticosa*.[9]

The ^{1}H-NMR spectra of compounds **4** and **5** were very similar, differing only in the presence of an additional acetate methyl group (δ 1.95) in the spectrum of the latter compound. An extra oxymethine proton signal (δ 4.1) and the downfield shift of the H-9 resonance (δ 2.0) were the two significant differences between the ^{1}H-NMR spectra of **4** and **5** and the major compound **1**. The deshielded H-9 signal in the ^{1}H-NMR spectra of compounds **4** and **5** suggested an oxygen substituent in ring C. A combination of COSY and selected NOE difference experiments placed this functionality at C-11 and also established the β configuration of H-11. The molecular formulae of **4**, $C_{25}H_{34}O_3$ (*m/z* 358.2502 Δmmu −6) and **5**, $C_{23}H_{34}O_4$ (*m/z* 374.2450 Δmmu −7) were determined from HREIMS data. The three-bond HMBC correlation from the deshielded H-3 methine proton (δ 5.05) to the single acetate carbonyl resonance (δ 170.7) placed the acetate group at C-3. The chemical shift and splitting pattern of the C-4 methylene protons in the ^{1}H-NMR spectrum of **4** were consistent with **1** and thus supported a 3α configuration for the acetate moiety. The positioning of the single acetate moiety in **4** required the oxygen functionality at C-11 to be a hydroxyl group, while the remaining hydroxyl group, implied by the molecular formula, was placed at C-7 from a strong COSY correlation between H-6 and H-7. The β configuration of the H-7 oxymethine proton followed from the $J_{6,7}$ coupling constant (5.5 Hz), which was consistent with that observed for compounds **1** and **2**. The two α-acetate groups in **5** were similarly placed at C-3 and C-7 from the NMR data, while NOE difference experiments confirmed the 11α orientation of the single hydroxyl group in this compound.

HREIMS data established the molecular formulas of compounds **6** and **7** as $C_{25}H_{36}O_5$ (416.2568 Δmmu +5) and $C_{27}H_{38}O_7$ (474.2616 Δmmu −19), respectively. The absence of the characteristic resonance for the C-19 methyl group and the presence of two deshielded oxymethylene doublets (δ 3.94 and 4.53, *J* 11.9 Hz) in the ^{1}H-NMR spectra of both **6** and **7** suggested oxygenation at C-19. HMBC correlations from the two oxymethylene protons to an acetate carbonyl (δ 170.6) unequivocally placed an acetate group at this position in both these compounds. The positions and α configurations of the C-3 acetate and C-7 hydroxyl group in **6** and C-3, C-7 diacetoxy and C-11 hydroxyl moieties in **7** were established using the same methodology described for compounds **4** and **5**. The ^{13}C-NMR data for compounds **1**−**7** is presented in Table 1.

Pregnadiene compounds are rare in the marine environment, with only compounds **3** and **9**−**13** being previously reported from soft corals of the genera *Gersemia, Muricea, Capnella*, and an unidentified soft coral collected at Canton Island.[2,6,8,9] The biological role of compounds **1**−**7** is unknown.



9: $R^1 + R^2$ = O, R^3 = H
10: $R^1 + R^2$ = O, R^3 = OAc
11: $R^1 + R^2$ = O, R^3 = OH
12: R^1 = H, $R^2 = R^3$ = OAc
13: R^1 = H, $R^2 = R^3$ = OH

Experimental Section

General Experimental Procedures. Normal-phase HPLC was performed using a Whatman Partisil-10 column. The NMR and IR spectra were recorded on Bruker AMX400 and Perkin-Elmer 180 spectrometers, respectively. Optical rotatations were measured on a Perkin-Elmer 141 polarimeter. LRMS were recorded on a Hewlett-Packard 5988a spectrometer, while HRMS were obtained by Dr P. Boshoff of the Mass Spectrometry Unit at the Cape Technikon, Cape Town.

Invertebrate Material. The soft coral was collected in September 1994, by scuba (−15m), off Port Alfred, South Africa. The animal was identified as *Pieterfaurea unilobata* (class Anthozoa, order Alcyonacea Lamaouroux, 1816, family Nidaliidae) by Dr. Gary Williams. *P. unilobata* is distinguished by the possession of conspicuous permanent conical protuberances formed by a palisade-like arrangement of large spindles. Large tuberculate and clubbed spindles are present throughout the surface as well as in the interior of the colony, while anthocodial sclerites are absent.[10] Voucher specimens are deposited at the California Academy of Science and Rhodes University marine invertebrate collection (PA94-003).

Extraction and Isolation. The frozen specimens were diced and freeze-dried to give 143 g of dried material, which was extracted twice with EtOAc (1.25 L). Removal of the solvent under reduced pressure gave a viscous, dark-orange oil (2.3 g). Flash chromatography of this oil on Si gel using a solvent gradient of increasing polarity (*n*-hexane−EtOAc, 4:1, 3:1, 2:1, 1:1, 0:1) gave six fractions (A−F) as ajudged by TLC.

Compounds **1** (128 mg) and **5** (5 mg) were obtained from fraction D (313 mg) by recrystallization (*n*-hexane−EtOAc, 7:1.5) followed by HPLC (*n*-hexane−EtOAc, 7:2). HPLC (*n*-hexane−EtOAc, 7:1) of fraction E (193 mg) gave sterols **2** (19 mg), **6** (10 mg), and **7** (11 mg). Compound **3** (5 mg) was purified from fraction C (321 mg, mainly cholesterol) by HPLC (*n*-hexane−EtOAc, 3:2), while **4** (3.7mg) was recrystallized from fraction F (99 mg, *n*-hexane−EtOAc, 7:3).

Pregna-5,20 diene-3α,7α-diol 3α-acetate (1): colorless crystalline plates (*n*-hexane−EtOAc mp 156−159 °C); $[\alpha]^{25}_D$ −84° (*c* 0.02, $CHCl_3$); IR (KBr disk) ν_{max} 3490 (OH), 2940, 1700 (Ac C=O), 1380, 1285, 1030, 1010, 890 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 5.77 (1H, m, H-20), 5.55 (1H, dd, *J* = 5.55, 1.8 Hz, H-6), 5.05 (1H, m, H-3), 5.0 (2H, m, H-21), 3.84 (1H, br s, $W_{1/2}$ = 14 Hz, H-7), 2.55 (1H, dd, *J* = 15.3, 1.5 Hz, H-4), 2.25 (1H, dd, *J* = 15.3, 1.5

Hz, H-4), 2.03 (1H, m, H-17), 2.0 (3H, s, OAc), 1.85 (1H, m, H-15), 1.80 (2H, m, H-2), 1.72 (1H, m, H-12), 1.68 (1H, m, H-1), 1.60 (2H, m, H-11, H-16), 1.50 (2H, m, H-14, H-8), 1.45 (2H, m,H-1, H-11), 1.35 (1H, m, H-9), 1.25 (1H, m, H-15), 1.1 (1H, m, H-12), 1.0 (3H, s, CH_3-19), 0.60 (3H, s, CH_3-18); LREIMS (70 eV) *m/z* no M^+ 298 (100), 280 (19), 265 (15), 105 (16), 93 (17), 91 (25); HREIMS *m/z* 358.2522 ($C_{23}H_{34}O_3$ requires 358.2508).

Pregna-5,20 diene-3*β*,7α-diol 7α-acetate (2): yellow oil; $[\alpha]^{25}_D$ −198° (*c* 0.17, $CHCl_3$); IR (dry film) ν_{max} 3400 (OH), 2940, 1710 (Ac C=O), 1360, 1240, 1200, 1040, 100, 880 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ 5.75 (1H, m, H-20), 5.55 (1H, dd, J = 1.6, 5.2 Hz, H-6), 5.0 (3H, m, H-7, CH_2-21), 3.55 (1H, m, $W_{1/2}$ = 21 Hz H-3), 2.30 (2H, m, CH_2-4), 2.01 (1H, m, H-17), 2.0 (3H, s, OAc), 1.85 (2H, m, H-2, H-12), 1.80 (2H, m, H-1, H-16), 1.70 (1H, m, H-12), 1.60 (1H, m, H-11), 1.57 (2H, m, H-1, H-15), 1.52 (1H, m, H-2), 1.45 (1H, m, H-11), 1.37 (1H, m, H-9), 1.35 (1H, m, H-14), 1.20 (1H, m, H-15), 1.13 (1H, m, H-8), 1.00 (3H, s, CH_3-19), 0.60 (3H, s, CH_3-18); LREIMS (70 eV) *m/z* 298 (94), 265 (100), 211 (61), 143 (55), 105 (54), 91 (81); HREIMS *m/z* 358.2502 ($C_{23}H_{34}O_3$ requires 358.2508).

Pregna-5,20-dien-3*β*-ol (3): colorless oil; $[\alpha]^{25}_D$ −58° (*c* 0.05, $CHCl_3$); IR (dry film) ν_{max} 3020 (OH), 1210, 740 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ 5.75 (1H, m, H-20), 5.35 (1H, m, H-6), 5.0 (2H, m, CH_2-21), 3.55 (1H, m, $W_{1/2}$ = 22.2 Hz, H-3), 2.30 (2H, m, CH_2-4), 2.0 (2H, m, H-7, H-17), 1.85 (2H, m, H-1, H-2), 1.73 (2H, m, H-12, H-15), 1.55 (4H, m, H-7, H-11, H-16), 1.50 (2H, m, H-1, H-2), 1.45 (1H, m, H-11), 1.23 (1H, m, 15), 1.10 (2H, m, H-8, H-12), 1.03 (1H, m, H-14), 1.0 (3H, s, CH_3-19), 0.97 (1H, m, H-9), 0.6 (3H, s, CH_3-18); LREIMS (70 eV) *m/z* 267 no M^+(52), 213 (30), 145 (33), 107 (28), 105 (40), 91 (44), 85 (52), 83 (100); HREIMS *m/z* 300.2470 ($C_{21}H_{32}O$ requires 300.2453).

Pregna-5,20-diene-3α,7α,11α-triol 3α-acetate (4): white powder; mp 226−229 °C (with dec); $[\alpha]^{25}_D$ −65° (*c* 0.08, $CHCl_3$); IR (KBr disk) ν_{max} 3300 (OH), 2920, 1760 (Ac C=O), 1260 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ 5.75 (1H, m, H-20), 5.60 (1H, m, H-6), 5.05 (1H, m, H-3), 5.0 (2H, m, CH_2-21), 4.1 (1H, br s, $W_{1/2}$ = 22.6 Hz, H-11), 3.8 (1H, br s, $W_{1/2}$ = 13.3, H-7), 2.57 (1H, dd, J = 1.6, 15.1 Hz, H-4), 2.42 (1H, m, H-1), 2.27 (1H, dd, J = 1.6, 15.1 Hz, H-4), 2.06 (1H, m, H-17), 2.05 (3H, s, OAc), 2.02 (1H, m, H-9), 1.9 (1H, m, H-16), 1.85 (1H, m, H-15), 1.80 (2H, m, CH_2-2), 1.60 (1H, m, H-16), 1.57 (2H, m, H-1, H-12), 1.45 (2H, m, H-8, H-14), 1.25 (3H, s, CH_3-19), 1.23 (1H, m, H-15), 1.1 (1H, m, H-12), 0.65 (3H, s, CH_3-18); LREIMS (70 eV) *m/z* no M^+ 314 (100), 157 (10), 145 (16), 121 (11), 107 (17), 105 (17), 93 (13), 91 (22); HREIMS *m/z* 374.2450 ($C_{23}H_{34}O_4$ requires 374.2457).

Pregna-5,20 diene-3α,7α,11α-triol 3α,7α-diacetate (5): colorless oil; $[\alpha]^{25}_D$ −113° (*c* 0.09, $CHCl_3$); IR (dry film) ν_{max} 3450 (OH), 2940, 1750 (Ac C=O), 1450, 1420, 1370, 1240, 1120 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ 5.73 (1H, m, H-20), 5.6 (1H, dd, H-6), 5.0 (2H, m, CH_2-21), 4.95 (1H, m, H-3), 4.92 (1H, m, H-7), 4.10 (1H, br s, H-11), 2.50 (1H, dd, H-4), 2.35 (1H, m, H-1), 2.25 (1H, dd, H-4), 2.10 (1H, m, H-17), 2.06 (3H, s, OAc), 2.0 (1H, m, H-9), 1.95 (3H, s, OAc), 1.85 (1H, m, H-15), 1.75 (2H, m, CH_2-2), 1.65 (1H, m, H-8), 1.62 (2H, m, H-15, H-16), 1.58 (2H, m, H-1, H-14), 1.43 (1H, m, H-12), 1.20 (1H, m, H-16), 1.17 (3H, s, CH_3-19), 1.15 (1H, m, H-12), 0.60 (3H, s, CH_3-18); LREIMS (70 eV) *m/z* no M^+, 314 (100), 298 (29), 296 (22), 149 (42), 145 (30), 91 (33); HREIMS *m/z* 416.2563 ($C_{25}H_{36}O_5$ requires 416.2563).

Pregna-5,20 diene-3α,7α,19-triol 3α,19-diacetate (6): colorless oil; $[\alpha]^{25}_D$ −82° (*c* 0.04, $CHCl_3$); IR (dry film) ν_{max} 3500 (OH), 2930, 1730(Ac C=O), 1365, 1230, 1030, 1010, 950, 890 cm^{-1}; ^{1}H-NMR ($CDCl_3$) δ 5.80 (1H, m, H-6), 5.75 (1H, m, H-20), 5.07 (1H, m, H-3), 5.00 (2H, m, CH_2-21), 4.55 (1H, d, J = 11.9 Hz, H-19), 3.95 (1H, d, J = 11.9 Hz, H-19), 3.80 (1H, br s, $W_{1/2}$ = 11.8 Hz H-7), 2.60 (1H, d, J = 15.2 Hz, H-4), 2.30 (1H, d, J = 15.4 Hz, H-4), 2.10 (6H, s, 2 × OAc), 2.0 (1H, m, H-17), 1.90 (2H, m, H-1, H-16), 1.87 (1H, m, H-15), 1.85 (1H, m, H-8), 1.80 (1H, m, H- 2), 1.75 (1H, m, H-12), 1.70 (1H, m, H-2), 1.67 (1H, m, H-11), 1.60 (1H, m, H-16), 1.50 (2H, m, H-1, H-11), 1.45 (1H, m, H-14), 1.42 (1H, m, H-9), 1.27 (1H, m, H-15), 1.10 (1H, m, H-12), 0.65 (3H, s, CH_3-18); LREIMS (70 eV) *m/z* no M^+, 356 (83), 296 (86), 265 (70), 143 (72), 105 (71), 91 (98), 83 (100); HREIMS *m/z* 416.2568 ($C_{25}H_{36}O_5$ requires 416.2563).

Pregna-5,20 diene-3α,7α,11α,19-tetrol 3α,7α19-triacetate (7): a yellow oil; $[\alpha]^{25}_D$ −114° (*c* 0.013, $CHCl_3$); IR (dry film) ν_{max} 3500, 2930, 1750, 1365, 1240, 1010 cm^{-1}; ^{1}H-NMR($CDCl_3$) δ 5.85 (1H, d, J = 4.4 Hz, H-6), 5.75 (1H, m, H-20), 5.03 (1H, m, H-3), 5.00 (2H, m, CH_2-21), 4.95 (1H, m, H-7), 4.85 (1H, dd, J = 11.9 Hz, H-19), 4.22 (1H, dd, J = 11.9 Hz, H-19), 4.13 (1H, br s, H-11), 2.60 (1H, d, J = 14.9 Hz, H-4), 2.56 (1H, m, H-1), 2.30 (1H, d, J = 15.0 Hz, H-4), 2.10 (1H, m, H-17), 2.05 (3H, s, OAc), 2.02 (3H, s, OAc), 2.0 (1H, m, H-12), 1.95 (3H, s, OAc), 1.87 (1H, m, H-9), 1.85 (1H, m, H-16), 1.80 (1H, m, H-8), 1.70 (2H, m, CH_2-2), 1.63 (2H, m, H-14, H-16), 1.60 (2H, m, H-1, H-15), 1.20 (1H, m, H-15), 1.18 (1H, m, H-12), 0.65 (3H, s, H-18); LREIMS (70 eV) *m/z* no M^+ 312 (75), 276 (67), 263 (86), 143 (76), 141 (86), 131 (78), 129 (63), 91 (100); HREIMS *m/z* 474.2616 ($C_{27}H_{38}O_7$ requires 474.2635).

LAH Reduction of Compound 1. LAH (50 mg) was added to stirred solution of **1** (19 mg) in dry ether (6 mL). The solution was cooled in ice and 5% KOH (8 mL) added. The basic solution was extracted with ether, dried over anhydrous Na_2SO_4, filtered, and concentrated to yield pregna-5,20-dien-3α,7α-diol as a clear oil (15 mg): $[\alpha]^{25}_D$ −122° (*c* 0.08, $CHCl_3$); IR (dry film) ν_{max} 3360(OH), 2940, 1420, 1370, 1210, 1110, 1000, 985, 890, 745 cm^{-1}; ^{1}H-NMR ($CDCl_3$); δ 5.75 (1H, m, H-20), 5.60 (1H, m, H-6), 4.98 (2H, m, CH_2-21), 4.10 (1H, br s, H-3), 3.85 (1H, br s, H-7), 2.60 (1H, m, H-4), 2.20 (1H, m, H-4), 2.05 (1H, m, H-17), 1.90 (1H, m, H-16), 1.89 (1H, m, H-15), 1.80 (1H, m, H-2), 1.75 (1H, m, H-12), 1.65 (2H, m, H-1, H-11), 1.60 (1H, m, H-16), 1.58 (1H, m, H-1), 1.50 (1H, m, H-14), 1.45 (1H, m, H-11), 1.43 (1H, m, H-9), 1.42 (1H, m, H-8), 1.25 (1H, m, H-2), 1.23 (1H, m, H-15), 1.10 (1H, m, H-12), 1.00 (3H, s, CH_3-19), 0.60 (3H, s, CH_3-18); ^{13}C-NMR (100 MHz, $CDCl_3$) δ 12.6 (q, C-18), 17.8 (q, C-19), 20.2 (t, C-11), 24.8 (t, C-15), 27.3 (t, C-16), 28.8 (t, C-2), 33.1 (t, C-1), 37.0 (t, C-12), 37.8 (d, C-8), 38.2 (s, C-10), 39.6 (t, C-4), 42.7 (d, C-9), 43.2 (s, C-13), 48.7 (d, C-14), 55.3 (d, C-17), 64.3 (d, C-7), 67.4 (d, C-3), 114.6 (t, C-21), 125.6 (d, C-6), 139.8 (d, C-20), 144.0 (s, C-5); LREIMS *m/z* no M^+ 298 (100), 265 (33), 145 (230, 119 (23), 105 (26), 93 (22), 91 (35).

X-ray Diffraction of 1. A suitable crystal was flash cooled in a stream of N_2 gas to 223(2)K. Lattice parameters were determined from the setting angles of 25 reflections well distributed in reciprocal space meas-

ured on an Enraf Nonius CAD-4 diffractometer. Intensity data were collected on the diffractometer using graphite monochromated molybdenum radiation and an ω-2θ variable speed scan technique. Three orientation controls were monitored to assess any crystal movement during the experiment. The intensities of three reflections measured at the beginning, end, and every 3 h of exposure time showed a variation of 4.5%. Data were corrected for this variation and for Lorentz and polarization effects. Equivalent reflections, but not Friedel mates, were averaged. The structure was solved by direct methods using the SHELXS program[11] and refined using the SHELXL-93 program.[12] Positions for non-hydrogen atoms were eventually refined with anisotropic displacement parameters. With the exception of H-7 (which was refined) the hydrogen atoms were included in idealized positions riding on the atom to which they are attached with isotropic displacement factors assigned as a constant (1.2) times $\boldsymbol{U}_{\mathbf{eq}}$ of the attached atom. The full-matrix least-squares refinement (on F^2) of 239 parameters converged ($\Delta/\sigma_{max} = 0.00$) to values of the conventional crystallographic residuals $R = 0.0482$ for 2053 observed data [$I > 2\sigma(I)$] and $R = 0.0674$ ($wR_2 = 0.1610$) for all 2412 data. The function minimized was $\Sigma w (F_o^2 - F_c^2)^2$. Weights, w, were eventually assigned to the data as $w = 1/[\sigma^2(F_o^2) + (0.0999P)^2 + 1.7571P]$, where $P = [MAX(F_o^2,0) + 2F_c^2]/3$. A final difference Fourier map showed residual density between + 0.36 and −0.25eÅ^{-3}. Values of the neutral atom scattering factors and real and imaginary dispersion corrections were taken from the *International Tables for X-ray Crystallography*.[13]

Crystal data:[14] clear colorless prisms; monoclinic; unit cell dimensions a, 14.989(1) Å; b 6.211(1) Å; c 21.715(1) Å; β 95.26(1)°; volume 2013.1(4) Å^3; space group C2; Z 4; density (calcd) 1.183 Mg m^{-3}; $F(000)$ 784; linear absorption coefficient μ(Mo Kα, $\lambda = 0.710\ 73$ Å) 0.076 mm^{-1}.

Acknowledgment. We would like to thank Dr. Gary Williams of the California Academy of Sciences for identifying *P. unilobata* and both Mr. Dennis Croukamp of the Kowie Dive School and Mr. G. Hooper of Rhodes University for their assistance with the collection of *P. unilobata*. Financial support for this research from Rhodes University, the Foundation for Research Development and SmithKline Beecham Pharmaceuticals (USA) is gratefully acknowledged.

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(14) X-ray data for compound **1** have been deposited at the Cambridge Crystallographic Data Centre and are available upon request from Dr. Olga Kennard, University Chemical Laboratory: 12 Union Road, Cambridge CB2 1EZ, UK.

NP960684H