Withanolides from *Vassobia lorentzii*

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Eight new withanolides were isolated from the aerial parts of *Vassobia lorentzii* and characterized by spectroscopic methods and with the aid of molecular modeling. The compounds were identified as (17*S*,20*R*,22*R*)-5*β*,6*β*:18,20-diepoxy-18-hydroxy-1-oxowitha-2,5,24-trienolide (**1**); (17*S*,20*R*,22*R*)-18,20-epoxy-4*β*,18-dihydroxy-1-oxowitha-2,5,24-trienolide (**2**); (17*S*,18*R*,20*R*,22*R*)-4*β*-hydroxy-18,20-epoxy-18-methoxy-1-oxowitha-2,5,24-trienolide (**3**); (17*S*,18*S*,20*R*,22*R*)-4*β*-hydroxy-18,20-epoxy-18-methoxy-1-oxowitha-2,5,24-trienolide (**4**); (17*S*,20*R*,22*R*)-4*β*-hydroxy-18,20-epoxy-1,18-dioxowitha-2,5,24-trienolide (**5**); (17*S*,18*R*,20*R*,22*R*)-18,20-epoxy-18-methoxy-1,4-dioxowitha-2,5,24-trienolide (**6**); (17*S*,18*S*,20*R*,22*R*)-18,20-epoxy-18-methoxy-1,4-dioxowitha-2,5,24-trienolide (**7**); and (17*S*,20*R*,22*R*)-5*β*,6*β*-epoxy-4*β*,18,20-trihydroxy-1-oxowitha-2,24-dienolide (**8**). Compounds **1** and **2** were obtained as epimeric mixtures at C-18.

Vassobia Rusby is a genus in the Solanaceae represented by five species that grow in South America. It has been related to *Acnistus*, *Dunalia*, and *Iochroma* species,[1] and, considering the different opinions concerning the systematic position of this plant at the trivial level, identification of the withanolides may be important from a chemotaxonomic point of view.

We now report the isolation of eight new withanolides (**1**−**8**) from *Vassobia lorentzii* (Dammer) A. T. Hunziker. All of these compounds have a functionalized C-18 at various oxidation levels (alcohol, aldehyde, and lactone) and they are closely related to those isolated from *Dunalia brachyacantha*[2] and to the withaphysalins from *Physalis minima*.[3] It is noteworthy that all plants originally classified as *Dunalia* (i.e., *Acnistus*, *Dunalia*, *Iochroma*, and *Vassobia*) have yielded 18-oxygenated withanolides.[4] In contrast to a previous report on *V. lorentzii* (at that time *Acnistus lorentzii*),[5] withaferin A was not detected in the present study. Structures of the new compounds were determined using a combination of techniques including 2D NMR, molecular modeling, and chemical transformations.

Results and Discussion

Withaphysalin F (**1**), $C_{28}H_{36}O_7$, did not show a molecular ion in its EIMS but a peak at *m*/*z* 466 (0.3%) corresponding to the $[M - H_2O]^+$ ion was observed; an intense fragment at *m*/*z* 125 (34%) was indicative of an unsaturated *δ*-lactone side chain, typical of many withanolides. The HRFABMS (*m*-nitrobenzyl alcohol, NaCl) showed a $[M + Na]^+$ ion at *m*/*z* 507.2375 that was consistent with the proposed formula. The ^{1}H NMR spectrum of **1** indicated that it was an approximately 1:1 mixture of two stereoisomers, which we were unable to separate (Table 1). Despite this, the low-field part of the spectrum, in conjunction with the H-4 doublet at *δ* 3.77 (*J* = 5.8 Hz), indicated a 4*β*-hydroxy-2-en-1-one system in ring A, identical to that described for withaferin A. The H-6 signal at *δ* 3.24 confirmed the presence of a 5*β*,6*β*-epoxide.[6] The rest of the spectrum was identical to that of lactol **9**,[2] the only difference being absence of the acetate at C-4. The ^{13}C NMR spectrum of **1** (Table 2) was also almost identical to that of **9**, except for the resonances of carbons C-1 to C-6 due to the absence of the C-4 acetate. Lack of a C-18 methyl signal and the presence of two methine carbon resonances at *δ* 101.0 and 103.6 indicated the presence of a mixture of epimeric hemiacetals at C-18. This agreed with two signals at *δ* 5.15 (s) and 5.26 (s) in the ^{1}H NMR spectrum, which were assigned to H-18.

Withaphysalin G (**2**) $C_{28}H_{36}O_6$, showed a $[M + Na]^+$ ion at *m*/*z* 491.2449 in the HRFABMS (*m*-nitrobenzyl alcohol, NaCl). The EIMS did not show a molecular ion, but a peak at *m*/*z* 450 (8%) corresponding to the $[M - H_2O]^+$ ion; an intense fragment at *m*/*z* 125 was indicative of an unsaturated *δ*-lactone side chain. The ^{1}H and ^{13}C NMR spectra of **2** were very similar to those of **1** and indicated that withaphysalin G was also an approximately 1:1 mixture of hemiacetals, epimeric at C-18 (Tables 1 and 2). As in the previous case, these stereoisomers could not be separated, and their spectral data were obtained from the mixture. The ^{1}H NMR spectrum of compound **2** (Table 1) exhibited signals typical of a 2,5-dien-1-one system with a substituent at C-4.[7] A 4*β*-hydroxy group was inferred from the presence of a doublet at *δ* 4.64 (*J* = 4.4 Hz) corresponding to H-4; coupling of the latter to H-3 confirmed the *β* stereochemistry of this substituent.[8] The ^{13}C NMR spectrum (Table 2) showed only four methyl signals and two methine carbons (*δ* 101.1 and 103.8) assigned to C-18 of the epimeric hemiacetals. The ^{1}H and ^{13}C NMR data for rings C−D, the side chain and the *γ*-hemiacetal ring were almost identical to that of **1** and of the 5,6-epoxy analogue recently isolated from *D. brachyacantha* in which the isomer with the higher chemical shift for H-18 had the 18*S* configuration.[2] Acetylation of the epimeric mixture **2** (Ac_2O/pyridine, 25 °C) gave a single diacetyl derivative (**10**). The close similarity of ^{13}C chemical shifts for carbons 13−22 when compared to (18*R*)-**2** supported the 18*S* stereochemistry for diacetate **10**.

The ^{1}H and ^{13}C NMR spectra of compounds **3** and **4** (Tables 1 and 2) were very similar to those of **2**, indicating the same substitution pattern for rings A−C and the side chain. Both compounds showed a single C-18 resonance

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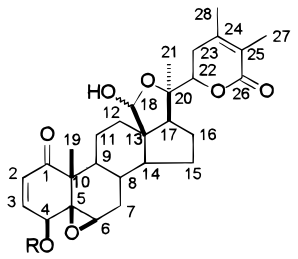
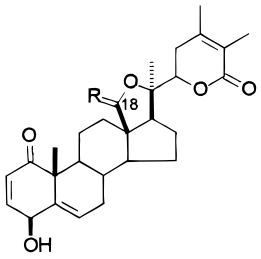
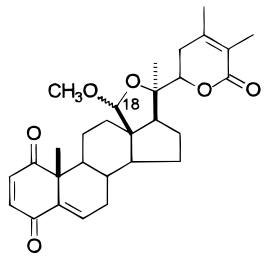


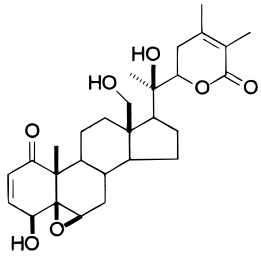
1 R = H; (18*R*/*S*)
9 R = Ac; (18*R*/*S*)



2 R = H, OH; (18*R*/*S*)
3 R = H, OCH_3; (18*R*)
4 R = H, OCH_3; (18*S*)
5 R = O
10 R = H, OAc; (18*S*)



6 18*R*
7 18*S*



8

shifted downfield about 7 ppm with respect to **2**, and an additional methyl signal at δ 55−56. These data were consistent with compounds **3** and **4** being the corresponding 18-methylacetals of each epimer of withaphysalin G (**2**). Withaphysalin H (**3**) showed a singlet in its ^{1}H NMR spectrum at δ 4.62 (H-18) and a sharp three-proton singlet at δ 3.40 corresponding to the 18-methoxy group (assignments confirmed from HETCOR spectrum). As compared with **3**, withaphysalin I (**4**) showed a downfield shift of the H-18 signal (δ 4.75) and upfield shifts of the H-21 (to δ 1.35) and methoxyl (to δ 3.31) resonances. The FABMS (glycerol) of both compounds showed an intense quasimolecular ion [M+1] at *m*/*z* 483, indicating an additional methyl group, and HRFABMS data was consistent with the molecular formula $C_{29}H_{38}O_6$ for both compounds. Intense fragments at *m*/*z* 125 (100%) in the EIMS in both cases indicated a δ-lactone unsaturated side chain.

The stereochemistry at C-18 was determined by the combined analysis of the H-21 shifts, the NOESY spectrum, and molecular modeling of both (18*R* and 18*S*) epimers. Thus, in the 18*R* epimer (**3**), the 1,3-pseudodiaxial relationship between methyl-21 and the 18-methoxy group would give rise to a deshielding effect on the former. In the NOESY spectrum of **4** the correlations observed for the pairs H-12β/H-18 and H-12β/21-CH_3 are only possible for the 18*S* stereoisomer in which H-18 is positioned on the same side as H-12β and H-21. On the other hand, in the NOESY spectrum of **3** a correlation was observed for the pair H-18/H-15β as expected from the geometries obtained by semiempirical AM1 calculations. No correlation was observed for the pair H-18/21-CH_3 in either epimer in accordance with the AM1 calculated structures, which predicted this distance to be larger than 3.5 Å. Thus, the structure of **3** was determined to be (17*S*,18*R*,20*R*,22*R*)-4β-hydroxy-18,20-epoxy-18-methoxy-1-oxowitha-2,5,24-trienolide, and that of **4** as its 18*S* epimer.

The HRFABMS (*m*-nitrobenzyl alcohol, NaCl) of withaphysalin J (**5**) showed a $[M + Na]^+$ ion at *m*/*z* 489.2274 consistent with the formula $C_{28}H_{34}O_6$. The EIMS of **5** showed the molecular ion at *m*/*z* 466 (10%) and a fragment at *m*/*z* 125 (35%), indicative of an α,β-unsaturated δ-lactone ring. The ^{1}H and ^{13}C NMR chemical shifts of compound **5** (Tables 1 and 2) were closely related to those of **2**. The singlet for the 21-methyl at δ 1.53 indicated the presence of an oxygen function at C-20. The lack of an 18-methyl signal in conjunction with the presence of an IR band at 1748 cm^{-1} (γ-lactone carbonyl) and the presence of a carbonyl carbon signal at δ 177.6 in the ^{13}C NMR spectrum (Table 2) led to the conclusion that withaphysalin J (**5**) contained an 18,20-γ-lactone as in other withaphysalins.[9] The ^{1}H and ^{13}C NMR data for rings C−D, the side chain, and γ-lactone ring were coincident with that found for the synthetic lactone obtained by oxidation of an 18,20-hemiacetal of *D. brachyacantha*.[2]

The ^{1}H NMR spectrum (Table 1) of compounds **6** and **7** were almost identical to those of compounds **3** and **4**, with differences in the substitution pattern of rings A and B. In the low-field region, an AB system at δ 6.74 and 6.67 (J = 10.5 Hz) and the proton at δ 6.84 (dd, J = 5.5; 2.3 Hz) suggested the presence of a 2,5-diene-1,4-diketone system in rings A and B.[7] The substitution pattern in ring A was further corroborated by the signals at δ 202.0 and 187.7 in the ^{13}C NMR spectrum (Table 2) that were assigned to C-1 and C-4, respectively. The ^{13}C NMR spectrum of the 18*R* epimer (withaphysalin K, **6**) showed five methyl groups, four of which were coincident with C-21, C-27, C-28, and the methoxyl group of compound **3**. The methine signal at δ 108.1 (C-18) confirmed the presence of a 18-methylacetal group. Its ^{1}H NMR spectrum showed signals for H-18 and the methoxyl group also coincident with those in **3**. The 18*S* epimer (withaphysalin L, **7**) had H-18 shifted downfield to δ 4.75 and showed the expected singlet at δ 3.30 (as in **4**) assigned to the 18-methoxy group. The ^{13}C NMR spectrum also showed a methoxyl group at the same chemical shift as in **4**. The HRFABMS of **6** and **7** gave quasimolecular ions [M + Na] consistent with the molecular formula $C_{29}H_{36}O_6$.

The HRFABMS (*m*-nitrobenzyl alcohol, NaCl) of 18-hydroxy-withanolide D (**8**) showed a $[M + Na]^+$ ion at *m*/*z* 509.2538 consistent with the formula $C_{28}H_{38}O_7$. Compound **8** did not have an 18,20-hemiacetal group but showed in its ^{1}H NMR spectrum (Table 1) only four methyl singlets assigned to C-19, C-21, C-27, and C-28. The missing methyl-18 signal and the appearance of an AB system at δ 3.59 and 3.60 (J = 10.9 Hz) indicated that C-18 was present as a hydroxymethyl group. The ^{1}H resonances were coincident with those of its 18-*O*-acetyl derivative isolated from *Iochroma fuschsioides*,[10] except for H-18 which was shifted upfield, supporting the presence of a free hydroxyl at C-18. The ^{13}C NMR (Table 2) was also almost identical to that of the known 18-acetate, confirming the proposed structure for **8**. The spectral data of **8** were also similar to the 20-deoxy analogue recently isolated from *D. brachyacanta*.[2]

Experimental Section

General Experimental Procedures. ^{1}H and ^{13}C NMR spectra were recorded on a Bruker AC-200 NMR spectrometer at 200.13 and 50.32 MHz, respectively, using $CDCl_3$ as solvent. Multiplicity determinations (DEPT) and 2D spectra (COSY-45, HETCOR, NOESY) were obtained using standard Bruker software. Chemical shifts are given in parts per million (ppm) downfield from TMS as internal standard. EIMS were collected on a VG Trio-2 mass spectrometer at 70 eV by direct inlet; FABMS were measured on a VG ZAB−BEqQ mass spectrometer, and HRFABMS were measured on a VG 7070-HF mass spectrometer. IR and UV spectra were measured on a Nicolet 5-SXC−FTIR and a Shimadzu UV 260 spectrophotometer,

Table 1. ^{1}H NMR Spectral Data for the Relevant Protons of Compounds **1**−**8** and **10** ($CDCl_3$, 200.13 MHz)[a]

proton	**1**[b]	**2**[b]	**3**	**4**	**5**	**6**	**7**	**8**	**10**
2	6.18 d (9.9) [6.19 d (9.9)]	5.95 d (10.0)	5.95 d (10.0)	5.95 d (10.0)	5.96 d (10.0)	6.74 d[c] (10.5)	6.74 d[d] (10.5)	6.19 d (9.9)	6.00 dd (10.0)
3	6.94 dd (9.9; 5.8) [6.78 dd (9.9; 5.8)]	6.76 dd (10.0; 4.4) [6.78 dd (10.0; 4.4)]	6.80 dd (10.0; 4.5)	6.78 dd (10.0; 4.5)	6.79 dd (10.0; 4.4)	6.67 d[c] (10.5)	6.67 d[d] (10.5)	6.92 dd (9.9; 5.5)	6.70 dd (10.0; 4.6)
4α	3.77 d (5.8)	4.64 d (4.4)	4.66 d (4.5)	4.66 d (4.5)	4.65 d (4.4)			3.77 d (5.5)	5.79 d (4.6)
6	3.24 br s [3.26 br s]	5.95 br d (3.7)	5.93 br d (5.5)	5.93 br d (5.5)	5.94 br d (3.7)	6.84 dd (5.5; 2.3)	6.84 dd (6.0; 2.1)	3.24 br s	6.07 br d (4.4)
7α	1.34 m	1.60 m	1.71 m	1.60 m	1.35 m	1.83 m		1.28 m	1.80m
7β	2.21 m	2.15 m	2.18 m	2.19 m	2.23 m	2.37 m	2.37 m	2.15 m	2.22 m
18	5.15 s [5.26 s]	5.18 s [5.30 s]	4.62 s	4.75 s		4.61 s	4.75 s	3.63 d (10.9) 3.59 d (10.9)	6.22 s
19	1.39 s [1.41 s]	1.42 s [1.44 s]	1.44 s	1.43 s	1.53 s	1.35 s	1.34 s	1.43 s	1.48 s
21	1.48 s [1.28 s]	1.49 s [1.29 s]	1.40 s	1.35 s	1.53 s	1.43 s	1.37 s	1.43 s	1.27 s
22	4.43 dd (13.2; 3.4) [4.52 dd (13.2; 3.4)]	4.44 dd (13.3; 3.5) [4.54 dd (13.3; 3.5)]	4.48 dd (13.1; 3.2)	4.42 dd (13.0; 3.0)	4.54 dd (13.2; 3.7)	4.45 dd (13.0; 3.0)	4.45 dd (13.0; 3.0)	4.28 dd (13.1; 3.6)	4.50 dd (13.5; 3.5)
23α	2.02 m	2.05 m [2.20 m]	2.04 m	2.13 m	2.03 m	2.10 m	2.09 m	2.10 m	2.09 m
23β		2.44 m [2.51 m]	2.42 m	2.40 m	2.42 m	2.43 m		2.37 m	2.49 m
27	1.88 br s	1.87 br s	1.89 br s	1.89 br s	1.89 br s	1.89 br s	1.90 br s	1.89 br s	1.89 br s
28	1.93 br s	1.96 br s	1.96 br s	1.94 br s	1.96 br s	1.94 br s	1.94 br s	1.96 br s	1.94 br s
OMe			3.40 s	3.31 s		3.39 s	3.30 s		
OAc									2.07 s 2.08 s

[a] Chemical shifts are in ppm (δ) downfield from TMS, *J* couplings (in parentheses) are in Hz. [b] Chemical shift data correspond to the 18*R* epimer. Distinct resonances for the 18*S* epimer observed in the spectrum of the epimeric mixture are shown in square brackets. [c,d] Assignments may be interchanged.

Table 2. ^{13}C NMR Spectral Data of Compounds **1**−**8** and **10** ($CDCl_3$, 50.32 MHz)

	1[a]		**2**[a]								
carbon	(18*R*)	(18*S*)	(18*R*)	(18*S*)	**3** (18*R*)	**4** (18*S*)	**5**	**6** (18*R*)	**7** (18*S*)	**8**	**10**
1	202.5	202.5	203.6	203.9	203.4	203.6	203.6	202.0	202.3	202.3	203.0
2	131.9	132.0	130.5	131.1	130.4	131.0	130.5	140.2	140.1	132.0	133.2
3	142.1	142.4	143.2	143.0	143.1	143.1	143.0	139.0	139.2	141.9	140.2
4	69.7	69.7	69.2	69.2	69.2	69.3	69.3	187.7	187.7	69.7	69.7
5	63.7	63.9	138.9	138.6	139.0	138.7	139.2	139.6	139.1	63.8	134.5
6	62.6	62.7	128.8	128.8	128.8	128.9	128.9	137.1	137.7	62.8	130.7
7	31.3[b]	31.3	31.1[d]	31.1	31.1[f]	31.3	31.3	31.3	31.4	31.6	31.0[i]
8	30.9	30.3	33.5	32.9	33.8	33.3	30.6	31.0	31.0	29.6	33.2
9	43.9	44.4	42.4	42.9	42.3	42.8	42.0	41.9	42.5	44.5	42.3
10	47.8	47.8	49.4	49.4	49.4	49.3	49.6	51.4	51.4	47.9	49.1
11	24.6	24.6	25.3	25.3	26.1	25.4	22.8	24.5	25.0	22.1	26.3
12	34.6	37.0	34.7	37.2	34.7	36.9	35.3	34.7	34.7	34.8	34.8
13	58.0	58.8	58.0	58.8	57.9	59.0	55.4	58.0	59.2	47.7	57.9
14	56.4[c]	54.7	56.4[e]	54.7	56.4	54.0	55.9	56.0[g]	56.1	55.1[h]	56.5
15	25.4	25.4	25.2	25.2	25.5	24.9	25.9	25.5	25.6	23.7	24.9
16	26.2	27.5	26.2	27.5	26.0	27.1	27.4	26.1	26.2	31.4	25.5
17	57.0[c]	57.0	56.9[e]	56.9	56.3	56.5	52.6	56.3[g]	56.5	55.9[h]	56.5
18	101.0	103.6	101.1	103.8	108.3	110.4	177.6	108.1	110.3	58.9	99.3
19	17.6	17.8	22.9	22.9	22.7	22.8	22.9	23.6	23.7	17.8	21.3
20	85.1	84.8	85.1	84.8	84.8	84.5	83.6	84.8	84.6	75.5	86.6
21	21.8	23.7	21.6	24.5	20.9	23.7	23.4	21.0	23.8	21.6	21.1
22	80.5	80.5	80.6	80.5	80.7	81.5	79.0	80.7	81.4	80.6	80.2
23	31.5[b]	32.1	31.3[d]	32.0	31.0[f]	32.2	31.2	32.7	32.3	31.6	31.3[i]
24	147.7	148.6	148.1	148.9	147.9	148.1	147.2	147.7	147.9	148.4	147.4
25	122.3	122.0	122.2	122.0	122.2	122.2	122.6	122.4	122.4	122.3	122.6
26	165.7	165.7	165.9	165.9	166.0	166.0	165.1	165.9	165.9	165.5	165.9
27	12.5	12.4	12.5	12.4	12.5	12.4	12.5	12.5	12.5	12.5	12.5
28	20.5	20.4	20.5	20.5	20.3	20.0	20.4	20.3	20.4	20.6	20.4
OMe					55.7	55.1		55.7	55.1		
OAc											170.1, 170.2 21.5

[a] Chemical shifts (δ) determined in the *R*/*S* mixture (ca. 1:1). [b−i] Assignments may be interchanged.

respectively. Melting points were obtained on a Fisher-Johns apparatus and are uncorrected. Chromatographic separations were performed by vacuum liquid chromatography (VLC), column chromatography on Si gel 60 (40−63 μm), and preparative TLC (Si gel 60 G F_{254} 2 mm thick).

Plant Material. Aerial parts of *V. lorentzii* were collected in November 1994, in Department of Andalgalá, Catamarca Province, Argentina. A voucher specimen is deposited at the Museo Botánico, Universidad Nacional de Córdoba [CORD].

Extraction and Isolation. The dried and pulverized aerial part of *V. lorentzii* (1.83 kg) was extracted with EtOH at room temperature, the solvent was evaporated, and the residue (78 g) was partitioned between hexane, MeOH, and H_2O (30:3:1). The aqueous MeOH layer was washed with hexane, concentrated, and extracted with $CHCl_3$. The residue (21.5 g) obtained after evaporation of the solvent was fractionated by VLC eluting with hexanes−EtOAc mixtures of increasing polarity (100:0−0:100) to yield four fractions containing withanolides.

These fractions, eluted with hexanes−EtOAc from 50:50 to 10:90, were further fractionated by flash chromatography to yield a fraction (354 mg) containing the mixture **2** of epimers as the major component. The minor fractions were purified by flash chromatography and preparative TLC to yield withanolides **1** (8.3 mg), **3** (22 mg), **4** (6 mg), **5** (6 mg), **6** (3 mg), **7** (4 mg), and **8** (90 mg).

(17*S*,20*R*,22*R*)-5β,6β:18,20-diepoxy-18-hydroxy-1-oxowitha-2,5,24-trienolide (1, 18*R* and 18*S*): white crystals (hexanes−EtOAc); mp 256−258 °C; $[\alpha]^{25}_D$ +46.9° (*c* 0.06, $CHCl_3$); UV (MeOH) λ_{max} 225 nm; IR (film) ν_{max} 3485, 2978, 2825, 1701, 1689 cm^{-1}; ^1H and ^{13}C NMR, (Tables 1 and 2; EIMS *m*/*z* $[M]^+$ absent, 466 (0.3) $[M - H_2O]^+$, 152 (6), 125 (34); FABMS (glycerol, KCl) *m*/*z* 523 (100) $[M + K]^+$, 467 (44) $[M - H_2O + 1]^+$; HRFABMS (*m*-nitrobenzyl alcohol, NaCl) *m*/*z* $[M + Na]^+$ 507.2375 (calcd for $C_{28}H_{36}O_7Na$ 507.2359).

(17*S*,20*R*,22*R*)-18,20-Epoxy-4β,18-dihydroxy-1-oxowitha-2,5,24-trienolide (2, 18*R* and 18*S*): white crystals (hexanes−EtOAc); mp 264−266 C° (dec); $[\alpha]^{25}_D$ +97.6° (*c* 0.13, MeOH); UV (MeOH) λ_{max} 224 nm; IR (film) ν_{max} 3350, 2958, 2849, 1703, 1684 cm^{-1}; ^1H and ^{13}C NMR, Tables 1 and 2; EIMS *m*/*z* $[M]^+$ absent, 450 (8) $[M - H_2O]^+$, 432 (8), 343 (17), 171 (45), 152 (46), 125 (94), 105 (62), 91 (100) and 55 (81); FABMS (glycerol, NaCl) *m*/*z* 491 (17) $[M + Na]^+$, 451 (17) $[M - H_2O + 1]^+$; HRFABMS (*m*-nitrobenzyl alcohol, NaCl) *m*/*z* $[M + Na]^+$ 491.2449 (calcd for $C_{28}H_{36}O_6Na$ 491.2410).

Acetylation of Compound 2. The mixture **2** (10 mg) was dissolved in Ac_2O/pyridine (1:1, 0.3 mL) and left for 18 h at 25 °C. Dilution with CH_2Cl_2 and evaporation under a stream of nitrogen afforded acetate **10** as a white powder; ^1H and ^{13}C NMR data, see Tables 1 and 2.

(17*S*,18*R*,20*R*,22*R*)-4β-Hydroxy-18,20-epoxy-18-methoxy-1-oxowitha-2,5,24-trienolide (3): white amorphous powder; $[\alpha]^{25}_D$ +47.9° (*c* 0.20, $CHCl_3$); UV (MeOH) λ_{max} 224 nm; IR (film) ν_{max} 3427, 2953, 2881, 2828, 1704, 1687, 1666 cm^{-1}; ^1H and ^{13}C NMR, Tables 1 and 2; EIMS *m*/*z* $[M]^+$ absent, 450 (3) $[M - CH_3OH]^+$, 433 (8), 432 (14), 406 (7), 388 (4), 357 (3), 313 (3), 171 (47), 169 (17), 152 (48), 125 (100); FABMS (glycerol) *m*/*z* 483 (25) $[M + 1]^+$, 467 (10), 451 (100) $[M - OCH_3]^+$; HRFABMS (*m*-nitrobenzyl alcohol, NaCl) *m*/*z* $[M + Na]^+$ 505.2579 (calcd for $C_{29}H_{38}O_6Na$ 505.2566).

(17*S*,18*S*,20*R*,22*R*)-4β-Hydroxy-18,20-epoxy-18-methoxy-1-oxowitha-2,5,24-trienolide (4): white amorphous powder; $[\alpha]^{25}_D$ +54.9° (*c* 0.18, $CHCl_3$); UV (MeOH) λ_{max} 223 nm; IR (film) ν_{max} 3480, 2973, 2897, 1700, 1690 cm^{-1}; ^1H and ^{13}C NMR, Tables 1 and 2; EIMS *m*/*z* $[M]^+$ absent, 464 (4) $[M - H_2O]^+$, 450 (9) $[M - CH_3OH]^+$, 432 (23), 357 (30), 152 (69), 125 (100); FABMS (glycerol) *m*/*z* 483 (28) $[M + 1]^+$; HRFABMS (*m*-nitrobenzyl alcohol, NaCl) *m*/*z* $[M + Na]^+$ 505.2566 (calcd for $C_{29}H_{38}O_6Na$ 505.2566).

(17*S*,20*R*,22*R*)-4β-Hydroxy-18,20-epoxy-1,18-dioxowitha-2,5,24-trienolide (5): white amorphous powder (hexanes−EtOAc); mp 215−217 °C (dec); $[\alpha]^{25}_D$ +60.4° (*c* 0.15, $CHCl_3$); UV (MeOH) λ_{max} 226, 312 nm; IR (film) ν_{max} 3456, 1748, 1688, 1655, 1230, 1035 cm^{-1}; ^1H and ^{13}C NMR, Tables 1 and 2; EIMS *m*/*z* 466 (10) $[M]^+$, 448 (3), 152 (10), 151 (10), 125 (35), 109 (34), 97 (18), 43 (100); FABMS (glycerol, KCl) *m*/*z* 505 (100) $[M + K]^+$, 467 (8) $[M + 1]^+$; HRFABMS (*m*-nitrobenzyl alcohol, NaCl) *m*/*z* $[M + Na]^+$ 489.2274 (calcd for $C_{28}H_{34}O_6Na$ 489.2253).

(17*S*,18*R*,20*R*,22*R*)-18,20-Epoxy-18-methoxy-1,4-dioxowitha-2,5,24-trienolide (6): $[\alpha]^{25}_D$ +41.1° (*c* 0.25, $CHCl_3$); UV (MeOH) λ_{max} 224, 284, 362 nm; IR film (AgCl) ν_{max} 3545, 1700, 1676, 1668, 1620 cm^{-1}; ^1H and ^{13}C NMR, Tables 1 and 2; EIMS *m*/*z* $[M]^+$ absent, 465 (1) $[M - CH_3]^+$, 449 (5) $[M - OCH_3]^+$, 355 (52), 307 (15), 187 (7), 171 (7), 125 (36); FABMS (glycerol, KCl) *m*/*z* 481 (9) $[M + 1]^+$, 465 (6), 449 (100) $[M - OCH_3]^+$; HRFABMS (*m*-nitrobenzyl alcohol, NaCl) *m*/*z* $[M + Na]^+$ 503.2440 (calcd for $C_{29}H_{36}O_6Na$ 503.2410).

(17*S*,18*S*,20*R*,22*R*)-18,20-Epoxy-18-methoxy-1,4-dioxowitha-2,5,24-trienolide (7): $[\alpha]^{25}_D$ +38.9° (*c* 0.04, $CHCl_3$); UV (MeOH) λ_{max} 226, 280, 362 nm; IR film (AgCl) ν_{max} 3550, 1700, 1680, 1665, 1620 cm^{-1}; ^1H and ^{13}C NMR, Tables 1 and 2; EIMS *m*/*z* $[M]^+$ absent, 355 (4), 237 (3), 171 (4), 125 (28); FABMS (glycerol, KCl) *m*/*z* 481 (6) $[M + 1]^+$, 449 (100) $[M - OCH_3]^+$; HRFABMS (*m*-nitrobenzyl alcohol, NaCl) *m*/*z* $[M + Na]^+$ 503.2447 (calcd for $C_{29}H_{36}O_6Na$ 503.2410).

(17*S*,20*R*,22*R*)-5β,6β-Epoxy-4β,18,20-trihydroxy-1-oxowitha-2,24-dienolide (8): white crystals (hexanes−EtOAc); mp 175−177 °C (dec); $[\alpha]^{25}_D$ +59.8° (*c* 0.26, MeOH); UV (MeOH) λ_{max} 223 nm; IR (film) ν_{max} 3385, 2930, 2880, 1690 cm^{-1}; ^1H and ^{13}C NMR, Tables 1 and 2; EIMS *m*/*z* $[M]^+$ absent, 361 (1.5), 343 (9), 325 (4), 169 (11), 152 (3), 125 (30), 55 (29); FABMS (*m*-nitrobenzyl alcohol, K_2CO_3) *m*/*z* 525 (100) $[M + K]^+$, 467 (44) $[M - H_2O + 1]^+$; HRFABMS (*m*-nitrobenzyl alcohol, NaCl) *m*/*z* $[M + Na]^+$ 509.2538 (calcd for $C_{28}H_{38}O_7Na$ 509.2515).

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Supporting Information Available: AM1 calculated structures and NOE data of **3** and **4**. This material is available free of charge via the Internet at http://pubs.acs.org.

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