New Oplopane and Eremophilane Derivatives from *Robinsonecio gerberifolius*[§]

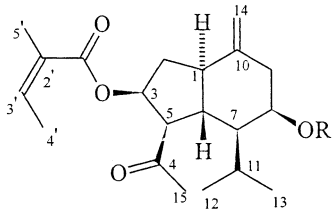
Amira Arciniegas,[†] Ana-L. Pérez-Castorena,[†] Sibet Reyes,[†] José Luis Contreras,[‡] and Alfonso Romo de Vivar*[,†]

Instituto de Química, Universidad Nacional Autónoma de México, Circuito Exterior, Ciudad Universitaria, Coyoacán 04510, D.F., México, and Herbario y Jardín Botánico, Benemérita Universidad Autónoma de Puebla, Ciudad Universitaria, 18 Sur y Av. San Claudio, San Manuel 72590, Puebla, México

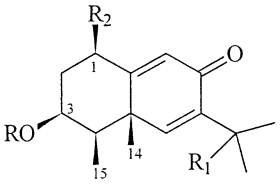
Received August 30, 2002

A phytochemical study of *Robinsonecio gerberifolius* afforded six new sesquiterpenoids, two oplopane (**1** and **2**) and four eremophilane derivatives (**3**−**6**). The structures of these compounds were elucidated on spectroscopic grounds, and the absolute configurations of compounds **3** and **4** were established from CD analysis. The known 3β-angeloyloxy-1,10-epoxyfuranoeremophilane (**7**) was also isolated, and its stereochemistry was revised. The cytotoxic activities of compounds **1**−**7** were determined against five human cancer cell lines.

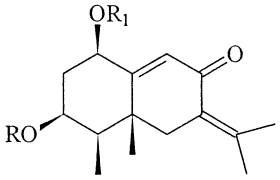
Robinsonecio (Asteraceae, Senecioneae, Tussilagininae) is a small genus that consists of only two species endemic to the high mountains of Mexico and Guatemala. Due to the fact that *Robinsonecio* was recently segregated from the genus *Senecio* on taxonomic grounds,[1] we became interested in its chemical composition. The most characteristic secondary metabolites of the genus *Senecio* are pyrrolizidine alkaloids and sesquiterpenes of the eremophilane type.[2] Oplopanes have also been isolated from *Senecio*,[3] although they are found in other genera of the Asteraceae[4] and even in other families.[5] As a result of a chemical study of *Robinsonecio gerberifolius*, we describe herein the isolation, structure elucidation, and cytotoxicity of two new oplopanes (**1** and **2**) and four new eremophilanes (**3**−**6**). Four common known compounds and the furanoeremophilane **7**, whose stereochemistry was revised, were also found.



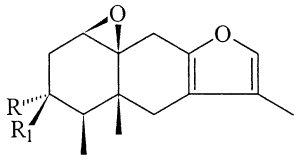
1 R = Epang
2 R = Ac



3 R = Ang $R_1 = R_2 = OH$
4 R = Ang $R_1 = OOH$ $R_2 = OH$
8 R = H $R_1 = R_2 = OH$
9 R = H $R_1 = OH$ $R_2 = H$



5 R = Ang $R_1 = H$
6 $R = R_1 =$ Ang
10 $R = R_1 = H$



7 R = H R_1 = OAng
7a R = OAng $R_1 = H$

Results and Discussion

Compound **1** exhibited a molecular formula of $C_{25}H_{36}O_6$ (HRFABMS *m/z* 433.2594 $[M + 1]^+$) indicative of eight degrees of unsaturation. It exhibited IR bands at 1736, 1715, and 1656 cm^{-1} due to carbonyl groups and double bonds. Its ^1H NMR spectrum (Table 1) showed signals that corresponded to an exocyclic double bond (δ 4.69 and 4.63), a methyl ketone (δ 2.17 s), and an isopropyl moiety (δ 0.97 d, 0.79 d, and 1.60 m), suggesting an oplopene skeleton.[3] Signals at δ 5.56 and 5.38 were assigned to hydrogen atoms attached to C-3 and C-8, respectively. These carbons also supported ester functions which were identified as angeloyloxy and epoxyangeloyloxy groups by their characteristic signals observed in the ^1H and ^{13}C NMR spectra (Tables 1 and 2). The epoxyangeloyloxy substituent was placed at C-8 because its carbonyl correlated with H-8 and with its α-methyl group in a FLOCK experiment.[6] Accordingly, the angeloyloxy group could be attached to C-3 as shown in structure **1**. This structure was confirmed and its relative stereochemistry established by X-ray crystallographic analysis (Figure 1).

Compound **2** exhibited a molecular formula of $C_{22}H_{32}O_5$, established from HRFABMS (*m/z* 377.2321 $[M + 1]^+$). Its ^1H and ^{13}C NMR spectra (Tables 1, 2) were similar to those of **1** except for the presence of an acetoxy group attached to C-8 according to a FLOCK experiment. NOE effects of

[§] Contribution No. 1758 of the Instituto de Química, UNAM.

* To whom correspondence should be addressed. Tel: (5255) 56-224412. Fax: (5255) 56-162217. E-mail: alrovi@correo.unam.mx.
[†] Instituto de Química, UNAM.
[‡] Herbario y Jardín Botánico, Benemérita Universidad Autónoma de Puebla.

10.1021/np0203739 CCC: $25.00
Published on Web 01/17/2003

Table 1. ^{1}H NMR Spectral Data of Compounds **1**−**6** (300 MHz, $CDCl_3$)[a]

position	**1**[b]	**2**[c]	**3**	**4**[d]	**5**	**6**[e]
1	under 4′	under 4′	4.58 br t (3.0)	4.53 dd (3.6, 1.8)	4.37 br t (3.6)	5.59 dd (4.4, 2.2)
2a	2.47 ddd (12.0, 7.2, 2.1)	2.46 ddd (12.0, 7.2, 4.8)	2.50 dt (15.9, 2.4)	2.49 dt (15.3, 2.4)	2.42 dt (15.6, 3.0)	2.57 dt (16.2, 2.4)
2b	dd (12.2, 7.8)	1.76 br dd (12.6, 8.2)	1.93 dt (15.9, 3.9)	1.90 dt (15.3, 4.2)	1.98 dt (15.6, 4.2)	2.05 dt (16.0, 4.4)
3	5.56 ddd (9.6, 7.8, 7.2)	5.57 ddd (9.9, 7.9, 7.3)	5.27 dt (3.9, 3.6)	5.28 br dd (4.2, 3.6)	5.3 br dd (4.2, 3.3)	5.11 br dd (3.6, 3.3)
4			1.84 qd (7.2, 3.6)	1.84 qd (7.2, 3.6)	1.84 qd (7.2, 3.6)	1.83 qd (6.9, 3.5)
5	3.20 t (10.2)	3.15 t (9.9)				
6a	2.75 q (11.4)	2.68 q (11.1)	6.93 s	7.30 s	2.95 d (13.5)	2.97 d (13.2)
6b					2.19 d (13.5)	2.17 d (13.2)
7	1.42 ddd (10.1, 4.5, 1.8)	1.38 ddd (11.3, 4.1, 2.0)				
8	br dd (4.5, 2.7)	5.28 br dd (4.7, 2.0)				
9a	2.55 dd (12.0, 3.0)	dd (15.2, 3.3)	6.22 s	6.14 s	5.94 s	6.06 s
9b	2.22 dd (12.0, 1.5)	2.18 dd (15.2, 2.0)				
11	1.60 m	1.52 dhept (7.0, 4.1)				
12	0.97[f] d (6.6)	0.96[f] d (6.6)	1.47[f] s	1.53[f] s	2.11 s	2.14 s
13	0.79[f] d (6.9)	0.73[f] d (7.2)	1.46[f] s	1.49[f] s	1.87 s	1.87 s
14a	4.69 d (1.8)	4.68 br d (3.0)	1.57 s	1.60 s	1.36 s	1.34 s
14b	4.63 d (1.8)					
15	2.17 s	2.19 s	1.23 d (6.9)	1.26 d (7.2)	1.10 d (7.2)	1.13 d (6.9)
3′	6.11 qq (7.2, 1.5)	6.09 qq (7.2, 1.5)	6.14 qq (7.2, 1.5)	6.14 qq (7.2, 1.5)	6.11 qq (7.5, 1.5)	6.03 qq (7.5, 1.38)
4′	1.98 dq (7.2, 1.5)	1.98 dq (7.2, 1.5)	2.05 dq (7.2, 1.5)	2.05 dd (1.2, 1.2)	2.04 dq (7.2, 1.5)	1.97 dq (7.6, 1.38)
5′	1.84 quint (1.5)	1.84 br s	1.96 quint (1.5)	1.96 quint (1.5)	1.98 br s	1.89 quint (1.38)

[a] Assigments are based on COSY, long-range HETCOR, and FLOCK experiments. [b] Epang signals: δ 3.03 (q, J = 5.4 Hz, H-3″), 1.38 (d, J = 5.4 Hz, H-4″), 1.55 (s, H-5″). [c] Ac signal at δ 2.08 s. [d] δ 10.0 (OOH). [e] Ang signals: δ 6.01 (qq, J = 7.2, 1.38 Hz, H-3″), 1.92 (dq, J = 7.2, 1.38 Hz, H-4″), 1.78 (quint, J = 1.38 Hz, H-5″). [f] Exchangeable signals.

H-5 with H-3 and H-7 and between H-7 and H-8, observed in a NOESY experiment, suggested the same relative stereochemistry for compounds **1** and **2**.

Compound **3** showed a protonated molecular ion peak at m/z 349.2006 $[M + 1]^+$ in the HRFABMS, consistent with a molecular formula of $C_{20}H_{28}O_5$. The IR spectrum exhibited bands for hydroxyl and α,β-unsaturated carbonyl groups (3603, 3537, 1712, and 1661 cm^{-1}). The ^{1}H NMR spectrum (Table 1) showed three vinylic protons, two singlets at δ 6.93 and 6.22 assigned to H-6 and H-9, respectively, and a quartet of quartets at δ 6.14 corresponding to an angelic proton. The signals at δ 5.27 (dt, J = 6.6, 3.5 Hz) and 4.58 (br t, J = 3.0 Hz) were assigned by means of COSY and long-range HETCOR experiments to the equatorial protons H-3 and H-1, geminal to an ester function and to a hydroxyl group, respectively. The second hydroxyl group could be located at C-11 since its ^{13}C NMR signal appeared as a singlet at δ 71.4. The ^{1}H NMR spectrum showed at high field, in addition to the typical signals of the angeloyloxy methyl groups, the four methyl signals of an eremophilane skeleton at δ 1.47 s, 1.46 s, 1.55 s, and 1.23 d (J = 6.9 Hz) assigned to C-12, C-13, C-14, and C-15, respectively. The relative stereochemistry depicted in **3** was suggested by the NOE effects of H-3 with H-4, and that of CH_3-14 with CH_3-15 observed in the NOESY spectrum.

Compound **4** exhibited a molecular formula of $C_{20}H_{28}O_6$ determined from the HRFABMS (m/z 365.1957 $[M + 1]^+$). Its ^{1}H NMR spectrum differed from that of **3** (Table 1) only in the presence of a broad signal at δ 10.2 and an observed paramagnetic shift of the H-6 signal ($\Delta\delta$ 0.37). The ^{13}C NMR spectrum (Table 2) displayed the C-11 and C-6 signals with downfield shits ($\Delta\delta$ 11.1 and 2.4, respectively) and those of C-7, C-12, and C-13 with upfield shifts ($\Delta\delta$ 2.4, 3.9, and 4.4, respectively) with respect to the same signals of **3**. On the basis of the previous data a hydroperoxy group attached to C-11 of structure **4** was proposed and confirmed by X-ray crystallographic analysis (Figure 2).

Alkaline hydrolysis of **3** and **4** produced the same triol (**8**), whose CD curve showed a negative Cotton effect, similar to that observed in petasitol (**9**), whose absolute stereochemistry has already been determined.[7] Therefore, compounds **3** and **4** should have the CH_3-14 and CH_3-15 β-oriented as in petasitol, with the absolute configurations 1*S*, 3*S*, 4*R*, 5*S*.

Compound **5**, with a molecular formula of $C_{20}H_{28}O_4$ (HRFABMS m/z 333.2072 $[M + 1]^+$), exhibited IR bands for hydroxyl and conjugated carbonyl groups (3593, 1713, and 1661 cm^{-1}). Its ^{1}H and ^{13}C NMR spectra (Tables 1 and 2) indicated the same substitution pattern in ring A as in compound **4**. Compound **5** differed from the latter in the

Table 2. ^{13}C NMR Spectral Data of Compounds **1**−**6** (75 MHz, $CDCl_3$)[a]

carbon	**1**[b]	**2**[c]	**3**	**4**	**5**	**6**[d]
1	47.9 d	47.9 d	72.4 d	72.3 d	71.7 d	71.1 d
2	34.9 t	34.9 t	37.8 t	37.9 t	36.1 t	34.0 t
3	73.8 d	73.6 d	71.6 d	72.0 d	72.1 d	71.4 d
4	205.5 s	205.7 s	42.3 d	42.6 d	43.7 d	43.3 d
5	57.9 d	58.3 d	42.0 s	42.1 s	39.8 s	40.0 s
6	45.6 d	45.9 d	149.6 d	152.0 d	43.1 t	42.6 t
7	51.7 d	51.6 d	140.3 s	137.9 s	127.5 s	127.5 s
8	72.3 d	70.5 d	187.6 s	185.8 s	191.6 s	191.6 s
9	40.5 t	40.5 t	127.0 d	127.4 d	129.5 d	131.7 d
10	144.3 s	144.5 s	164.0 s	162.7 s	163.5 s	159.1 s
11	29.3 d	29.5 d	71.4 s	82.5 s	145.3 s	145.4 s
12	22.7 q[e]	22.4 q[e]	28.6 q[e]	24.7 q[e]	22.5 q[e]	22.5 q[e]
13	19.1 q[e]	18.1 q[e]	28.5 q[e]	24.1 q[e]	27.8 q[e]	22.8 q[e]
14	107.9 t	107.4 t	20.6 q	20.8 q	20.3 q	19.2 q
15	31.7 q	31.8 c	12.2 q	12.4 q	11.8 q	11.8 q
1′	167.3 s	167.3 s	166.7 s	167.1 s	167.0 s	167.5 s
2′	127.2 s	127.2 s	126.9 s	127.2 s	127.2 s	127.3 s
3′	139.5 d	139.3 d	139.6 d	138.2 d	139.6 d	139.1 d
4′	15.7 q	15.7 q	15.6 q	15.6 q	15.7 q	15.6 q
5′	20.3 q	20.3 q	20.8 q	20.8 q	20.8 q	20.6 q

[a] Assigments are based on DEPT, HETCOR, long-range HETCOR, and FLOCK experiments. [b] Epang signals: δ 168.9 (s, C-1″), 59.8 (s, C-2″), 59.9 (d, C-3″), 13.7 (q, C-4″), 19.1[e] (q, C-5″). [c] Ac signals at δ 21.6 q and 170.5 s. [d] Ang signals: δ 166.5 (s, C-1″), 127.9 (s, C-2″), 138.4 (d, C-3″), 15.6 (q, C-4″), 20.3 (q, C-5″). [e] Exchangeable signals.

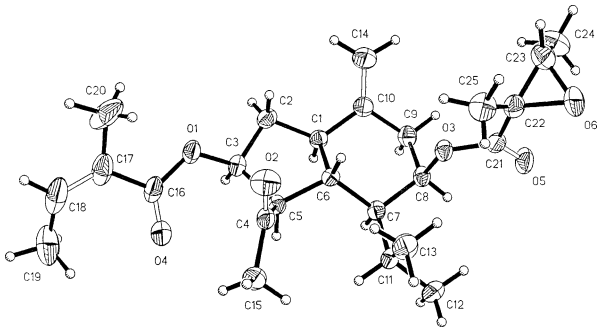


Figure 1. ORTEP projection of **1** (crystallographic numbering).

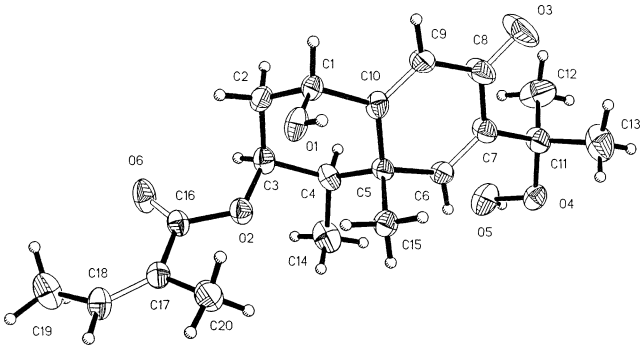


Figure 2. ORTEP projection of **4** (crystallographic numbering).

presence of a 7(11) double bond. This was deduced from the paramagnetic shifts of the C-11 methyl signals in the ^1H NMR spectrum (Table 1) and the C-6, C-7, and C-11 chemical shifts observed in the ^{13}C NMR spectrum (Table 2). Compound **5** was assigned with the same stereochemistry as **4** since a NOESY experiment showed interactions between H-1 and H-9 and between H-3 and H-4.

Compound **6**, with a molecular formula $C_{25}H_{34}O_5$ (HRFABMS *m*/*z* 415.2481, $[M + 1]^+$), was the angelate of compound **5**. Hydrolysis of compounds **5** and **6** resulted in the same derivative **10**, thus confirming the stereochemistry of both compounds.

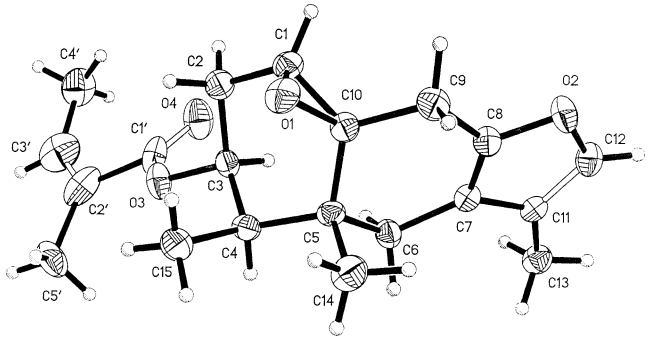


Figure 3. ORTEP projection of **7** (crystallographic numbering).

Compound **7** showed the same IR, MS, and ^1H NMR spectral data as those reported by Bohlmann and Zdero[8] for **7a**. However, the coupling constants of H-3 (ddd, J = 11.7, 7.2, 4.2 Hz) were in agreement with the β-orientation of the ester group, as in similar compounds.[9] X-ray crystallographic analysis of this compound (Figure 3) provided unequivocal evidence that its structure should be depicted as **7** instead of **7a**.

The co-occurrence of eremophilane and oplopane sesquiterpenoids makes the chemistry of *R. gerberifolius* rather unusual. The presence of eremophilanes containing the 1-hydroxy-6,9-dien-8-one system seems to be another distinctive feature of this species. However, it is necessary to investigate the chemical composition of *R. porphyresthes*, the other species of the genus, to reach a definitive conclusion about the chemistry of the newly established genus.

Compounds **1**−**7** were tested against colon (HCT-15), breast (MCF-7), central nervous system (U-251), prostate (PC-3), and leukemia (K562) human cancer cells (Table 3) following protocols established by the National Cancer Institute (Bethesda, Maryland).[10] Of the oplopane derivatives tested, compound **1** showed selective cytotoxicity against PC-3 cells, while compound **2** was nearly active against all of the tested cell lines. Among the eremophilanes, compound **3** was not active, compounds **4** and **5** were selective to U-251 and PC-3 cells, and compound **6** was cytotoxic for all the cell lines tested. The furanoeremophilane **7** did not show any activity at the dose range tested.

Experimental Section

General Experimental Procedures. Melting points were determined on a Fisher-Jones melting point apparatus and are uncorrected. Optical rotations were determined on a JASCO DIP-360 digital polarimeter. IR spectra were recorded on a Nicolet Magna-IR 750 spectrometer. ^1H NMR and ^{13}C NMR data were obtained on a Varian Unity 300 instrument. Chemical shifts were referred to TMS (δ 0). Standard Varian programs were used for COSY and NOESY spectra at 300 MHz. HETCOR experiments were obtained for $^1J_{CH} = 140$ Hz at 75 MHz. Long-range HETCOR, COLOC, and FLOCK experiments were obtained for $^nJ_{CH} = 9$ Hz at 75 MHz. EIMS data were determined on a JEOL JMS-AX505HA mass spectrometer at 70 eV. FABMS were obtained on a JEOL JMS-SX102A mass spectrometer operated with an acceleration voltage of 10 kV, and samples were desorbed from a nitrobenzyl alcohol matrix using 6 kV xenon atoms. High-resolution MS measurements in the FAB mode were performed at 10 000 resolution using electric field scans and poly(ethylene glycol) ions (Fluka 200 and 300) as the reference material. Column chromatography was carried out on Kieselgel G (Merck, Darmstadt, Germany). TLC was performed on Si gel 60 and preparative TLC on Si gel GF_{254} (Merck), layer thickness 2.0 mm.

Plant Material. *Robinsonecio gerberifolius* (Sch. Bip. in Hemsley) T. M. Barkley & J. P. Javonec was collected at Pico de Orizaba, Veracruz, Mexico, in October 2000. A voucher

Table 3. Cytotoxicity Data for Compounds **1**−**6** (IC_{50} μM)[a]

compound	HCT-15	MCF-7	U251	PC-3	K562
1	>100	>100	>100	13.6 ± 4.2	>100
2	43.6 ± 3.3	33.9 ± 11.6	13.0 ± 0.95	78.7 ± 13.0	13.0 ± 1.2
4	>100	>100	33.7 ± 3.4	16.5 ± 5.6	46.3 ± 3.3
5	>100	>100	24.1 ± 1.5	33.2 ± 1.5	> 100
6	12.8 ± 0.65	21.0 ± 2.0	17.6 ± 1.88	10.7 ± 0.65	51.1 ± 8.6
doxorubicin	0.23 ± 0.01	0.14 ± 0.01	0.09 ± 0.02	0.32 ± 0.02	0.28 ± 0.01

[a] Compounds **3** and **7** were not active.

specimen (HUAP 10365) is deposited at the Herbario y Jardín Botánico de la Benemérita Universidad Autónoma de Puebla.

Extraction and Isolation. The dried and ground leaves (764 g) of *R. gerberifolius* were extracted exhaustively with MeOH. The solvent was eliminated under reduced pressure to obtain 93.48 g of extract. The same procedure was applied to the rhizomes (367 g) and roots (130 g) to afford 57.6 and 23 g of extract, respectively. The extracts gave negative Dragendorff tests.

The leaf extract was submitted to vacuum-column chromatography (VCC) with a gradient of hexane−EtOAc (500 mL fractions) as follows: hexane (fr. 1−12), hexane−EtOAc, 49:1 (fr. 21−30), hexane−EtOAc, 19:1 (fr. 31−43), hexane−EtOAc, 9:1 (fr. 44−100), hexane−EtOAc, 4:1 (fr. 100−200), hexane−EtOAc, 7:3 (fr. 200−216), hexane−EtOAc, 1:1 (fr. 217−130), hexane−EtOAc, 3:7 (fr. 230−240), EtOAc (fr. 240−250), and MeOH (fr. 250−260). Fractions 8−46 were combined and purified by VCC with hexane−EtOAc (9:1) to give **2** (3.44 g). Fractions 51−61 afforded after recrystallization with EtOAc 148.7 mg of **1**. Fractions 62−84, submitted to VCC with hexane−EtOAc (9:1), yielded *p*-hydroxyacetophenone (269 mg) and a fraction A. Purification of A by flash column chromatography using Si gel 230−400 μm and hexane−acetone (99:1) as eluent afforded **6** (97.8 mg). Further purification of fractions 85−104 by VCC using hexane−EtOAc (17:3) produced **4** (1.85 g) and sitosterol−stigmasterol as a mixture (218 mg). Fractions eluted with hexane−EtOAc (3:7) produced β-sitosterol glucoside (1.51 g).

The root extract, purified using the same procedure described above, produced **1** (1.56 g) from fractions eluted with hexane−EtOAc (9:1). Fractions eluted with hexane were submitted to further purification by VCC using hexane as eluent to afford **7** (949 mg). Fractions eluted with MeOH afforded sucrose (416 mg).

The rhizome extract was purified by VCC with a hexane−acetone gradient (250 mL fractions). Fractions eluted with hexane−acetone (24:1) afforded **1** (1.85 g). Fractions eluted with hexane−acetone (17:3) were combined and purified by VCC using hexane−acetone (9:1) as eluent to yield **3** (133.2 mg) and fraction B, which produced **5** (898 mg) by VCC using hexane−acetone (9:1) as eluent.

p-Hydroxyacetophenone was identified by comparison of its physical constants and spectral data with those described in the literature.[11] β-Sitosterol glucoside, sitosterol, stigmasterol, and sucrose were identified by direct comparison with authentic samples.

3-Angeloyloxy-8-epoxyangeloyloxy-10(14)-oplopen-4-one (1): white crystals from EtOAc; mp 123−5 °C; $[\alpha]_D$ −109.5° (*c* 0.20, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 219 (4.01) nm; IR ($CHCl_3$) ν_{max} 2946, 1736, 1715, 1656 cm^{-1}; ^1H NMR data, see Table 1; ^{13}C NMR data, see Table 2; EIMS *m/z* 432 $[M]^+$ (3), 316 (8), 217 (100), 173 (27), 83 (65); HRFABMS *m/z* 433.2594 $[M + 1]^+$ ($C_{25}H_{37}O_6$ requires 433.2590).

3-Angeloyloxy-8-acetoxy-10(14)-oplopen-4-one (2): colorless oil; $[\alpha]_D$ 55° (*c* 0.20, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 219 (3.79) nm; IR ($CHCl_3$) ν_{max} 2962, 1717, 1656 cm^{-1}; ^1H NMR data, see Table 1; ^{13}C NMR data, see Table 2; EIMS *m/z* 376 $[M]^+$ (5), 316 (12), 217 (55), 173 (53), 131 (35), 83 (100), 55 (40), 43 (55); HRFABMS *m/z* 377.2321 $[M + 1]^+$ ($C_{22}H_{33}O_5$ requires 377.2328).

3-Angeloyloxy-1,11-dihydroxyeremophila-6,9-dien-8-one (3): colorless needles from EtOAc; mp 165−7 °C; $[\alpha]_D$ −23° (*c* 0.20, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 242 (4.08) nm; IR ($CHCl_3$) ν_{max} 3604, 3484, 1712, 1661, 1619 cm^{-1}; ^1H NMR data, see Table 1; ^{13}C NMR data, see Table 2; FABMS *m/z* 349 $[M + 1]^+$ (60), 331 (15), 307 (25), 154 (100), 136 (75); HRFABMS *m/z* 349.2006 $[M + 1]^+$ ($C_{20}H_{29}O_5$ requires 349.2015).

1-Hydroxy-3-angeloyloxy-11-hidroperoxyeremophila-6,9-dien-8-one (4): white crystals from EtOAc; mp 178−80 °C; $[\alpha]_D$ +19° (*c* 0.20, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 241 (4.44) nm; IR ($CHCl_3$) ν_{max} 3602, 3537, 1711, 1663, 1627 cm^{-1}; ^1H NMR data, see Table 1; ^{13}C NMR data, see Table 2; FABMS *m/z* 365 $[M + 1]^+$ (100), 331 (20), 231 (40), 213 (35), 154 (60), 136 (45), 83 (55); HRFABMS *m/z* 365.1957 $[M + 1]^+$ ($C_{20}H_{39}O_6$ requires 365.1964).

1-Hydroxy-3-angeloyloxyeremophila-9,7(11)-dien-8-one (5): colorless oil; $[\alpha]_D$ +90° (*c* 0.20, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 281 (3.76), 274 (3.75), 209 (4.19) nm; IR ($CHCl_3$) ν_{max} 3593, 2980, 1713, 1661, 1613 cm^{-1}; ^1H NMR data, see Table 1; ^{13}C NMR data, see Table 2; EIMS *m/z* 332 $[M]^+$ (90), 232 (65), 217 (35), 199 (50), 175 (55),149 (95), 97 (55), 83 (100), 55 (75); HRFABMS *m/z* 333.2072 $[M + 1]^+$ ($C_{20}H_{29}O_4$ requires 333.2066).

1,3-Diangeloyloxyeremophila-9,7(11)-dien-8-one (6): colorless oil; $[\alpha]_D$ +174.5° (*c* 0.20, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 285 (3.84), 273 (3.82), 215 (4.48) nm; IR ($CHCl_3$) ν_{max} 2969, 1712, 1662, 1614 cm^{-1}; ^1H NMR data, see Table 1; ^{13}C NMR data, see Table 2; EIMS *m/z* 432 $[M]^+$ (3), 414 (35), 314 (20), 231 (15), 83 (100), 55 (35); HRFABMS *m/z* 415.2481 $[M + 1]^+$ ($C_{25}H_{35}O_5$ requires 415.2484).

3β-Angeloyloxy-1,10-epoxyfuranoeremophilane (7):[8] white crystals from hexane; mp 148−149 °C; $[\alpha]_D$ −12.5° (*c* 0.2, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 218.6 (4.39) nm.

Alkaline Hydrolysis of Compounds 3−6. Compounds **3**−**6** (0.2 mmol), in 10 mL of 2 M Na_2CO_3 methanolic solution, were each stirred at room temperature for 72 h. The solvent was evaporated, and the residue was extracted with EtOAc, purified by VCC using hexane−acetone (9:1) as eluent, and subsequently purified by preparative TLC with hexane−acetone (3:1) as eluent. Compounds **3** and **4** yielded **8** (13 and 15 mg, respectively), and compounds **5** and **6** produced **10** (12 and 14 mg, respectively).

1,3,11-Trihydroxyeremophila-6,9-dien-8-one (8): white crystals from hexane−EtOAc; mp 78−80 °C; $[\alpha]_D$ −20.8° (*c* 0.12, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 246 (4.01) nm; IR ($CHCl_3$) ν_{max} 3692, 3610, 3517, 161, 1660 cm^{-1}; CD $[\theta]_{209}$ + 8600.8, $[\theta]_{243}$ − 18707.3, $[\theta]_{278}$ + 499.8; ^1H NMR ($CDCl_3$, 300 MHz) δ 4.64 (1H, t, J = 2.9 Hz, H-1), 2.54 (1H, dt, J = 14.8, 2.8 Hz, H-2a), 1.79 (1H, dt, J = 14.8, 3.3 Hz, H-2b), 3.99 (1H, br d, J = 3.0 Hz, H-3), 1.57 (1H, dq, J = 7.0, 2.9 Hz, H-4), 6.93 (1H, s, H-6), 6.18 (1H, s, H-9), 1.46, 1.47 (3H each, s, H-12, H-13), 1.51 (3H, s, H-14), 1.36 (3H, d, J = 7.0 Hz, H-15); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 72.5 (d, C-1), 39.8 (t, C-2), 74.6 (d, C-3), 43.3 (d, C-4), 42.7 (s, C-5), 150.3 (d, C-6), 139.9 (s, C-7), 188.2 (s, C-8), 126.7 (d, C-9), 165.2 (s, C-10), 71.9 (s, C-11), 28.8 (q, C-12 or C-13), 28.9 (q, C-12 or C-13), 22.0 (q, C-14), 12.7 (q, C-15); FABMS *m/z* 267 $[M + 1]^+$ (35), 249 (15), 231 (18), 213 (10).

1,3-Dihydroxyeremophila-9,7(11)-dien-8-one (10): colorless oil; $[\alpha]_D$ +29.6° (*c* 0.25, $CHCl_3$); UV (MeOH) λ_{max} (log ϵ) 244 (3.84), 207 (3.85) nm; IR ($CHCl_3$) ν_{max} 3684, 3607, 1661, 1604 cm^{-1}; ^1H NMR ($CDCl_3$, 300 MHz) δ 4.44 (1H, dq, J = 7.0, 2.7 Hz, H-1), 2.44 (1H, dt, J = 14.9, 2.8 Hz, H-2a), 1.83 (1H, dt, J = 15, 3.3 Hz, H-2b), 3.97 (1H, br d, J = 2.1 Hz, H-3), 1.56 (1H, dq, J = 7.0, 2.7 Hz, H-4), 2.95 (1H, d, J = 13.9 Hz, H-6a), H-6b under H-12, 5.87 (1H, s, H-9), 2.13 (3H, s, H-12), 1.87 (3H, s, H-13), 1.32 (3H, s, H-14); 1.20 (3H, d, J = 7.0 Hz, H-15); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 72.4 (d, C-1), 37.7 (t, C-2), 73.8 (d, C-3), 43.8 (d, C-4), 39.9 (s, C-5), 45.4 (t, C-6),

127.6 (s, C-7), 192.2 (s, C-8), 129.2 (d, C-9), 164.5 (s, C-10), 154.8 (s, C-11), 22.9 (q, C-12 or C-13), 22.7 (q, C-12 or C-13), 21.3 (q, C-14), 12.5 (q, C-15); EIMS *m*/*z* 250 $[M]^+$ (100), 248 (10), 230 (10), 281 (25).

X-ray Diffraction Structure Determination for Compound 1.[12] Crystal data: $C_{25}H_{36}O_6$; crystal size (mm) 0.62 × 0.36 × 0.28 colorless prism; crystal system orthorhombic; space group $P2_12_12_1$; unit cell dimensions a = 10.872(1) Å, b = 14.883(1) Å, c = 15.121(1); volume 2446.7(3) Å^3; Z = 4; formula weight 432.54; density (calcd) 1.174 Mg/m^3; absorption coefficient 0.083 mm^{-1}; F(000) 936. The reflection data were collected on a Siemens P4, using graphite-monochromated radiation Mo Kα (λ = 0.71073 Å). A total of 4882 reflections were collected in the range 1.50° ≤ θ ≤ 25.00°, of which 4299 were unique reflections with $I > 2\sigma(I)$, and were used for refinement. The final R and R_w were 0.0687 and 0.1512, respectively. The structure was solved by the direct methods using the program SIR97. No hydrogen atoms were refined with anisotropic displacement parameters. Hydrogen atoms were included at calculated positions, except for those bonded to oxygen atoms, and were not refined.

X-ray Diffraction Structure Determination for Compound 4.[12] Crystal data: $C_{20}H_{28}O_6$; crystal size (mm) 0.60 × 0.24 × 0.20 colorless prism; crystal system orthorhombic; space group $P2_12_12_1$; unit cell dimensions a = 7.124(1) Å, b = 14.218(1) Å, c = 19.643(1) Å; volume 1989.6(3) Å^3; Z = 4; formula weight 364.42; density (calcd) 1.217 Mg/m^3; absorption coefficient 0.089 mm^{-1}; F(000) 784. The reflection data were collected on a Siemens P4, using graphite-monochromated radiation Mo Kα (λ = 0.71073 Å). A total of 2029 reflections were collected in the range 1.50° ≤ θ ≤ 25.00°, of which 2028 were unique reflections with $I > 2\sigma(I)$, and were used for refinement. The final R and R_w were 0.0592 and 0.1190, respectively. The structure was solved as described for compound **1**.

X-ray Diffraction Structure Determination for Compound 7.[12] Crystal data: $C_{20}H_{26}O_4$; crystal size (mm) 0.22 × 0.20 × 0.10 colorless plates; crystal system orthorhombic; space group $P2_12_12_1$; unit cell dimensions a = 6.991(1) Å, b = 7.753(1) Å, c = 33.047(1) Å; volume 1791.4 (4) Å^3; Z = 4; formula weight 330.41; density (calcd) 1.225 Mg/m^3; absorption coefficient 0.084 mm^{-1}; F(000) 712. The reflection data were collected on a Bruker Smart Apex CCD diffractometer, using graphite-monochromated radiation Mo Ka (λ = 0.71073 Å). A total of 14 559 reflections were collected in the range 2.47° ≤ θ ≤ 24.99°, of which 3161 were unique reflections with $I > 2\sigma(I)$m and were used for refinement. The final R and R_w were 0.0575 and 0.0636, respectively. The structure was solved as described for compound **1**.

Acknowledgment. We are indebted to Rubén Alfredo Toscano, María Teresa Ramírez Apan, Alejandrina Acosta, Isabel Chávez, Nieves Zavala, Héctor Ríos, Angeles Peña, Elizabeth Huerta, Rocío Patiño, Miryam Adaya, Luis Velasco, Javier Pérez, and Gabriela Salcedo for technical assistance.

Supporting Information Available: X-ray crystallographic data of compounds **1**, **4**, and **7**. This material is available free of charge via the Internet at http//pubs.acs.org.

References and Notes

(1) Barkley, T. M.; Javonec J. P. *Sida* **1996**, *17*, 77−81.
(2) Robins, D. J. In *The Biology and Chemistry of the Compositae*; Heywood, J. H., Harborne, J. B., Turner, B. L., Eds.; Academic Press: London, 1977; Chapter 30, pp 831−850.
(3) Joseph-Nathan, P.; Villagómez, J. R.; Román, L. U.; Hernández, J. D. *Phytochemistry* **1990**, *29*, 977−979.
(4) Marco, A.; Sanz-Cervera, J. F.; Sancenon, F.; Jakupovic, J.; Rustaiyan, A.; Mohamadi, F. *Phytochemistry* **1993**, *34*, 1061−1065.
(5) Takeda, K.; Minato, H.; Ishikawa, M. *Tetrahedron* **1966**, Suppl. 7, 219−225.
(6) Reynolds, W. F.; McLean, S.; Perpick-Dumont, M.; Enriquez, R. *Magn. Reson. Chem*. **1989**, *21*, 162−169.
(7) Naya, K.; Takagi, I. *Tetrahedron Lett*. **1968**, 629−632.
(8) Bohlmann, F.; Zdero, C. *Phytochemistry* **1978**, *17*, 1135−1153.
(9) Wang, W.; Gao, K.; Jia, Z. *J. Nat. Prod*. **2002**, *65*, 714−717.
(10) Monks, A.; Scudiero, D.; Skehan, P.; Shoemaker, R.; Paul, K.; Vistica, D.; Hose, C.; Langley, J.; Cronise, P.; Vaigro-Wolff, A.; Gray-Goodrich, M.; Campbell, H.; Mayp, J.; Boyd, M. *J. Natl. Cancer Inst*. **1991**, *38*, 757−766.
(11) IR spectrum No. 28289, ^{1}H NMR spectrum No. 10590. *The Sadtler Standard Spectra*; Sadtler Research Laboratories, Inc.: Philadelphia, 1971.
(12) X-ray data for compounds **1**, **4**, and **7** have been deposited in the Cambridge Crystallographic Data Centre (CCDC 197755, 197756, and 197757, respectively). Copies of the data can be obtained free of charge on application to the Director, CCDC, 12 Union Rd., Cambridge CB2 1EZ, UK (fax: +44-(1223-336033; e-mail: deposit@ccdc.cam.ac.uk).

NP0203739