

TERPENOIDS FROM *VIGUIERA EXCELSA* AND
VIGUIERA OAXACANA

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Viguiera constitutes one of the largest genera in the subtribe Helianthinae of the Compositae and is exclusively located on the American continent (1). As part of a continuing chemical study of this genus which elaborates diterpenes (2-4), flavanol compounds (5), and sesquiterpene lactones (6), some of which display cytotoxic activity (7,8), we wish to describe the isolation and characterization of the terpenoids of *Viguiera excelsa* (Willd.) B. & H. and *Viguiera oaxacana* (Greenm.) Blake.

RESULTS AND DISCUSSION

Extensive chromatography of the CHCl_3 extract of *V. excelsa* furnished the cytotoxic lactone budlein A (**1**) as the major constituent. This compound was described earlier from *Helianthus* (9), *Calea* (10), and several other *Viguiera* species (6). Usual acetylation of budlein A (**1**) afforded the rearranged derivative **3**, previously obtained (11). The unrearranged ester (**2**) was prepared *in situ* on treatment with trichloroacetyl isocyanate (TAI) (see Experimental section). The not previously reported ^{13}C -nmr data of **2** and **3** are shown in Table 1 and agree with the proposed structures.

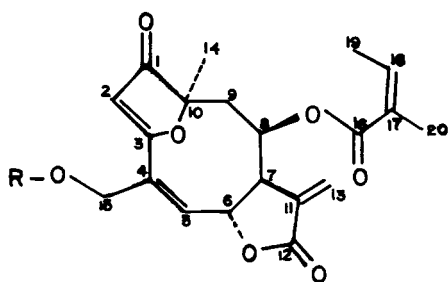
Minor constituents of *V. excelsa* were a mixture of acids, which was treated with CH_2N_2 and resolved by silica gel chromatography. The following compounds were obtained in the form of methyl esters: *ent*-kaur-9(11),16-dien-19-oic acid (**4**) (2), *ent*-kaur-16-en-19-oic acid (**12**) (2), *ent*-12-oxo-kaur-9(11),16-dien-19-oic acid (**6**) (13), 16 α , 17-dihydroxy-*ent*-kauran-19-oic

acid (**14**) (14,15), 12 β -hydroxy-*ent*-kaur-9(11),16-dien-19-oic acid (**8**) (13), and 12 β -ethoxy-*ent*-kaur-9(11),16-dien-19-oic acid (**10**), recently isolated from *Stevia eupatoria* (16). The identification of these compounds was carried out by standard methods (ir, ^1H -nmr, mp, mmp, ms), direct comparison with authentic samples and several derivatives described in the experimental.

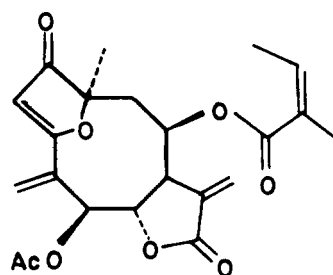
The most polar constituent isolated from this specimen was clovandiol (**18**), previously isolated from *Dipterocarpus pilosus* (17) and *Salvia canariensis* (18). This molecule was further characterized by its derivatives **19** and **20**. The not previously reported ^{13}C -nmr data of **18** are shown in Table 1.

The principal constituents of *V. oaxacana* were *ent*-kaur-9(11),16-dien-19-oic acid (**4**) and *ent*-kaur-16-en-19-oic acid **12**, which were characterized by direct comparison with authentic samples. Stigmasterol was also isolated from this specimen.

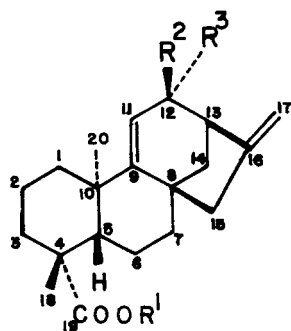
In spite of the limited chemical studies of *Viguiera* species, it has been found that the distribution of the secondary metabolites is in agreement with the proposed phylogenetic subdivision of the genus (1). *V. excelsa*, which belongs to the subgenus Amphilepsis, contains the 3(2H)-furanone heliangolide budlein A (**1**), like other members of this group (*Viguiera buddleiaeformis*, *Viguiera angustifolia*, *Viguiera bemsleyana*, *Viguiera hypochlora* and *Viguiera scultzii*), as previously suggested (6). On the other hand, *V. oaxacana* (subgenus Calantictaria, section Chloracra, series



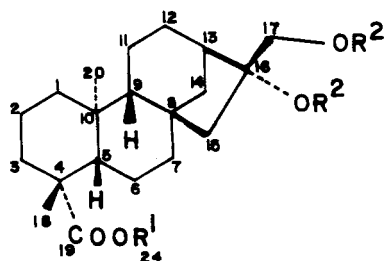
- 1 R=H
2 R=CONHCOCCL₃



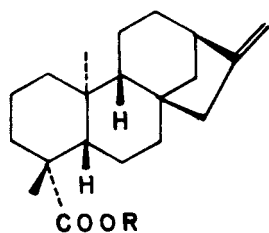
3



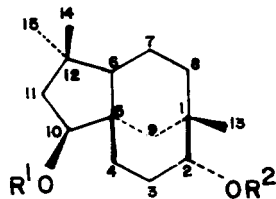
- 4 R¹=R²=R³=H
5 R¹=CH₃, R²=R³=H
6 R¹=H, R²=R³=O
7 R¹=CH₃, R²=R³=O
8 R¹=H, R²=OH, R³=H
9 R¹=CH₃, R²=OH, R³=H
10 R¹=H, R²=OEt, R³=H
11 R¹=CH₃, R²=OEt, R³=H



- 14 R¹=R²=H
15 R¹=CH₃, R²=H
16 R¹=CH₃, R²=CONHCOCCL₃
17 R¹=CH₃, R²=>C(CH₃)₂



- 12 R=H
13 R=CH₃



- 18 R¹=R²=H
19 R¹=H, R²=Ac
20 R¹=, R²=Ac

Maculatae) contains *ent*-kaurenoid acids, previously found in other members of this series [*Viguiera insignis* (2,3) and *Viguiera maculata* (19)].

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Ir spectra were taken on Perkin-Elmer 283b instrument. ¹H-nmr and ¹³C-nmr spectra were obtained on Varian FT-80 spectrometer, in

CDCl₃ solutions with TMS as internal standard. Mass spectra were recorded on a Hewlett-Packard 5985-B spectrometer.

PLANT MATERIAL.—Aerial parts of *V. excelsa* were collected 3 km east of Nopala, Hidalgo, on 11 August 1982 (voucher deposited in the National Herbarium, Instituto de Biología de la U.N.A.M., Reg. No. 219957). Aerial parts of *V. oaxacana* were collected near La Luz Nagore, Oaxaca, on 3 October 1982 (voucher deposited in

TABLE 1. ^{13}C -nmr (CDCl_3 Solution) Data of **2**, **3**, **17**, and **18**

Carbon	Chemical Shift			
	2	3	17	18
1	204.90 (<i>s</i>)	204.37 (<i>t</i>)	40.89 (<i>t</i>)	34.90 (<i>s</i>)
2	105.34 (<i>d</i>)	105.31 (<i>d</i>)	19.24 (<i>t</i>)	75.18 (<i>d</i>)
3	180.71 (<i>s</i>)	182.89 (<i>s</i>)	38.47 (<i>t</i>)	26.57 (<i>t</i>) ^b
4	138.44 (<i>s</i>)	139.50 (<i>s</i>)	43.94 (<i>s</i>)	20.85 (<i>t</i>)
5	139.12 (<i>d</i>)	78.50 (<i>s</i>)	57.21 (<i>d</i>)	37.44 (<i>s</i>)
6	74.28 (<i>d</i>) ^a	74.52 (<i>d</i>)	22.17 (<i>t</i>)	50.75 (<i>d</i>)
7	48.12 (<i>d</i>)	43.43 (<i>d</i>)	41.73 (<i>t</i>)	26.39 (<i>t</i>) ^b
8	75.12 (<i>d</i>) ^a	75.05 (<i>d</i>)	44.61 (<i>s</i>)	35.78 (<i>t</i>) ^a
9	42.16 (<i>t</i>)	42.54 (<i>t</i>)	55.62 (<i>d</i>)	33.42 (<i>t</i>) ^a
10	88.03 (<i>s</i>)	89.32 (<i>s</i>)	39.57 (<i>s</i>)	81.05 (<i>d</i>)
11	149.59 (<i>s</i>)	135.13 (<i>s</i>)	19.15 (<i>t</i>)	47.90 (<i>t</i>)
12	168.60 (<i>s</i>)	169.47 (<i>s</i>)	27.16 (<i>t</i>)	44.61 (<i>s</i>)
13	124.01 (<i>t</i>)	122.45 (<i>t</i>)	45.80 (<i>d</i>)	31.62 (<i>q</i>)
14	21.17 (<i>q</i>)	21.84 (<i>q</i>)	38.25 (<i>t</i>)	25.50 (<i>q</i>) ^c
15	65.35 (<i>t</i>)	128.50 (<i>t</i>)	56.77 (<i>t</i>)	28.39 (<i>q</i>) ^c
16	165.77 (<i>s</i>)	165.84 (<i>s</i>)	89.16 (<i>s</i>)	
17	126.41 (<i>s</i>)	126.46 (<i>s</i>)	70.15 (<i>t</i>)	
18	141.17 (<i>d</i>)	140.94 (<i>d</i>)	28.78 (<i>q</i>)	
19	15.70 (<i>q</i>)	15.67 (<i>q</i>)	176.86 (<i>s</i>)	
20	19.94 (<i>q</i>)	19.99 (<i>q</i>)	15.64 (<i>q</i>)	
21	157.90 (<i>s</i>)	167.97 (<i>s</i>)	108.42 (<i>s</i>)	
22	129.96 (<i>s</i>)	20.84 (<i>q</i>)	26.99 (<i>q</i>)	
23	91.75 (<i>s</i>)		26.89 (<i>q</i>)	
24			51.03 (<i>q</i>)	

^{a,b,c}Assignment may be interchanged.

the National Herbarium, Instituto de Biología de la U.N.A.M., J.L. Villaseñor and G. Delgado, collection No. 268).

ISOLATION OF TERPENOIDS FROM *V. EXCELSA* AND FORMATION OF DERIVATIVES.—Air-dried aerial parts of *V. excelsa* (4.9 kg) were extracted with CHCl_3 at room temperature to give 150 g syrup. This was extensively chromatographed over silica gel (1.5 kg) with hexane and hexane-EtOAc gradient solvent system. From the less polar fractions, 435 mg of stigmasterol was isolated, and from the more polar, 605 mg of budlein A (**1**) was obtained, which was further characterized utilizing the trichloroacetyl carbamate (**2**): ^1H -nmr (80 MHz) δ 6.35 (1H, d, $J=3$ Hz, H-13), 6.18 (1H, dt, $J=1.5, 6$ Hz, H-5), 6.08 (1H, m, H-18), 5.67 (1H, s, H-2), 5.66 (1H, d, $J=3$ Hz, H-13'), 5.31 (1H, m, H-6), 5.25 (1H, m, H-8), 4.97 (2H, br s, H-15, H-15'), 3.75 (1H, m, H-7), 2.57 (1H, dd, $J=4, 16$ Hz, H-9), 2.27 (1H, dd, $J=6, 16$ Hz, H-9'), 1.93 (3H, dq, 19 CH_3 -), 1.80 (3H, dq, 20 CH_3 -), 1.50 (3H, s, 14 CH_3 -); ^{13}C -nmr (20 MHz) Table 1. The rearranged derivative **3** was obtained as previously described (6) [^{13}C -nmr (20 MHz) Table 1]. In addition, 36 mg of clovandiol (**18**) was isolated from the more polar fractions [^{13}C -nmr (20 MHz) Table 1]. This

substance was further characterized, obtaining its derivatives **19** and **20** (17). The fractions of medium polarity (eluted with hexane-EtOAc, 8:2 and 7:3) of the initial column chromatography were combined and the residue (6.7 g) was treated with CH_2N_2 in Et_2O . The reaction mixture containing the methyl esters was chromatographed over silica gel (300 g) using hexane and hexane- C_6H_6 gradient solvent system. From this column chromatography we obtained 215 mg of **5**, 118 mg of **13**, 42 mg of **9** as colorless oil (12), 33 mg of **7** as colorless oil (12), and 44 mg of methyl 12 β -ethoxy-*ent*-kaur-9(11),16-dien-19-oate (**11**), mp and mmp 85-87°, ir (CHCl_3) 1725, 1655, 880 cm^{-1} ; ^1H -nmr (80 MHz) δ 5.32 (1H, br d, H-11), 4.97 (1H, br s, H-17), 4.87 (1H, br sb, H-17'), 3.82 (1H, m, H-12), 3.65 (3H, s, OCH_3), 3.53 (2H, q, $J=7$ Hz, O-CH_2 -), 2.90 (1H, m, H-13), 1.20 (3H, t, $J=7$ Hz, $\text{CH}_2\text{-CH}_3$), 1.17 (3H, s, 18 CH_3 -), 0.98 (3H, s, 20 CH_3 -); ms m/z (rel. int.) 358 (M^+ , 90), 343 (34), 229 (75), 155 (21), 107 (34), 93 (29), 91 (100). These values are identical to those of the sample isolated from *Stevia eupatoria* (16). Finally, from the chromatography of the mixture of methyl esters, 54.2 mg of **15** was obtained. This substance was further characterized utilizing the *bis*-trichloro acetyl carbamate (**16**): ^1H -nmr (80 MHz) δ 8.30 (2H, br s, 2 NH), 5.12 (1H, d,

$J=12$ Hz, H-17), 4.69 (1H, d, $J=12$ Hz, H-17'), 3.65 (3H, s, OCH₃), 2.62 (1H, m, H-13), 1.16 (3H, s, 18 CH₃-), 0.84 (3H, s, 20 CH₃-). The acetonide **17** was obtained with usual procedures, yielding 24 mg of **17**, mp 126-127°, ir (CHCl₃) 2940, 1720, 1465, 1273 cm⁻¹; ¹H-nmr (80 MHz) δ 4.05 (1H, d, $J=9$ Hz, H-17), 3.85 (1H, d, $J=9$ Hz, H-17'), 3.65 (3H, s, OCH₃), 1.37 (3H, s, -C-CH₃), 1.35 (3H, s, -C-CH₃), 1.17 (3H, s, 18 CH₃-), 0.81 (3H, s, 20 CH₃); ¹³C-nmr (20 MHz); Table 1; ms *m/z* (rel. int.) 390 (M⁺, 1), 375 (100), 315 (60), 255 (75), 121 (44).

EXTRACTION AND FRACTIONATION OF V. OAXACANA.—Dried aerial parts of *V. oaxacana* (3.6 kg) were extracted as previously described for *V. excelsa*, providing 43.8 g of residue which was chromatographed over 1.2 kg of silica gel, eluting with hexane and hexane-EtOAc gradient elution system. The chromatography afforded a mixture of hydrocarbons that were rejected, namely, 302 mg of stigmasterol, 64 mg of **4**, and 238 mg of **12**, identified by direct comparison with authentic samples.

ACKNOWLEDGMENTS

The authors thank Mr. José L. Villaseñor (National Herbarium, Instituto de Biología de la U.N.A.M.) for identification of plant material and Miss Laura Alvarez and Mr. Eduardo Huerta (Instituto de Química de la U.N.A.M.) for technical assistance. We are grateful to Dr. A. Ortega for providing a sample of **11**. This work was supported in part by the Consejo Nacional de Ciencia y Tecnología, México, Proyecto PCCBBNA-002049.

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Received 18 May 1984