Phytochemistry, Vol 23, No 10, pp 2285–2288, 1984
Printed in Great Britain

0031–9422/84 $3 00 + 0 00

ENT-BEYERENE AND *ENT*-ATISENE DITERPENES FROM *VIGUIERA INSIGNIS*

GUILLERMO DELGADO, ALFONSO ROMO DE VIVAR, JORGE CARDENAS, ROGELIO PEREDA-MIRANDA* and EDUARDO HUERTA

*Instituto de Química de la Universidad Nacional Autónoma de México†, Circuito Exterior, Ciudad Universitaria, Coyoacán 04510, México, D F, *Departamento de Química Orgánica, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, 11340 México, D F*

(*Revised received* 10 *February* 1984)

Key Word Index—*Viguiera insignis*, Compositae, Heliantheae, diterpenoids, *ent*-beyer-15-ene-12α,19-diol, *ent*-atis-13-en-3β,16α-diol

Abstract—The new diterpenes, *ent*-beyer-15-ene-12α,19-diol and *ent*-atis-13-en-3β,16α-diol, were isolated as minor constituents from the aerial part of *Viguiera insignis* The structures were established by biomimetic transformation, spectroscopic means and comparison with closely related compounds

INTRODUCTION

In a previous communication [1], we reported the diterpenoids **1**–**6** as the major constituents of *Viguiera insignis* Miranda In continuation of our studies on the constituents of *Viguiera* genus [2–6], we now report the isolation and structure determination of two additional new diterpenes which occur as minor constituents of this species

The structures of the new diterpenoids, *ent*-beyer-15-ene-12α,19-diol (**8**) and *ent*-atis-13-en-3β,16α-diol (**12**), were established by a biomimetic transformation, spectroscopic means and comparison with closely related compounds

RESULTS AND DISCUSSION

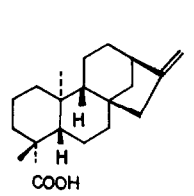
Exhaustive chromatography of the chloroform extract of *V insignis* afforded, in addition to **1**–**7**, two minor isomeric new diols **8** and **12** The first diol **8**, had molecular formula $C_{20}H_{32}O_2$ and its IR spectrum showed hydroxyl (3380 cm^{-1}) and olefin (730 cm^{-1}) absorptions The ^1H NMR spectrum (Table 1) exhibited typical signals of an *ent*-beyer-15-ene derivative an AB system (δ5 68, 5 52, J = 5 7 Hz) of a disubstituted double bond of a cyclopentene which disappeared in the dihydroderivative **9** and three methyl groups [δ1 05 (13Me), 0 97 (4Me) and 0 66 (10Me)] A second AB system centred at δ3 72, 3 41 (2H, J = 11 5 Hz), which shifted downfield on acylation in **10** and **11**, indicated that a primary alcohol was located at C-19 [7] Treatment of **8** with TAI *in situ* [8] afforded a bis-trichloro-acetyl carbamate **11**, therefore showing that the compound was a diol The second hydroxyl group was secondary and axially oriented, because its geminal proton appeared at δ3 62 (1H, *m*, $W_{1/2}$ = 7 Hz) in the ^1H NMR spectrum of **8** and shifted to δ4 94 (1H, *m*, $W_{1/2}$ = 7 Hz) in **11** Upon addition of Eu(fod)$_3$ to a solution of **8**, the signal of the methyl at C-13 showed a significant downfield shift only concordant with a 12β-ol (Table 1) An identical behaviour has been previously found for **6** [1] Therefore, the structure of this diterpene was established as *ent*-beyer-15-ene-12α,19-diol (**8**)

The second new diterpene (**12**, $C_{20}H_{32}O_2$) showed hydroxyl (3600 cm^{-1}) and olefin (860 cm^{-1}) absorptions in the IR spectrum Prolonged treatment of **12** with acetic anhydride–pyridine gave only a monoacetyl derivative **13**, whose IR spectrum still showed hydroxyl absorptions, indicating the presence of a tertiary hydroxyl group The ^1H NMR spectrum of **12** (Table 1) revealed four quaternary methyl groups The singlets at δ1 00 and 0 77 which shifted to 0 87 and 0 84 in the monoacetyl derivative **13** are indicative of a 4,4′-*gem*-dimethyl group adjacent to 3-equatorial hydroxyl (geminal proton δ3 20 *dd*, J = 10, 5 4 Hz) and acetoxyl (geminal proton δ4 45 *dd*, J = 10, 5 4 Hz) groups, respectively [9] In addition, the ^1H NMR spectrum of **12** displayed an ABX pattern (δ5 77, *d*, 1H, J = 8 Hz, 6 06, *dd*, 1H, J = 7, 8 Hz, 2 25 *m*, 1H, $W_{1/2}$ = 12 Hz) characteristic of a bicyclo [2 2 2] octene system of the *ent*-atis-13-ene skeleton [10] The tertiary hydroxyl group was located at C-13, since the quaternary methyl geminal to this group resonated at δ1 13

Furthermore, the chemical shift of the C-10 (δ0 60) and C-13 (δ1 13) methyl groups in **12** indicated that they were shielded by the double bond and therefore *endo* to the olefinic linkage as depicted in **12** [10] Catalytic hydrogenation of **12** gave **14** and this resulted in a significant downfield shift of the signals for the C-10 (δ0 95, Δδ = 0 35) and C-13 (δ1 28, Δδ = 0 15) methyl groups as expected Finally, proof of the structure and stereochemistry of **12** was obtained by chemical correlation with *ent*-beyer-15-ene-3β,12α-diol (**6**)

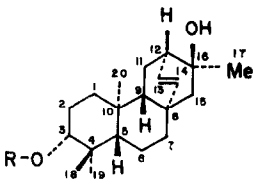
The biomimetic transformation of an *ent*-beyer-15-ene to an *ent*-atis-13-ene derivative has been previously reported under reductive [11] and acid conditions [12] In this case, treatment of **6**, a compound also occurring in *V insignis* and whose structure was confirmed by crystallographic methods [1], with aqueous methanolic hydro-

†Contribution No 673



1 9 (11)-dehydro

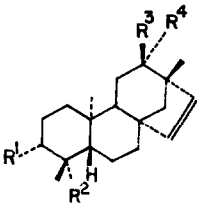
3



12 R = H

13 R = Ac

14 R = H, 13,14-dihydro



	R^1	R^2	R^3	R^4	
2	H	COOH	H	H	
4	OH	CH_3	H	H	
5	OH	CH_2OH	H	H	
6	OH	Me	OH	H	
7	H	CH_2OH	H	H	
8	H	CH_2OH	OH	H	
9	H	CH_2OH	OH	H	15,16 dihydro
10	H	CH_2OAc	OH	H	
11	H	CH_2OTAC	OTAC	H	
15	OAc	Me	OH	H	
16	OAc	Me	= O		

TAC = $CONHCOCCl_3$

chloric acid gave a complex mixture from which *ent*-atis-13-en-3β,16α-diol (**12**) was identified in *ca* 12% yield Better preparative results were obtained when the 3-monoacetyl *ent*-beyer-15-ene derivative **15** previously obtained [1], was treated with Jones reagent to afford the *ent*-beyer-15-en-12-keto compound **16** and the *ent*-atis-13-ene derivative **13** identical in all respects with the acetylation product of *ent*-atis-13-en-3β,16α-diol (**12**), isolated from the natural source

EXPERIMENTAL

Mps are uncorr Aerial parts (1 5 kg) of *V insignis*, collected on October 2, 1981 *ca* 19 km SSE Izúcar de Matamoros, Puebla, Hwy 190, (voucher deposited in the National Herbarium, Instituto de Biología de la UNAM), were extracted and fractionated as previously described [1] Part of the non-polar fraction was treated with CH_2N_2 and chromatographed on silica gel impregnated with $AgNO_3$, affording **1**–**3** as methyl esters In addition, **4** was isolated from these fractions and monogynol **7**, mp 119–120° (lit [13] 119–120°, lit [14] 118–120°), not previously reported from this specimen, was isolated (0 035% of the dry wt) and characterized by standard methods and comparison with an authentic sample

The polar residue (19 5 g) was chromatographed on silica gel and elution with $CHCl_3$–Me_2CO (20 1) gave 245 mg of **8** Mp 143–144° (from *iso*-Pr_2O-EtOAc) $[\alpha]_D^{25}$ + 12 7° (MeOH, *c* 0 110) IR ν_{max}^{KBr} cm^{-1} 3380, 3040, 2920, 2860, 1442, 1363, 1022, 730; ^1H NMR (80 MHz, $CDCl_3$) see Table 1, EIMS (direct inlet) 75 eV, *m/z* (rel int) 304 $[M]^+$ (35 0), 286 (16), 273 (22), 268 (4), 255 (40), 246 (55), 217 (14), 215 (15), 135 (30), 119 (40), 107 (71), 106 (65), 105 (69), 91 (100), 81 (58), 79 (53), 77 (42) [Calculated for $C_{20}H_{32}O_2$ MW 304 2400 Found MW (MS) 304 2390]

Subsequent fractions eluted with the same solvent system gave 355 mg of **5** The fractions eluted with $CHCl_3$–Me_2CO (10 1) gave *ent*-atis-13-en-3β,16α-diol (**12**) (114 mg) Mp 192–193° (from *iso*-Pr_2O–EtOAc) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3600, 3015, 2925, 2860, 2840, 1460, 1440, 1365, 1090, 1021, 1010, 860, ^1H NMR (80 MHz, $CDCl_3$) see Table 1, EIMS (direct inlet) 75 eV *m/z* (rel int) 304 $[M]^+$ (2), 245 (20), 246 (100), 228 (21), 213 (20), 137 (27), 136 (26), 135 (98), 124 (16), 123 (20), 91 (58), 81 (15), 79 (13), 77 (11) [Calculated for $C_{20}H_{32}O_2$ MW 304 2400 Found MW (HRMS) 304 2402] Fractions eluted with $CHCl_3$–Me_2CO (4 1) afforded 415 mg of **6**

Table 1 ^{1}H NMR spectra of compounds **8**–**14** and **16** (80 MHz, $CDCl_3$)*

	8	8a†	8b†	8c†	9	10	11	12	13	14	16
H-3	—	—	—	—	—	—	—	3 20 *dd* (10, 5 4)	4 45 *dd* (10, 5 4)	3 20 *dd* (10, 4 5)	4 45 *dd* (10, 5 4)
H-12	3 62 *m* ($W_{1/2}$ = 7)	5 15 *m* ($W_{1/2}$ = 7)	6 76 *m* ($W_{1/2}$ = 7)	8 50 *m* ($W_{1/2}$ = 7)	3 50 *m* ($W_{1/2}$ = 7)	3 64 *m* ($W_{1/2}$ = 7)	4 94 *m* ($W_{1/2}$ = 7)	2 25 *m* ($W_{1/2}$ = 12)	2 25 *m* ($W_{1/2}$ = 12)	—	—
H-13	—	—	—	—	—	—	—	6 06 *dd* (7, 8)	6 06 *dd* (7, 8)	—	—
H-14	—	—	—	—	—	—	—	5 77 *d* (8)	5 77 *d* (8)	—	—
H-15	5 68 *d* (5 7)	6 06 *d* (5 7)	6 44 *d* (5 7)	6 82 *d* (5 7)	—	5 68 *d* (5 7)	5 80 *d* (5 7)	—	—	—	5 55 *d* (5 7)
H-16	5 52 *d* (5 7)	5 88 *d* (5 7)	6 26 (5 7)	6 67 *d* (5 7)	—	5 52 *d* (5 7)	5 57 *d* (5 7)	—	—	—	6 02 *d* (5 7)
H-17	1 05 *s*	2 15 *s*	3 19 *s*	4 20 *s*	1 00	1 05 *s*	1 04 *s*	1 13 *s*	1 12 *s*	1 28 *s*	1 09 *s*
H-18	0 97 *s*	1 76 *s*	2 50 *s*	3 30 *s*	0 96	0 95 *s*	1 04 *s*	1 00 *s*	0 87 *s*	0 99 *s*	0 87 *s*
H-19	3 72 *d* (11 5)	5 95 *d* (11 5)	7 93 *d* (11 5)	9 87 *d* (11 5)	3 38 *d* (11 5)	4 22 *d* (11 5)	4 48 *d* (11 5)	0 77 *s*	0 84 *s*	0 79 *s*	0 87 *s*
	3 41 *d* (11 5)	5 63 *d* (11 5)	7 62 *d* (11 5)	9 53 *d* (11 5)	3 73 *d* (11 5)	3 83 *d* (11 5)	4 05 *d* (11 5)				
H-20	0 66 *s*	1 17	1 65 *s*	2 15 *s*	0 89 *s*	0 68 *s*	0 70 *s*	0 60 *s*	0 62 *s*	0 95 *s*	0 77 *s*
Others						2 04 (Ac)	8 24 (2NH–)		2 03 (AC)		2 04 (Ac)

*Coupling constants (Hz) in parentheses

†Addition of (**8a**) 0 282, (**8b**) 0 547 and (**8c**) 0 823 mol $Eu(fod)_3$ per mol substrate

Derivatives of **8** *Acetylation* Ac_2O–pyridine treatment of **8** (50 mg) for 1 hr at room temperature yielded the derivative **10** (37 mg after crystallization), mp 130–131° (from *iso*-Pr_2O–EtOAc), IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3600, 2920, 2860, 1725, 1450, 1390, 1370, 1240, 1925, ^{1}H NMR (80 MHz, $CDCl_3$) see Table 1, EIMS (direct inlet) 75 eV *m/z* (rel int) 346 $[M]^+$ (32), 328 (13), 288 (30), 255 (21), 135 (46), 120 (43), 107 (100), 105 (81), 93 (64), 91 (94), 81 (48), 43 (81) *Hydrogenation* Compound **8** (70 mg) in 8 ml MeOH was hydrogenated using Pd–C (9 mg) to afford 65 mg of **9**, Mp 184–186° (from *iso*-Pr_2O–MeOH) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3620, 3003, 2925, 2870, 1460, 1370, 1280, 1130, 1075, ^{1}H NMR (80 MHz, $CDCl_3$) see Table 1, EIMS (direct inlet) 75 eV *m/z* (rel int) 306 $[M]^+$ (6), 276 (13), 275 (56), 258 (27), 257 (100), 186 (13), 175 (24), 123 (33), 95 (37), 81 (31), 79 (52)

Derivatives of **12** *Acetylation* Ac_2O–pyridine treatment of **12** (40 mg) for 72 hr at room temp yielded the derivative **13** (34 mg), mp 123–124° (from *iso*-Pr_2O–EtOAc) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3600, 3000, 2942, 2922, 2860, 1725, 1450, 1395, 1372, 1250, 1120, 1100, 1030, 975, ^{1}H NMR (80 MHz, $CDCl_3$) see Table 1, EIMS (direct inlet) 75 eV *m/z* (rel int) 346 $[M]^+$ (< 1), 328 (1), 288 (100), 228 (24), 137 (50), 135 (99), 124 (33), 123 (35), 92 (31), 91 (75), 81 (22), 79 (19), 43 (66), 41 (12) *Hydrogenation* A soln of **12** (46 9 mg) in EtOH (7 ml) was hydrogenated with Pd–C (8 mg) to give 39 2 mg of **14**, mp 185–186° (from *iso*-Pr_2O–EtOH) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3618, 3000, 2940, 2888, 1470, 1450, 1390, 1370, 1095, 1043, 1020, 995, ^{1}H NMR (80 MHz, $CDCl_3$) see Table 1, EIMS (direct inlet) 75 eV *m/z* (rel int) 306 $[M]^+$ (1), 288 (52), 273 (100), 270 (43), 255 (56), 227 (22), 159 (21), 121 (40), 112 (42), 110 (48), 91 (52), 81 (43), 43 (55)

Conversion of **6** *to* **12** To a chilled soln of *ent*-beyer-15-ene-3β,12α-diol (**6**, 80 mg) in absolute MeOH (5 ml), conc HCl was added (0 11 ml) The soln was then stirred at 0–5° for 3 hr, diluted with H_2O and extracted with $CHCl_3$ The residue obtained was a complex mixture of unidentified products from which 9 6 mg of **12** were separated by silica gel column chromatography, eluted with $CHCl_3$–Me_2CO (20 1)

Conversion of **15** *to* **13** *and* **16** To a soln of 135 mg of **15** in Me_2CO (50 ml) at 0–5°, 0 3 ml of Jones reagent was added, the reaction being monitored by TLC After 15 min, the soln was filtered and usual work-up afforded 96 4 mg residue, which was chromatographed on a silica gel column using *n*-hexane–EtOAc (20 1) as eluent The more mobile component was **16** (19 1 mg) Mp 108–109° (from *iso*-Pr_2O–EtOAc) IR $\nu_{max}^{CHCl_3}$ cm^{-1} 3000, 2980, 2940, 2860, 1729, 1705, 1450, 1390, 1380, 1255, 1090, 1030, 1010, 980, ^{1}H NMR (80 MHz, $CDCl_3$) see Table 1, EIMS (direct inlet) 75 eV *m/z* (rel int) 344 $[M]^+$ (63), 316 (24), 241 (20), 159 (5), 81 (20), 79 (28), 76 (39), 43 (100), 41 (14) The less mobile compound, 32 8 mg, was identical by direct comparison (mmp, IR, ^{1}H NMR) with an authentic sample of **13**

Acknowledgements—We thank Professor W Herz (Florida State University, U S A) for the HRMS, Professor F Walls (Instituto de Química de la U N A M) for an authentic sample of monogynol, and Mr José L Villaseñor (National Herbarium, Instituto de Biología de la U N A M) for identification of plant material This work was supported in part by the Consejo Nacional de Ciencia y Tecnología, México Proyecto PCCBBNA-002049

REFERENCES

1 Delgado, G, Romo de Vivar, A, Ortega, A, Cárdenas, J and Schlemper, E O (1983) *Phytochemistry* **22**, 1227
2 Romo de Vivar, A, Guerrero, C, Díaz, E, Bratoeff, E and Jiménez, L (1976) *Phytochemistry* **15**, 525
3 Romo de Vivar, A, Delgado, G, Guerrero, C, Reséndiz, J and Ortega, A (1978) *Rev Latinoam Quím* **9**, 171
4 Romo de Vivar, A, Bratoeff, E, Ontiveros, E, Lankin, D C and Bhacca, N S (1980) *Phytochemistry* **19**, 1795
5 Delgado, G, Romo de Vivar, A and Herz, W (1982) *Phytochemistry* **21**, 1305

6 Delgado, G, Alvarez, L and Romo de Vivar, A *Phytochemistry* **23**, 675
7 Wenkert, E and Beak, P (1961) *Tetrahedron Letters* 358
8 Samek, Z and Budesinsky, M (1979) *Collect Czech Chem Commun* **44**, 558
9 Hanson, J R (1970) *Tetrahedron* **26**, 2711
10 Ayer, W, Ball, J H, Rodríguez, B and Valverde, S (1974) *Can J Chem* **52**, 2792
11 Pegel, K H, Piacenza, P L, Phillips, L and Waight, E S (1973) *J Chem Soc Chem Commun* 552
12 von-Carsten-Lichterfelde, C, Panizo, F M, de Quesada, T G, Rodríguez, B, Valverde, S, Ayer, W A and Ball, J H (1975) *Can J Chem* **53**, 1172
13 McCrindle, R, Martin, A and Murray, R D H (1968) *J Chem. Soc* (*C*) 2349
14 Caballero, Y and Walls, F (1970) *Bol Inst Quim Univ Nacl Autón Mex* **22**, 79