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LABDANE DITERPENES FROM *BRICKELLIA VERONICAEFOLIA**

J. S. CALDERÓN, L. QUIJANO, M. CRISTIÁ, F. GÓMEZ and T. RÍOS

Instituto de Química de la Universidad Nacional Autónoma de México, Circuito Exterior, Ciudad Universitaria, Coyoacán 04510, México, D. F., Mexico

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Key Word Index—*Brickellia veronicaefolia*; Compositae; Eupatorieae; labdane type diterpenes; 2α,3α-dihydroxycativic acid; 2α-hydroxy-3α-(2-hydroxy-2-methyl-butyryloxy)cativic acid.

Abstract—The aerial parts of *Brickellia veronicaefolia* afforded two new labdane type diterpenes, the structures and stereochemistries of which were established by spectroscopic methods and chemical transformations.

INTRODUCTION

The few species of the genus *Brickellia* (tribe Eupatorieae) which have been investigated chemically have contained thymol derivatives [1], diterpenes [1–4], flavones [5–8] and nerolidol derivatives [2, 9]. *B. veronicaefolia* (H.B.K.) A. Gray has been shown to contain flavones [8], two diterpenes and a nerolidol derivative [1]. In the present paper, we report the isolation and structure elucidation of two new labdane type diterpenes besides the known triterpenes, taraxasterol and taraxasteryl acetate.

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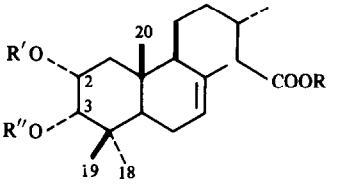
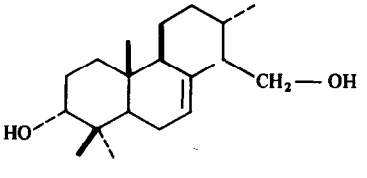
RESULTS AND DISCUSSION

2α,3α-Dihydroxycativic acid (**1a**), $C_{20}H_{34}O_4$, M^+ at *m/z* 338, $[\alpha]_D = -1.45$, was isolated as a crystalline compound, mp 149–150°. The presence in **1a** of a carboxylic acid group was shown by IR absorptions at 3500–2500 and 1700 cm^{-1}, and by the formation of the methyl ester, **1b**, upon treatment with an ethereal solution of diazomethane. It also contained hydroxyl groups (IR absorption at 3250 cm^{-1}) which could be acetylated under mild conditions to give the monoacetate, **1c**, and the diacetate, **1d**. The ^1H NMR spectrum (Table 1) of **1a** showed signals due to three tertiary methyl groups (δ 0.79, 0.90 and 0.99), one secondary methyl group (δ 0.99, *d*, *J*



1a R′=R″=R=H
1b R′=R″=H, R=Me
1c R′=Ac, R″=H, R=Me
1d R′=R″=Ac, R=Me
1e R′=R″=Me—C(Me)—, R=H
1f R′=Ts, R″=R=H
1g R′=H, R″=—CO—C(OH)Me—CH_2—Me, R=H
1h R′=H, R″=—CO—C(OH)Me—CH_2—Me, R=Me
1i R′=Ac R″=—CO—C(OH)Me—CH_2—Me, R=Me



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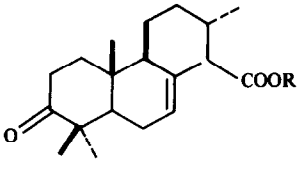

3a R=H
3b R=Me

Table 1. ^{1}H NMR data of compounds **1a** and **1g** and their derivatives (60 MHz, $CDCl_3$, TMS as int. standard)

	1a	1b	1c	1d	1e	1g*	1i
H-2	4.0 *m*	3.99 *ddd*	5.24 *ddd*	5.23 *ddd*	4.14 *td*	4.20 *ddd*	5.24 *ddd*
H-3	3.44 *d*	3.43 *d*	3.55 *d*	5.0 *d*	3.64 *d*	4.95 *d*	5.05 *d*
H-7	5.34 *br s*	5.38 *br s*	5.40 *br s*	5.41 *br s*	5.34 *br s*	5.40 *br s*	1.70 *br s*
H-16	0.99 *d*	0.95 *d*	0.94 *d*	0.95 *d*	1.03 *d*	0.95 *d*	1.03 *d*
H-17	1.67 *br s*	1.65 *br s*	1.66 *br s*	1.68 *br s*	1.68 *br s*	1.72 *br s*	1.70 *br s*
H-18	0.99 *s*	1.00 *s*	0.99 *s*	1.05 *s*	1.02 *s*	1.05 *s*	1.09 *s*
H-19	0.90 *s*	0.91 *s*	0.99 *s*	0.87 *s*	0.97 *s*	0.93 *s*	0.89 *s*
H-20	0.79 *s*	0.80 *s*	0.86 *s*	0.87 *s*	0.77 *s*	0.85 *s*	0.89 *s*
				1.98 *s*			
Ac	—	—	2.1 *s*	2.08 *s*	—	—	1.97 *s*
MeO	—	3.66 *s*	3.68 *s*	3.67 *s*	—	—	3.66 *s*
Gem-diMe	—	—	—	—	1.28 *s* 1.48 *s*	—	—

*Run at 80 MHz; –CO–C(OH)MeEt 1.82 *m*, 1.0 *t*, 1.46 *s*.

J (Hz): 1β,2β = 5.5; 1α,2β = 11; 2β,3β = 2.5; 13,16 = 6.5. Compound **1e**. 1β,2β = 2β,3β = 5; 1α,2β = 10.

= 6 Hz) and a trisubstituted double bond (vinyl proton at δ 5.34 and a vinyl methyl group at 1.67).

The ^{1}H NMR spectrum (Table 1) of the methyl ester **1b** clearly showed two signals due to the methine protons on carbons bearing two secondary hydroxyl groups, as a doublet at δ 3.43 (*J* = 2.5 Hz) and a doublet of doublets of doublets at δ 3.99 (*J* = 2.5, 5.5, 11 Hz). The coupling constants of these signals showed that both hydroxyl groups had to be α-orientated and were placed at C-3 and C-2, respectively. Confirmation of the relative position of these groups was achieved by preparation of the acetonide, **1e**. A similar substitution pattern has been found in diterpenes isolated from plants of the same genus [1, 3].

The structure of **1a** was confirmed by chemical correlation with 3-oxocativic acid (**3a**), a diterpene isolated from *B. veronicaefolia* [1]. Treatment of **1a** with *p*-toluensulfonyl chloride afforded the tosylate, **1f**, which was reduced with lithium aluminium hydride to give the diol, **2**. Oxidation of **2** with an excess of Jones' reagent in acetone gave **3a** which was treated with diazomethane to afford the ester, **3b**, whose IR, ^{1}H NMR and mass spectra were identical to those previously published [1].

2α-Hydroxy-3α-(2-hydroxy-2-methyl-butyryloxy) cativic acid, (**1g**), $C_{25}H_{42}O_6$, M^+ at *m/z* 438, had a carboxylic acid group since on treatment with diazomethane it gave the methyl ester, **1h**. Its ^{1}H NMR spectrum (Table 1) was very similar to that of **1a** showing that both acids had the same substitution pattern but the low field signal at δ 4.95 indicated that the hydroxyl group at C-3 was esterified. The presence of a 2-hydroxy-2-methylbutyrate ester (1715 cm^{-1}) was indicated by fragmentation ions in the mass spectrum at *m/z* 320 $[M - C_5H_{10}O_3]^+$ (12%) and 73 $[C_4H_9O]^+$ (100%) and the signal for the tertiary methyl group at δ 1.46 in the ^{1}H NMR spectrum of **1g**. Acetylation of **1h** with acetic anhydride pyridine gave the acetate methyl ester, **1i** (δ 1.97; 1740 cm^{-1}), the IR spectrum of which showed the presence of a hydroxyl group (3510 cm^{-1}) indicating that **1h** had one tertiary and one secondary hydroxyl group. Final confirmation of the structure of **1g** was achieved by alkaline hydrolysis which furnished the acid **1a**.

Based on all these facts, we propose that the structures **1a** and **1g** as the more likely ones for the new diterpenes.

EXPERIMENTAL

Mps are uncorr. Known compounds were identified by comparison of the IR and ^{1}H NMR spectra. Elemental analyses were performed by Dr. F. Pascher, Germany.

Brickellia veronicaefolia (H.B.K.) A. Gray, was collected in México City at U.N.A.M., in January 1977. A voucher, Calderón 37 A, has been deposited at the Herbarium of the Instituto de Biología (U.N.A.M.). México. Air-dried leaves and flowers (629 g) were extracted with petrol, to give a crude syrup (76.5 g) which was separated into neutral and acid compounds. Chromatography of the neutral fraction gave taraxasterol, taraxasteryl acetate and a mixture of linear hydrocarbons.

2α-3α-Dihydroxycativic acid (**1a**). The acid fraction (42 g) was chromatographed over 500 g Si gel, using C_6H_6 and C_6H_6–EtOAc mixtures as eluants. Fractions eluted with C_6H_6–EtOAc (1:4) afforded 4.0 g **1a** as a crystalline compound, mp 149–150°. IR ν_{max}^{nujol} cm^{-1}: 3500–2500, 3250, 1700, 1480, 1260, 1035, 1050; EIMS (probe) 70 eV *m/z* (rel. int.): 338 $[M]^+$ (1), 320 $[M - H_2O]^+$ (6), 305 $[M - H_2O - Me]^+$ (2), 205 $[M - C_6H_{11}O_2 - H_2O]^+$ (30), 190 $[205 - Me]^+$ (28), 122 $[C_9H_{14}]^+$ (50). (Found: C, 70.90; H, 10.06; O, 18.91 $C_{20}H_{34}O_4$ requires: C, 70.97; H, 10.13; O, 18.91.)

$$[\alpha]_\lambda^{25} = \frac{589 \quad 578 \quad 546 \quad 435 \quad 365}{-1.45 \quad -1.45 \quad -2.2 \quad -9.5 \quad -26.3} (CHCl_3).$$

2α-3α-Dihydroxycativic acid methyl ester (**1b**). Esterification of 200 mg **1a** with CH_2N_2 afforded 160 mg **1b** as an oil. IR ν_{max}^{film} cm^{-1}: 3400, 1730, 1050; EIMS (probe) 70 eV *m/z* (rel. int.): 352 $[M]^+$ (3.5), 334 $[M - H_2O]^+$ (9), 319 $[M - H_2O - Me]^+$ (12), 234 (27), 205 $[M - C_7H_{13}O_2 - 18]^+$ (58), 187 $[205 - H_2O]^+$ (31), 149 (28), 129 $[C_7H_{13}O_2]^+$ (31), 122 $[C_9H_{14}]^+$ (100).

Acetylation of **1b**. 125 mg **1b** was acetylated with Ac_2O–pyridine to give the monoacetate, **1c** (60 mg): oil, IR $\nu_{max}^{CHCl_3}$ cm^{-1}: 3525, 1740, 1720, 1250, 1030 and 55 mg of the diacetate, **1d** oil, IR ν_{max}^{film} cm^{-1}: 1745, 1735, 1255, 1040; EIMS (probe) 70 eV *m/z* (rel. int.): 436 $[M]^+$ (1.3), 376 $[M - HOAc]^+$

(3.5), 316 $[M-2HOAc]^+$ (17), 301 $[M-2HOAc-Me]^+$ (13), 187 $[M-2HOAc-C_7H_{13}O_2]^+$ (41), 122 $[C_9H_{14}]^+$ (34), 43 $[C_2H_3O]^+$ (100).

2α-3α-Acetonide cativic acid (**1e**). 150 mg **1a** in dry Me_2CO (25 ml) and a drop of conc. HCl was refluxed for 24 hr and worked-up as usual to give, after TLC purification, 55 mg **1e**. Colourless oil. IR $\nu_{max}^{film}cm^{-1}$: 3500–2500, 1705, 1220 1055; EIMS (probe) 70 eV *m/z* (rel. int.): 378 $[M]^+$ (1), 363 $[M-Me]^+$ (2), 320 $[M-Me-C_3H_7]^+$ (7), 305 $[M-2Me-C_3H_7]^+$ (6), 220 $[M-C_6H_{11}O_2-C_3H_7]^+$ (17), 205 $[220-Me]^+$ (14), 187 $[M-C_3H_8O_2-C_6H_{11}O_2]^+$ (16), 122 $[C_9H_{14}]^+$ (64), 55 (68), 43 $[C_3H_7]^+$ (100).

Tosylate (**1f**). To a soln of **1a** (519 mg) in dry pyridine (3 ml), TsCl (550 mg) in pyridine (3 ml) was added and the mixture allowed to react at room temp. for 12 hr. After the usual work-up, the residue was purified by CC on Si gel (40 g) to give 262 mg of **1f**. Oil, IR $\nu_{max}^{film}cm^{-1}$: 3500–2500, 3420, 1710, 1450, 1260, 1120, 1060; ^1H NMR (60 MHz, $CDCl_3$): δ 0.70 (3H, *s*, H-20), 0.88 (3H, *s*, H-19), 0.95 (3H, *s*, H-18), 0.92 (3H, *d*, J = 6.5 Hz, H-16), 1.63 (3H, *br s*, H-17), 2.45 (3H, *s*, aromatic Me), 3.53 (1H, *d*, J = 2.5 Hz, H-3), 4.84 (1H, *ddd*, J = 2.5, 6.0, 10.5 Hz, H-2), 5.34 (1H, *m*, H-7), 7.32 and 7.82 (2H each, *d*, J = 8 Hz, aromatic protons).

Labd-7-ene-3α,15-diol (**2**). To a soln of the tosylate, **1f** (260 mg), in 30 ml dry Et_2O was added $LiAlH_4$ (500 mg) in small amounts, the reaction being monitored by TLC. After 2 hr the reaction was worked-up and the residue purified by TLC yielding 48 mg **2** as an oil. IR $\nu_{max}^{film}cm^{-1}$: 3360, 1475, 1250, 1060; ^1H NMR (60 MHz, $CDCl_3$): δ 0.78 (3H, *s*, H-20), 0.88 (3H, *s*, H-19), 0.99 (3H, *s*, H-18), 0.92 (3H, *d*, J = 6.5 Hz, H-16), 3.69 (2H, *t*, J = 6.5 Hz, H-15), 5.41 (*m*, H-7).

3-Oxo-cativic acid (**3a**). To a soln of **2** (48 mg) in Me_2CO (5 ml), Jones' reagent (0.4 ml) was added dropwise with cooling by ice, the reaction being monitored by TLC. After 30 min at room temp., the acid, **3a** (20 mg), was obtained by usual work-up. $[\alpha]_D^{25}$ −25.7° ($CHCl_3$; *c* 0.20) [lit. [1] $[\alpha]_D^{24}$ −35.4° ($CHCl_3$; *c* 1.3)]. IR $\nu_{max}^{film}cm^{-1}$: 3500–2500, 1725, 1705.

3-Oxo-cativic methyl ester (**3b**). Esterification of **3a** (20 mg) with CH_2N_2 provided 12 mg of the ester **3b** after TLC purification. IR, ^1H NMR and mass spectra were identical with those previously published [1].

2α-Hydroxy-3α(2-hydroxy 2-methylbutyryloxy)cativic acid (**1g**). From fractions eluted with C_6H_6–EtOAc (2:3) 6.5 g **1g**, as an amorphous solid was obtained. Mp 104–109°. $[\alpha]_D^{25}$ +11° ($CHCl_3$; *c* 0.38); IR $\nu_{max}^{CHCl_3}cm^{-1}$: 3500–2500, 1715, 1700, 1240; EIMS (probe) 70 eV *m/z* (rel. int.): 420 $[M-H_2O]^+$ (2.8), 320 $[M-C_5H_{10}O_3]^+$ (12), 222 (47), 203 (47), 122 $[C_9H_{14}]^+$ (87), 107 (64), 95 (66), 73 $[C_4H_9O]^+$ (100).

Ester **1h**. Esterification with CH_2N_2 of **1g** (527 mg) provided 415 mg of the ester, **1h**. Colourless oil. IR $\nu_{max}^{film}cm^{-1}$: 3450, 1730, 1715, 1240; ^1H NMR (60 MHz, $CDCl_3$): δ 0.83 (3H, *s*, H-20), 0.88 (3H, *s*, H-19), 0.95 (3H, *d*, J = 6.5 Hz, H-16), 1.02 (3H, *s*, H-18), 1.44 [3H, *s*, =C(OH)Me], 1.68 (3H, *br s*, H-17), 3.63 (3H, *s*, OMe), 4.17 (1H, *ddd*, J = 2.5, 4.5, 12 Hz, H-2), 4.92 (1H, *d*, J = 2.5 Hz, H-3), 5.37 (1H, *m*, H-7); EIMS (probe) 70 eV *m/z* (rel. int.): 452 $[M]^+$ (1), 334 $[M-EtCMe(OH)COOH]^+$ (23), 236 (48), 205 $[M^+-C_5H_{10}COOMe-C_5H_{10}O_3]^+$ (63), 187 $[205-H_2O]^+$ (48), 129 $[C_5H_{10}COOMe]^+$ (48), 122 $[C_9H_{14}]^+$ (88), 73 $[C_4H_9O]^+$ (100), 55 $[C_4H_7]^+$ (99).

2α-Acetyloxy,3α(2-hydroxy-2-methylbutyryloxy)cativic acid methyl ester (**1i**). Acetylation of **1h** (100 mg) with Ac_2O–pyridine gave 80 mg **1i** after TLC purification. Oil. IR $\nu_{max}^{film}cm^{-1}$: 3510, 1740, 1720, 1260, 1235, 1150, 1035; EIMS (probe) 70 eV *m/z* (rel. int.): 494 $[M]^+$ (2), 434 $[M-MeCOOH]^+$ (3.5), 376 $[M-EtC(OH)MeCOOH]^+$ (8), 316 $[376-MeCOOH]^+$ (60), 187 $[316-C_5H_{10}COOMe]^+$ (76), 122 $[C_9H_{14}]^+$ (70), 82 (77), 73 $[C_4H_9O]^+$ (83), 55 $[C_4H_7]^+$ (62), 43 $[MeCO]^+$ (100).

Hydrolysis of **1g**. To a soln of 530 mg **1g** in 20 ml MeOH, 300 mg NaOH was added. The reaction was monitored by TLC. After 1 hr the reaction was worked-up as usual to give 221 mg **1a** as a crystalline solid, mp 149–50°. IR and ^1H NMR spectra were identical to those of the natural compound.

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