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TWO TRITERPENES FROM *MAYTENUS CANARIENSIS*

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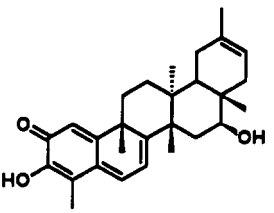
ABSTRACT.—Two new triterpenes with a friedo-oleane skeleton were isolated from the root bark of *Maytenus canariensis*. Their structures were elucidated by means of ^{1}H- and ^{13}C-nmr spectroscopic studies, including homonuclear and heteronuclear correlations and ^{1}H-^{13}C long-range correlation spectra with inverse detection (HMBC).

Maytenus canariensis (Loes.) Kunk. et Sund. (Celastraceae), known locally as "peralillo," is an endemic species that grows in the forest areas of the Canary Islands (1). Shepherds are known to chew the leaves of this species to ward off fatigue, in a similar way that *Catha edulis* Forsk. ("khat") is used in northeast Africa. We have reported in a previous paper that both species have a common ancestor (2).

Our earlier work on *M. canariensis*, in the context of an intensive study of bioactive metabolites from the Celastraceae, yielded dihydro-β-agarofuran sesquiterpenes with insecticidal and antifeedant activity (3,4), triterpenes (5,6), nortriterpene quinone methides showing antitumor activity (7,8), and phenolic nortriterpenes (9).

This paper reports the isolation and structural elucidation of two new compounds based on the friedo-oleane skeleton, the nortriterpene quinone methide, 16β-hydroxyiguesterin [**1**], and the triterpene, 2α-hydroxy-3-oxofriedolean-30-oic acid [**2**], related to cerine (10).

Repeated Sephadex LH-20 and Si gel cc of a hexane/Et_2O extract of the root bark of *M. canariensis* gave two new metabolites, **1** and **2**. Compound **1**, isolated as an amorphous orange-red solid, had the molecular formula $C_{28}H_{36}O_{30}$ (hreims), and its ir spectrum showed absorption bands for hydroxy (3778 cm^{-1}) and conjugated ketone (1725 cm^{-1}) groups; these data and its uv and ^{1}H-nmr (Table 1) spectra indicated that **1** was a



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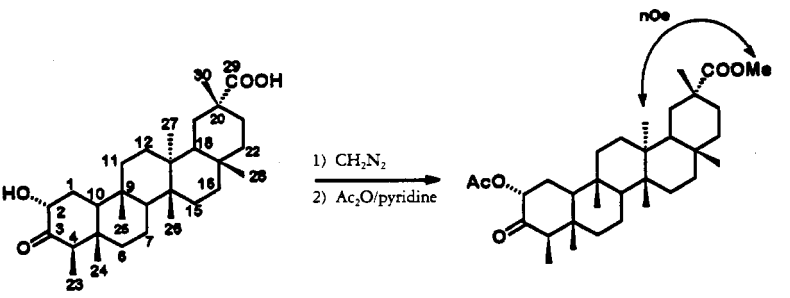

2 **3**

TABLE 1. ^{1}H-Nmr (200 MHz) Data (δ, $CDCl_3$) of Compounds **1–3** (*J* values are given in parentheses).

Proton	Compound		
	1	**2**[a]	**3**
H-1	6.52 d (1.4)		
H-2		4.00 dd (2.9,2.9)	4.95 br s
H-4		2.90 q	2.64 q
H-6	7.03 dd (7.1,1.4)		
H-7	6.32 d (7.1)		
H-16	4.29 br s		
H-18	1.78 t		
H-21	5.27 br s		
Me-23 ...	2.22 s	0.76 d (6.7)	0.88 d (5.3)
Me-24 ...			0.69 s
Me-25 ...	1.48 s		0.85 s
Me-26 ...	1.38 s		0.86 s
Me-27 ...	0.45 s		0.87 s
Me-28 ...	0.97 s		1.08 s
Me-30 ...	1.67 s	1.19 s	1.19 s
-OMe			3.66 s
-OOC-Me			2.17 s

[a] $C_5D_5N/CDCl_3$.

triterpene quinone methide related to iguesterin (7). The only difference between the ^{1}H-nmr spectra of **1** and iguesterin was the presence of a broad singlet at δ 4.29, assigned to a geminal proton of a hydroxyl group, located either in the C, D, or E ring. The 2D ^{1}H-^{13}C-nmr correlations (HMBC) (Figure 1) showed three-bond couplings between the carbon bearing the hydroxyl group with H-18 and Me-28, while in a ^{1}H-^{1}H COSY experiment, coupling between the methylene protons on C-15 and the proton geminal to the hydroxyl group was observed; analysis of these data allowed us to place the hydroxyl group at C-16. The geminal proton at C-16 was determined to be α-equatorial on the basis of molecular mechanics calculations and coupling constants of the preferred conformer (11). Therefore, the structure of compound **1** was established as 16β-hydroxyiguesterin.

Compound **2** was isolated as an amorphous off-white solid. Its ^{1}H-nmr spectrum (Table 1) displayed signals for six angular methyls as singlets and one methyl as a doublet at δ 0.76, which was coupled to a quartet at δ 2.90, assigned to a proton vicinal to a carbonyl group, and a methine proton signal at δ 4.00 as a double doublet for a geminal proton to a hydroxy group. These data suggested a friedo-oleane type triterpene structure for compound **2**. When **2** was treated with CH_2N_2, taken to dryness, and Ac_2O in pyridine was added, the corresponding acetate methyl ester derivative, **3**, was obtained. A detailed study of the spectroscopic data of **3** (Tables 1 and 2) showed that the most notable ^{1}H-nmr differences with respect to those of **2** were the presence of a singlet at δ 2.17 assigned to an acetate methyl, with its geminal proton as a broad singlet at δ 4.95, and a signal at δ 3.66 as a singlet, assignable to a methoxyl group. These data indicated that **3** was the acetate methyl ester derivative of compound **2**. An HMBC nmr

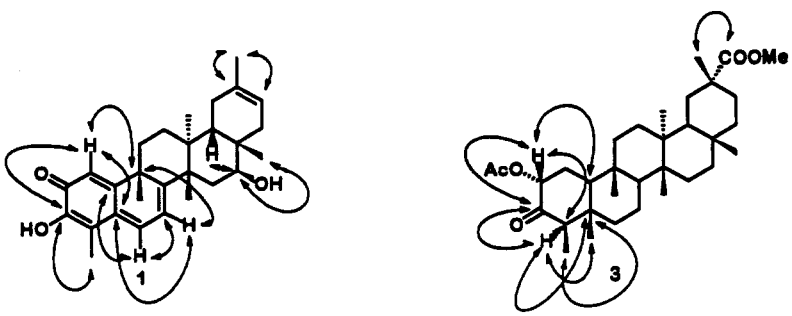

FIGURE 1. Selected HMBC correlations of **1** and **3**.

TABLE 2. ^{13}C-Nmr (100 MHz) Data (δ, $CDCl_3$) of Compounds **1** and **3**.

Carbon	Compound	
	1	**3**
1	120.9 d	28.24 t
2	178.7 s	76.48 d
3	146.4 s	208.08 s
4	117.5 s	54.30 d
5	127.9 s	43.13 s
6	134.3 d	40.96 t
7	118.1 d	18.19 t
8	170.0 s	50.36 d
9	43.2 s	36.65 s
10	165.5 s	53.35 d
11	34.7 t	35.12 t
12	29.7 t	30.10 t
13	41.1 s	39.16[a] s
14	44.5 s	39.35[a] s
15	28.7 t	30.40 s
16	70.4 d	36.16 t
17	36.8 s	29.12 t
18	45.8 d	44.49 d
19	28.0 t	36.93 t
20	136.7 s	40.61 s
21	123.8 d	29.89[b] t
22	29.7 t	29.41[b] t
23	10.7 q	6.47 c
24	39.8 q	14.07 q
25	39.8 q	18.35[c] q
26	16.9 q	16.07[c] q
27	22.2 q	17.48 q
28	25.4 q	31.83 q
30	23.6 q	31.92 q
-*C*OOCH$_3$		179.39 s
-COO*C*H$_3$		51.59 q
-OO*C*-CH$_3$		169.78 s
-OOC-*C*H$_3$		21.16 q

[a,b,c]Interchangeable values. Data are based on ^{1}H-^{13}C 2D experiments.

experiment (Figure 1) showed two-bond couplings between H-4 and the carbonyl carbon (C-3), and between the geminal proton of the acetate group (H-2), and C-3 and a three-bond coupling between H-2 and C-4, confirming the structure of the A ring. A coupling between the protons of an angular methyl (H-30) and the carboxyl carbon of the methyl ester was also observed, locating the carboxyl group at C-20. The stereochemistry of the carboxyl group was established as α by a nOe experiment and the characteristic shift of the Me-27 at δ 0.87 (12). All these data suggested the structure of **3** as shown and confirmed the structure of **2** as 2α-hydroxy-3-oxofriedolean-30-oic acid.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Ir spectra were taken on a Perkin-Elmer 681 spectrophotometer and ^{1}H- and ^{13}C-nmr spectra on a Bruker WP-200 SY instrument in $CDCl_3$ at 200 and 50 MHz, respectively, with TMS as internal standard. The HMBC nmr spectrum was run on a Bruker AMX 400 spectrometer at 400 MHz. Ms were recorded on VG Micromass LTD-ZAF-2F and Hewlett-Packard 5930 mass spectrometers. Optical rotations were measured on a Perkin-Elmer 241 polarimeter and the uv spectra were obtained on a Perkin-Elmer model 550-SE spectrophotometer. Schleicher-Schüell F-100/LS 254 and prep. 1510/LS 254 foils were used for tlc, with Si gel (0.2–0.63 mm) and Sephadex LH-20 employed for cc.

PLANT MATERIAL.—*Maytenus canariensis* was gathered at Icod, Tenerife, Canary Islands, Spain, in October 1986, and a voucher specimen (No. TFC 191641) is on file with the Departamento de Biología Vegetal, Facultad de Ciencias Biológicas, Universidad de La Laguna, La Laguna, Tenerife, Canary Islands, Spain.

EXTRACTION AND ISOLATION.—Root bark (1 kg) of the plant was extracted with *n*-hexane-Et_2O (1:1) (3 liters) in a Soxhlet apparatus. The extract (18 g) was repeatedly chromatographed on Sephadex LH-20 and Si gel using as solvents mixtures of *n*-hexane-$CHCl_3$-MeOH (2:1:1) and of *n*-hexane/EtOAc, respectively, to afford compounds **1** (2.3 mg) and **2** (7 mg).

16β-Hydroxyiguesterin [**1**].—Compound **1** was obtained as an amorphous orange-red solid: $[\alpha]^{20}D$ −100° (*c*=0.46, $CHCl_3$); uv λ max (EtOH) (ε) 414 (9015), 264 (23575) nm; ir ν max ($CHCl_3$) 3378, 2919, 2849, 1705, 1590, 1514, 1437, 1378, 1284, 1220, 1190, 1085, 1014 cm^{-1}; ^{1}H-nmr data (200 MHz), see Table 1; ^{13}C-nmr data (100 MHz), see Table 2; eims *m/z* 420 $[M]^+$ (23), 402 (92), 241 (85), 201 (100); hreims *m/z* 420.26714 (calcd for $C_{28}H_{36}O_3$, 420.26645).

2α-Hydroxy-3-oxofriedolean-30-oic acid [**2**].—Compound **2** was obtained as an amorphous off-white solid; ^{1}H nmr (200 MHz) ($C_5D_5N/CDCl_3$) δ 0.60 (3H, s), 0.77 (3H, s), 1.03 (3H, s), 1.08 (3H, s), 1.17 (3H, s) (for other signals, see Table 1); hreims *m/z* 457.33247 (calcd for $C_{29}H_{45}O_4$ $[M^+-Me]$ 457.33179).

Acetate methyl ester of 2α-hydroxy-3-oxofriedolean-30-oic acid [**3**].—Compound **2** dissolved in MeOH was treated with CH_2N_2 at 4° for 12 h, taken to dryness, and Ac_2O (4 drops) in

pyridine (2 drops) was added. The reaction mixture was left at room temperature for 12 h and purified by prep. tlc with a mixture of *n*-hexane-EtOAc (8:2), to give **3** (4 mg). Compound **3** was obtained as an amorphous off-white solid: $[\alpha]^{20}D$ $-22.5°$ ($c=0.4$, $CHCl_3$); uv λ max (EtOH) (ε) 261 (11807) nm; ir ν max ($CHCl_3$) 2931, 2861, 1743, 1725, 1455, 1373, 1231, 1138 cm^{-1}; ^1H nmr (200 MHz) δ 0.85 (3H, s), 0.87 (6H, s), 1.08 (3H, s), 2.34 (1H, d, $J=12.0$ Hz), 4.95 (1H, br s) (for other signals, see Table 1); ^{13}C-nmr data (100 MHz), see Table 2; eims *m/z* 528 $[M]^+$ (0.3), 468 (3), 385 (2), 332 (1), 271 (12), 249 (11), 223 (10), 190 (10), 164 (7), 109 (100), 81 (48), 43 (64); hreims *m/z* 528.37931 (calcd for $C_{33}H_{52}O_5$, 528.38148).

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