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8β-HYDROXYPIMAR-15-EN-19-OIC ACID FROM *TAXODIUM MUCRONATUM*

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Key Word Index—*Taxodium mucronatum*; Taxodiaceae; diterpenoid; pimarane derivative; 8β-hydroxypimar-15-en-19-oic acid; (+)-thermarol.

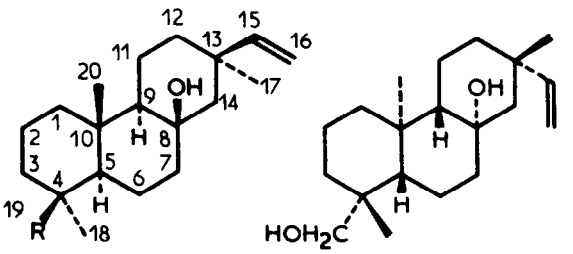
Abstract—A new diterpenoid has been isolated from the leaves and fruits of *Taxodium mucronatum*. Its structure, 8β-hydroxypimar-15-en-19-oic acid, was established by chemical and spectroscopic means.

In the continuation of our search for new natural diterpenoids [1, 2], we have investigated *Taxodium mucronatum* Tenore (*T. distichum* Richards var. *mucronatum* Henry), a typical tree of Mexico (local names: ciprés de Moctezuma, sabino, ahoehoetl), the leaves and fruits of which are used, in this country, in folk medicine [3]. From the leaves and fruits of this tree we isolated a diterpene acid whose structure, 8β-hydroxypimar-15-en-19-oic acid (**1**), was established by chemical and spectroscopic means.

The diterpenoid **1** has a molecular formula of $C_{20}H_{32}O_3$ and its IR spectrum showed hydroxyl (3570, 3460 cm^{-1}), carboxylic (3360–2500, 1710 cm^{-1}) and vinyl (3080, 1635, 980, 910 cm^{-1}) group absorptions. The ^1H NMR spectrum of **1** showed signals for three tertiary methyl groups at δ1.20, 0.88 and 0.79, and for a vinyl group attached to a fully-substituted sp^3 carbon atom (an ABX system, δ_A 5.10, δ_B 5.13, δ_X 5.97; J_{AB} = 1.4 Hz, J_{AX} = 14.2 Hz, J_{BX} = 18.0 Hz), but no signals which could be attributed to geminal protons of hydroxyl groups. On the other hand, the mass spectrum of **1** showed an ion fragment at *m/z* 87, which is typical [4] of diterpenes possessing a C-18 or C-19 carboxylic function.

All the above data indicated a pimar-15-ene or isopimar-15-ene structure with a C-18 or C-19 carboxylic group and a tertiary hydroxyl group for the new diterpenoid. Inspection of the ^{13}C NMR spectrum of compound **1** (Table 1) established that it possessed a C-19 axial carboxylic function, a C-8β hydroxyl group and a C-13β vinyl substituent in a pimarane hydrocarbon skeleton because all of its carbon atom resonances (Table 1) were in complete agreement [5–9] with a pimar-15-en-19-oic acid structure. The presence of the C-8β hydroxyl group was confirmed by the fact that this ^{13}C NMR spectrum was almost identical to that of (−)-thermarol (**2**), an *ent*-pimar-15-ene diterpenoid whose structure is well known [10]. In fact, the differences in the chemical shifts of the C-3–C-6 and C-18–C-20 carbon atoms of compounds **1** and **2** (Table 1) are only due to their different C-19 functions [5–11]. Moreover, the *cis* relationship between the tertiary hydroxyl group at C-8 and the C-13 vinyl side-chain was also confirmed by the ^1H NMR spectral pattern of the C-15 and C-16 protons (see above and refs. [10, 12–14]).

Finally, the structure and absolute configuration depicted in **1** for this new diterpenoid were firmly established by the fact that reduction with lithium aluminium hydride yielded a compound (**3**) which possessed an identical melting point, IR, ^1H NMR and mass spectra to those of



1 R = COOH **2**

3 R = CH_2OH

Table 1. ^{13}C NMR chemical shifts of compounds **1** and **2*** ($CDCl_3$, TMS as internal standard)

	1	2		1	2
C-1	40.0 *t*†	39.5 *t*	C-11	17.6 *t*	17.4 *t*
C-2	19.5 *t*	18.1 *t*	C-12	36.4 *t*	36.1 *t*
C-3	38.1 *t*	35.6 *t*	C-13	37.8 *s*	37.1 *s*
C-4	43.7 *s*	38.7 *s*	C-14	53.1 *t*	53.4 *t*
C-5	55 7 *d*	56 5 *d*	C-15	147.3 *d*	147.5 *d*
C-6	19.0 *t*	18.1 *t*	C-16	112.7 *t*	111.9 *t*
C-7	41 8 *t*	42.3 *t*	C-17	32.4 *q*	32.3 *q*
C-8	73.2 *s*	72.5 *s*	C-18	28.9 *q*	27.0 *q*
C-9	57.2 *d*	57.2 *d*	C-19	182.0 *s*	65 1 *t*
C-10	36.4 *s*	36.4 *s*	C-20	13 6 *q*	16.1 *q*

*Taken from ref. [10].
†SFORD multiplicity.

(−)-thermarol (**2**) [10]; the optical rotations were of the same magnitude although of opposite sign. Thus since the *ent*-pimarene structure of (−)-thermarol is well known [10], the new diterpenoid isolated from *Taxodium mucronatum* must be 8β-hydroxypimar-15-en-19-oic acid (**1**).

EXPERIMENTAL

Mps are uncorr. Plant materials were collected near Tarandacuao, Guanajuato (Mexico) in May 1983.

Extraction and isolation of the diterpenoid. Dried and finely-powdered leaves and fruits (3.5 kg) of *Taxodium mucronatum* were extracted with *n*-hexane in a Soxhlet for 48 hr. The extract (60 g) was chromatographed on a silica gel column (1 kg), which was eluted with *n*-hexane and *n*-hexane–EtOAc mixtures; elution with *n*-hexane–EtOAc (3:1) yielded compound **1** (1050 mg), mp 186–187° (from EtOAc–*n*-hexane); $[\alpha]_D^{26}$ +48.9° ($CHCl_3$, *c* 0.181); IR ν_{max}^{KBr} cm^{-1}: 3570, 3460 (OH group); 3360–2500, 1710 (COOH), 3080, 1635, 980, 910 (vinyl group); 2950, 2910, 2850, 1480, 1450, 1410, 1370, 1265, 1195, 930, 890, 845, 795, 750, 630, ^1H NMR (90 MHz, $CDCl_3$): see discussion of results; ^{13}C NMR (20.15 MHz, $CDCl_3$). see Table 1; EIMS (direct inlet) 75 eV, *m/z* (rel. int.): 320 $[M]^+$ (16), 305 (54), 302 (100), 287 (34), 275 (20), 274 (20), 260 (18), 259 (18), 257 (30), 256 (34), 241 (17), 209 (14), 167 (20), 148 (48), 135 (30), 121 (64), 109 (40), 107 (40), 95 (41), 93 (42), 87 (8), 81 (52), 67 (38), 55 (50), 43 (34), 41 (50) (Found: C, 74.81, H, 10.11. $C_{20}H_{32}O_3$ requires: C, 74.96; H, 10.06%.)

Transformation of compound **1** *into* (+)-*thermarol* (**3**). Compound **1** (18 mg) was treated with excess $LiAlH_4$ in an ethereal soln in the usual manner, yielding the derivative **3** (11 mg, after crystallization from *n*-hexane), mp 146–148°; $[\alpha]_D^{26}$ +16.1° ($CHCl_3$; *c* 0.242); IR ν_{max}^{KBr} cm^{-1}: 3575, 3420, 3390 (OH groups); 3080, 1635, 975, 907 (vinyl group); 2960, 2925, 2850, 1455, 1410, 1385, 1025, 1010, 925, 895, 840; ^1H NMR (90 MHz, $CDCl_3$): vinyl group as an ABX system, δ_A 5.05, δ_B 5.11, δ_X 5.98 (J_{AB} = 1.5 Hz, J_{AX} = 12 Hz, J_{BX} = 18 Hz, H-15 and 2H-16), δ 3.42 and 3.78 (AB system, J_{AB} = 12 Hz, 2H-19), C-Me singlets at δ 0.97 (3H) and 0.89 (6H), EIMS (direct inlet) 10 eV, *m/z* (rel. int.): 306 $[M]^+$ (16), 291 (24), 288 (70), 275 (40), 257 (100), 245 (34), 153 (38), 148 (64), 135 (12), 134 (12), 121 (13), 110 (20), 109 (12), 95 (8). (Found: C, 78.51; H, 11.09. $C_{20}H_{34}O_2$ requires. C, 78.38; H, 11.18%.) Lit [10] (−)-thermarol (**2**). mp 146–147°, $[\alpha]_D$ −17.1° ($CHCl_3$; *c* 0.70), identical IR, ^1H NMR and mass spectra

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