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7β-ACETOXYTRACHYLOBAN-18-OIC ACID FROM THE STEM BARK OF *XYLOPIA QUINTASII**

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Key Word Index—*Xylopia quintasii*; Annonaceae; diterpenes; 7β-acetoxytrachyloban-18-oic acid; chemotaxonomy.

Abstract—A novel diterpene has been isolated from the stem bark of *Xylopia quintasii* and identified as 7β-acetoxytrachyloban-18-oic acid. The taxonomic significance of trachylobanic acids is discussed briefly.

INTRODUCTION

Xylopia quintasii is a large tree of deciduous and evergreen forests distributed throughout tropical west Africa [1]. Its fibrous bark is resistant to decay and attack by termites and is widely used in thatching the walls of dwellings. Nothing is known of the chemistry of *X. quintasii* bark but studies on a number of other species have shown barks containing a range of different alkaloids including 1-benzyltetrahydroisoquinolines [2, 3], aporphines [2–5], 7-oxoaporphines [3, 4, 6] and protoberberines [5]. The fruit of *X. aethiopica*, a species of similar distribution to *X. quintasii*, produces a volatile oil [7] and a number of kaurane-derived diterpenes [8, 9]. In this paper we report the isolation and identification of a novel diterpene. 7β-acetoxytrachyloban-18-oic acid (**1**), from the stem bark of *X. quintasii*.

RESULTS AND DISCUSSION

Extraction of the bark with petrol followed by column chromatography over silica gel afforded a single crystalline compound (yield 0.3%) which analysed for $C_{22}H_{32}O_4$. The presence of an acetyl substituent was indicated by IR, ^{1}H NMR and mass spectral data suggesting that it was a diterpene acetate. A second carbonyl band at 1700 cm^{-1} together with a replaceable proton at δ10 were typical of a carboxylic acid substituent, which was confirmed by synthesis of the methyl ester **2**.

The ^{1}H NMR spectrum of **1** showed singlets at δ1.15 (6H) and 1.00 (3H) for three tertiary methyl groups. A single deshielded proton at 4.64 occurred as a triplet (W = 5 Hz) and was assigned to the ester junction (CHOCOMe). This was confirmed by the hydrolysis of **1** to **3** which caused a shielding of 1.04 ppm.

Compound **1** showed no UV absorption and could not be hydrogenated. No downfield signals, other than those already assigned, occurred in either ^{1}H or ^{13}C NMR spectra. These data preclude the possibility of unsaturation and therefore require **1** to have a pentacyclic skeleton. The presence of broad multiplets between δ0.55 and 0.90 in **1** and all derivatives were typical of a cyclopropane system and suggest that **1** belongs to the trachylobane series of diterpenes [10].

The carboxylic acid moiety of **1** was assigned to C-18 on the basis of the following observations. On conversion of **1** to **2** resonances for H-17, H-19 and H-20 were unchanged whereas in C-4 axial carboxylic acids (19 acids) H-20 occurs at δ0.79 [11]. Reduction of **1** to **4** causes shielding of 0.45 ppm in H-19, twice the observed shielding for an axial substituent [12]. The H-18 protons of **4** and **5** occur as AB quartets centred at δ3.14 and 3.20, whereas in the corresponding 19-hydroxymethyl compounds they occur below 3.50 [12]. Finally, reduction of the keto acid **7** gave trachyloban-18-oic acid (**8**) which was identified as its methyl ester (**9**) [10].

The occurrence of the C*H*-OCOMe proton as a triplet, showing coupling to only two other protons, restricts it to either C-1, C-3, C-7 or C-11. The absence of diaxial coupling, as evidenced by the small width of the signal, requires that it is in an equatorial configuration, thus permitting the acetoxy substituents to be assigned the axial position. Placement of the ester at C-3 is excluded by the ready formation of **7** whereas in 3-hydroxy-4-carboxylic acids decarboxylation occurs on attempted oxidation [10]. In neither of the two keto compounds, **7** and **10**, are resonances observed below δ2.60. By contrast published data for 1-keto diterpenes indicates considerable deshielding of H-2 and H-20 [13] and for 11-keto diterpenes similar, although less pronounced, deshielding of H-1 and H-12 [10].

These data clearly support the placement of the acetoxy substituent at C-7. Further evidence in favour of this is found from a study of the ^{1}H NMR spectra of **7** and **10**. In both the ABX system due to the isolated H-6α, H-6β and H-5α are clearly visible. The occurrence of H-6α as a double doublet centred at δ2.43 (cf. 2.93 in the corresponding 19-carboxylic acid [11]) adds support to the assignment of the carboxylic acid to an equatorial position in **1**. The relative configuration at C-4 and C-7 is

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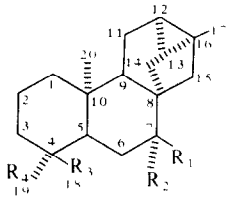

	R_1	R_2	R_3	R_4
1	OCOMe	H	COOH	Me
2	OCOMe	H	COOMe	Me
3	OH	H	COOH	Me
4	OCOMe	H	CH_2OH	Me
5	OH	H	CH_2OH	Me
6	OCOMe	H	CH_2OCOMe	Me
7	$R_1 = R_2 = O$		COOH	Me
8	H	H	COOH	Me
9	H	H	COOMe	Me
10	$R_1 = R_2 = O$		COOMe	Me
11	OH	H	Me	Me
12	H	H	Me	COOH
13	H	OH	Me	COOH

Table 1. ^{13}C NMR spectra of trachyloban 18 and 19 acids (shift values, ppm)

	1	**7**	**12** (18)	**13** (19)
C-1	38.4 *t*	39.8 *t*	39.5 *t*	39.7 *t*
C-2	16.1 *t*	17.2 *t*	18.7 *t*	19.4 *t*
C-3	37.0 *t*	37.8 *t*	37.8 *t*	39.3 *t*
C-4	46.8 *s*	46.4 *s*	43.7 *s*	43.6 *s*
C-5	48.8 *d*	44.8 *d*	57.0 *d*	54.0 *d*
C-6	21.1 *t*	34.0 *t*	21.8 *t*	24.9 *t*
C-7	78.3 *d*	214.7 *s*	39.2 *t*	74.2 *d*
C-8	44.3 *s*	53.3 *s*	40.8 *s*	47.4 *s*
C-9	42.7 *d*	36.9 *d*	52.2 *d*	52.2 *d*
C-10	37.7 *s*	37.2 *s*	38.9 *s*	32.2 *s*
C-11	19.2 *t*	19.7 *t*	19.7 *t*	22.3 *t*
C-12	20.4**d*	19.4**d*	20.6 *d*	24.0 *d*
C-13	23.9**d*	22.6**d*	24.3 *d*	21.0 *d*
C-14	33.1 *t*	29.8 *t*	33.1 *t*	38.5 *t*
C-15	50.4 *t*	46.7 *t*	50.4 *t*	45.4 *t*
C-16	22.4 *s*	24.5 *s*	22.4 *s*	20.1 *s*
C-17	20.5 *q*	16.0 *q*	20.6 *q*	20.9 *q*
C-18	184.7 *s*	183.5 *s*	28.9 *q*	29.2 *q*
C-19	28.9 *q*	20.2 *q*	184.7 *s*	179.9 *s*
C-20	12.5 *q*	14.0 *q*	12.5 *q*	13.1 *q*
COMe	171.9/20.4			

Spectra for **1**, **7** and **12** were run in $CDCl_3$, for **13** in C_5D_5N.
* Interchangeable signals.

also indicated by the equivalence of the H-18 protons in **6**. This phenomenon has been observed in other diterpenes where the C-4 substituent is equatorial and the C-7 substituents axial [14, 15], whereas in cases where both C-4 and C-7 substituents are equatorial the H-18 protons are observed as an AB quartet [15].

On the basis of the above facts the isolated diterpene was identified as **1**. This compound does not appear to have previously been isolated but a number of the derivatives have been prepared, and compounds **4**, **5** and **11** occur in *Sideritis canariensis* (Labiatae) [16, 17]. Comparison of data for these compounds with that already published showed good agreement.

During this study ^{13}C NMR spectra were obtained for **1** and **7**. Comparison of shift values with those reported for trachyloban-19-oic acid (**12**) [18] and ciliaric acid (**13**) [19] (Table 1) showed that in the 18 carboxylic acid series C-5 and particularly C-9 show marked shielding.

This represents the first record of a trachylobane diterpene from the Annonaceae. Its occurrence, together with the absence of detectable amounts of alkaloids, suggests an interesting chemotaxonomic dichotomy within the genus *Xylopia*. In the stem barks of all three west African species so far investigated, *X. quintasii*, *X. aethiopica* (Hasan, C. M., Healey, T. M. and Waterman, P. G., unpublished results) and *X. acutiflora* (ibid.), only diterpenes have been found. By contrast, the barks of five species from South America [4, 6], South-east Asia [2, 3] and Madagascar [6] have yielded alkaloids but no diterpenes.

In wider terms, trachyloban-18-oic diterpenes have now been found in three plant families, the Annonaceae, Labiatae [16, 17] and Leguminosae [10]. By contrast, trachylobane-19-oic derivatives have been found only in the Compositae, where they occur quite widely [11, 19, 20–24].

EXPERIMENTAL

Mps are uncorr. UV spectra were run in EtOH and IR spectra as KCl discs. ^1H NMR were recorded at 90 MHz in $CDCl_3$ using TMS as int. standard. ^{13}C NMR were recorded in the same solvent at 25.1 MHz using the FT mode. MS were recorded at 70 eV (probe).

Plant material. Stem bark of *Xylopia quintasii* Engl. *et* Diels was collected in the Douala-Edea Forest Reserve, west Cameroon, during June 1976. A voucher specimen, P. G. Waterman and D. McKey 863, has been deposited at the herbarium of the Royal Botanic Gardens, Kew.

Isolation and characterization of 7β-acetoxytrachyloban-18-oic acid (**1**). Ground stem bark (300 g) was extracted with petrol (bp 40–60°), then $CHCl_3$ and finally MeOH. The petrol extract was concd. and subjected to CC over Si gel. Elution with cyclohexane–EtOAc (9:1) gave **1** (800 mg) recrystallized from $CHCl_3$–petrol (bp 60–80°) as needles, mp 246–248° (lit. [16] 245–246°). $[\alpha]_D^{26.5}$ + 20.2° (*c* 0.1, $CHCl_3$). (Found: M^+ 360.2280; $C_{22}H_{32}O_4$ requires 360.2300.) IR ν_{max} cm^{-1}: 3200, 1725 (OCOMe), 1700 (COOH). ^1H NMR: δ 1.00 (3H, *s*, H-20), 1.15 (6H, *s*, H-17, H-19), 2.02 (3H, *s*, H-22), 4.64 (1H, *t*, $W = 5$ Hz, H-7). EIMS *m/z* (rel. int.): 360 $[M]^+$ (5), 301 (21), 300 (100), 285 (15), 255 (7), 157 (25). Compound **1** (100 mg), in dry THF at −15° under N_2, was treated with 1 M diborane (Me_2S complex) (0.7 ml) to give 7β-acetoxytrachyloban-18-ol (**4**) (97 mg), mp 146–150° (lit. [17] 156–160°). (Found: M^+ 346.2503; $C_{22}H_{34}O_3$ requires 346.2508.) ^1H NMR: δ 0.70 (3H, *s*, H-19), 3.14 (2H, AB*q*, $J = 12$ Hz, H-18). Compound **4** (76 mg) in pyridine (2.5 ml) was reacted with *p*-toluene sulphonyl chloride (161 mg) at 0° for 120 hr. Reduction of the resulting tosylate with $LiAlH_4$ gave a mixture which was sepd by CC over Si gel solvent: petrol (bp 60 80°)–EtOAc (4:1) to give 7β-hydroxytrachylobane (**11**) (50 mg), mp 95–100° (lit. [16] 105–106°). (Found: M^+ 288.2427; $C_{20}H_{32}O$ requires 288.2453.)

7β-Acetoxy-methyltrachyloban-18-oate (**2**). Compound **1** (150 mg), on treatment with CH_2N_2, gave **2** in quantitative yield. Needles from MeOH, mp 154–157°. (Found: M^+ 374.2456;

$C_{23}H_{34}O_4$ requires 374.2457.) IR ν_{max} cm^{-1}: 1720, 1244 (equatorial COOMe [25]). ^{1}H NMR: δ 0.99 (3H, *s*, H-20), 1.12 (3H, *s*, H-17), 1.14 (3H, *s*, H-19), 2.07 (3H, *s*, H-22), 3.60 (3H, *s*, COOMe), 4.60 (1H, *t*, $W_{\frac{1}{2}}$ = 5 Hz, H-7). EIMS *m/z* (rel. int.): 374 $[M]^+$ (5), 315 (44), 314 (100), 299 (17), 255 (39), 254 (32), 239 (16), 157 (22). Reduction of **2** with $LiAlH_4$ in dry THF gave 7β-hydroxytrachyloban-18-ol (**5**), mp 170–175° (lit. [16] 175–176°). (Found: M^+ 304.2399; $C_{20}H_{32}O_2$ requires 304.2402.) ^{1}H NMR: δ 0.70 (3H, *s*, H-19), 3.46 (2H, AB*q*, *J* = 12 Hz, H-18). Acetylation of **5** yielded 7β,18-diacetoxytrachylobane (**6**), mp 130–134° (lit. [17] 134–136°). (Found: M^+ 388.2616; $C_{24}H_{36}O_4$ requires 388.2613.) ^{1}H NMR: δ 3.68 (2H, *s*, H-18).

7β-Hydroxytrachyloban-18-oic acid (3). Saponification of **1** with alc. KOH for 1 hr gave **3** in 90% yield, recrystallized from $CHCl_3$–petrol (bp 60–80°) as needles, mp 215–220°. (Found: M^+ 318.2205; $C_{20}H_{30}O_3$ requires 318.2195.) IR ν_{max} cm^{-1}: 3350, 1705. ^{1}H NMR: δ 0.93 (3H, *s*, H-20), 1.15 (6H, *s*, H-17, H-19), 3.60 (1H, *br s*, $W_{\frac{1}{2}}$ = 5 Hz, H-7). EIMS *m/z* (rel. int.): 318 $[M]^+$ (39), 301 (49), 300 (100), 285 (33), 255 (32), 239 (22), 199 (19), 195 (18), 185 (52). Oxidation of **3** with Jones reagent gave 7-oxotrachyloban-18-oic acid (**7**) as an oil (Found: M^+ 316.2035; $C_{20}H_{28}O_3$ requires 316.2038.) ^{1}H NMR: δ 1.10 (3H, *s*, H-20), 1.19 (6H, *s*, H-17, H-19), 2.06 (1H, *dd*, J_1 = 16 Hz, J_2 = 2.5 Hz, Hβ-6), 2.43 (1H, *dd*, J_1 = 16 Hz, J_2 = 14 Hz, Hα-6). EIMS *m/z* (rel. int.): 316 $[M]^+$ (100), 270 (11), 220 (14), 174 (15), 147 (37). Treatment of **7** with CH_2N_2 gave 7-oxo-methyltrachyloban-18-oate (**10**), mp 106–110° (lit. [16] 110–111°). (Found: M^+ 330.2196. $C_{21}H_{30}O_3$ requires 330.2195.)

Trachyloban-18-oic acid (**8**). Compound **7** (32 mg) was refluxed with *p*-toluene sulphonyl hydrazide (37 mg) in EtOH for 24 hr. The reaction mixture was evapd and dissolved in 5 ml DMF–sulfolane (1:1). $NaBH_3CN$ (25 mg) and *p*-toluene sulphonic acid (50 mg) were added and the mixture heated to 110° for 10 hr with the addition of further reagents after 3 hr [26]. The reaction mixture was diluted with H_2O and extrd with Et_2O to give **8** (15 mg) as an oil. Methylation of **8** with CH_2N_2 gave methyltrachyloban-18-oate (**9**) mp 107–109° (lit. [10] 110–112°). (Found: M^+ 316.2417. $C_{21}H_{32}O_2$ requires 316.2402.)

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