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SHORT COMMUNICATION

TERPENOIDS—VII.[1]

CONSTITUENTS OF *EUPHORBIA LATERIFLORA* SCHUM. AND THONN.

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Abstract—The chemical constituents of *Euphorbia lateriflora* Schum. and Thonn. (Euphorbiaceae) have been investigated and the following compounds were isolated and identified: cholest-5-ene, moretenone, moretenol, lupeol, 24-methylene-cycloartanol and β-sitosterol. The occurrence in nature of a non-oxygenated steroid, cholest-5-ene and of moretenone is reported for the first time.

Euphorbia lateriflora Schum. and Thonn. (Euphorbiaceae) is a small shrub of common occurrence in wet regions of West Africa which is planted in every village because of its medicinal use. The leaves are thick and narrow with only the midrib showing through the smooth surface so that it appears like a microphyllous leaf; they are deciduous. The white milky juice is known to soften muscles locally and is used for extracting thorns. Patients infected with the Dracunculus soak their leg with a decoction of the stem.[2]

The present investigation was undertaken in order to characterize some of the chemical constituents of the plant. Chloroform extraction of the dried stems and leaves, yielded about 11 per cent of crude substance. This extract, when further treated with hexane, gave a mixture which showed at least seven components by TLC, using benzene–ethyl acetate (9:1) as solvent mixture. Through a combination of chromatographic procedures and fractional crystallizations, six pure compounds could be separated and identified as: cholest-5-ene, moretenone, β-sitosterol, moretenol, lupeol, and 24-methylene-cycloartanol.

The occurrence in nature of a non-oxygenated steroid, is hereby reported for the first time. It was separated from the least polar fraction following several crystallizations from chloroform–methanol and was identified by spectroscopic evidence and comparison with an authentic sample. Its mass spectrum showed the following peaks m/e 370 (M^+), 355 (M-15), 257 (M-C_8H_{17}), 216 (M-C_8H_{17}-41), 215 (M-C_8H_{17}-42), 122 (retro Diels Alder fragment).[3]

[1] D. LAVIE, MAHENDRA K. JAIN and J. KIRSON, *J. Chem. Soc.* 1347 (1967).

[2] J. HUTCHINSON and J. M. DALZIEL, *Useful Plants of West Africa*, p. 142. Crown Agents for Overseas Governments and Administrations, London (1937).

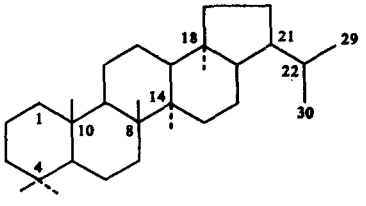
[3] H. BUDZIKIEWICZ, C. DJERASSI and D. H. WILLIAMS, *Structure Elucidation of Natural Products by Mass Spectrometry*, Vol. II, p. 94. Holden-Day, San Francisco (1964).

Moretenone (21-αH-hop-22(29)-en-3-one) is also reported for the first time to occur in nature.

The corresponding alcohol moretenol was characterized and identified with an authentic sample. From this compound moretanone, moretene, moretane and the acetoxy-norketone were prepared and all displayed the expected physical constants.[4] Moretenol has been reported to occur in *Ficus macrophylla*[4] and is the only compound of this series to have been isolated heretofore.

TABLE 1. NUCLEAR MAGNETIC RESONANCE SIGNALS OF HOPANE DERIVATIVES IN c/s

	Methyl signal at							
	4α*	4β*	10β*	8β	14α	18α	29H	30H
21 αH-hopane (moretane)	47·5	48·5	50·5	59	56·5	38	47 and 53·5 (J=6·5 c/s)	
21 αH-hopan-3-one (moretanone)	56	61	64	61	56	40	47 and 53·5 (J=6·5 c/s)	
21 αH-hop-22,29-en-3-one (moretenone)	57	61	64	61	57	42	100 (3H)	280 (2H)
21 αH-hop-22,29-en-3β-ol (moretenol)	45·5	50	59	59	56·5	41	100 (3H)	280 (2H)
21 αH-hop-22,29-ene-3β-acetate		50·0 (3)		58	56	41	100 (3H)	280 (2H)
21 αH-30-norhopan-22-one-3β-acetate		50·5 (3)		58	56·5	42	129 (3H)	—
21 αH-30-norhopan-3,22-dione	57	61	64	61	57	42	129 (3H)	—
21 αH-hopan-22,29-epoxy-3-one	56	60·5	64	60·5	56	39	75 (3H)	152 (2H)
21 αH-hopan-22,29-diol-3-one	56	60	64	60	56	40·5	75 (3H)	



* No specific allocations for these three groups

The NMR spectra of the various derivatives of moretenone are recorded in Table 1. The chemical shifts for the methyl groups at C(4α), (4β), (8β), (10β) and (14α) are essentially the same as those reported for 21-βH-hopane.[5] The C(18α) methyl group is subject to the deshielding effect of the groups of the side chain. The chemical shift reported for the methyl group at C(18α) in hopane (21-βH)[6] and in 21-αH-hopane is 42·5 and 38·0 c/s respectively. A similar difference has been noted for adiantone and iso-adiantone (35·5 and 42 c/s) respectively.[6] The chemical shifts for the methyl signals in other derivatives of hopane (21-βH) are not available and the comparison with the 21-αH-hopane series (reported herein) is there-

[4] M. N. GALBRAITH, C. J. MILLER, J. W. L. RAWSON, E. RITCHIE, J. S. SHANNON and W. C. TAYLOR, *Aust. J. Chem.* **18**, 226 (1965).

[5] S. HUNECK and J. M. LEHN, *Bull. Soc. Chim. Fr.* 1702 (1963).

[6] H. AGETA, K. IWATA, Y. ARAI, Y. TSUDA, K. ISOBE and S. FUKUSHIMA, *Tetrahedron Letters* 5679 (1966).

fore not possible. However, these values may be compared with those reported recently for 21-αH-30-nor-hopane (isoadiantane).[7]

From the second solvent fraction of the chromatography (ether–hexane 1:1 eluate) 24-methylene-cycloartanol was obtained. It could be identified by comparison with an authentic sample of the corresponding 24,31-epoxide.

EXPERIMENTAL

Melting points are uncorrected. Optical rotations were determined in $CHCl_3$. I.r. spectra were recorded in KBr pellet on a Perkin Elmer 137 spectrophotometer equipped with a NaCl prism; u.v. spectra were recorded on a Cary 14 instrument; NMR spectra were determined on a Varian A-60 spectrometer, for 5–10 per cent solutions in $CDCl_3$ containing tetramethylsilane as internal standard. Mass spectra were taken with an Atlas CH4 instrument, the samples being introduced directly into the source through a vacuum lock. Analyses were performed in the microanalytical laboratory of our Institute, under the direction of Mr. R. Heller.

The stems and leaves of *Euphorbia lateriflora* Schum. and Thonn. were cut into chips (3–5 cm long) immediately after collection and spread for drying until the moisture content was less than 15 per cent, they were then crushed and further air dried. The milled stems and leaves (2 kg) were soaked in $CHCl_3$ at room temperature and the solvent changed thrice. The combined extracts were evaporated to dryness and the residue was treated with hot hexane (3 × 1 l.). The hexane solution was decanted, evaporated and the residue chromatographed over Al_2O_3 (E. Merck, 40 g for 1 g of residue). Elution was carried out first with benzene then with ether–hexane (1:1).

1. *The Benzene Eluate*

The solvent was evaporated and the solid (5 g) rechromatographed over neutral Al_2O_3 (activity I, Woelm, 200 g) packed as a slurry in pentane. This column was eluted with pentane, and then with 2 per cent ether in pentane, 20-ml fractions being collected as follows: with pentane—fr. 1–3 waxy solid, fr. 4–6 waxy solid giving positive Liebermann–Burchard reaction, fr. 7–20 traces of waxy solid; with ether in pentane 2 per cent—fr. 21–24 resinous solid, fr. 25–36 crystalline solid, fr. 37–50 traces of oil.

Isolation of cholest-5-ene. The residue (600 mg) obtained from fractions 4–6 was rechromatographed over neutral Al_2O_3 (Woelm, activity I, 50 g) and 10-ml fractions were collected. The fractions 6–9 of this chromatography giving a positive Liebermann–Burchard reaction were combined, and the residue recrystallized five times from acetone, plates, m.p. 88–91°, u.v. end absorption (at 210 nm ϵ 3200); ν_{max} 1630 and 820 cm^{-1}; NMR δ5·2 (triplet 1H), 0·66 (3H) 0·82 (3H) 0·90 and 0·96 (6H). (Found: C, 91·5; H, 8·3 per cent; M^+, 370. $C_{27}H_{46}$ required: C, 91·74; H, 8·26 per cent. Mol wt., 370·5.) Compared with an authentic sample of cholest-5-ene it showed undepressed mixture m.p. and had identical i.r. and NMR spectra.

Isolation of moretenone. The fractions 25–36 were combined, evaporated and the residue was carefully crystallized from a mixture of $CHCl_3$–MeOH (3:1). The first crop, after five recrystallizations, gave needles, m.p. 202–203°, $[\alpha]_D$ +53° (*c.* 1·3); u.v. end absorption; ν_{max} 1710, 1640 and 885 cm^{-1}. (Found: C, 84·7; H, 11·1; M^+, 424. $C_{30}H_{48}O$ required: C, 84·8; H, 11·4 per cent; M wt., 424·6.)

Preparation of moretenol. Upon reduction of moretenone with $LiAlH_4$, the alcohol moretenol was obtained, m.p. 236°, $[\alpha]_D$ +27° (*c.* 1·6). (Found: C, 84·23; H, 11·6. $C_{30}H_{50}O$ required: C, 84·4; H, 11·8 per cent.) The acetate was prepared with acetic anhydride and pyridine at room temperature (24 hr), it crystallized from methanol, m.p. 285–286°, $[\alpha]_D$ +25° (*c.* 1·7); ν_{max} 1745, 1250, 1030 and 885 cm^{-1}. The identity of moretenol was confirmed by comparison with an authentic sample.

Ozonization of moretenone. It was effected as usual and worked-up by the zinc–acetic acid method yielding 30-*norketone-moretenone*, m.p. 276–277°, $[\alpha]_D$ +38° (*c.* 1·2). (Found: C, 81·6; H, 10·7. $C_{29}H_{46}O_2$ required: C, 81·6; H, 10·9 per cent.)

Moretanone-22,29-epoxide. Moretenone (100 mg) in benzene solution was treated with perbenzoic acid (60 mg) for 8 hr. The reaction mixture was worked-up as usual and the product crystallized from acetone, m.p. 196–198°, $[\alpha]_D$ +50° (*c.* 1·6). (Found: C, 81·5; H, 10·8. $C_{30}H_{48}O_2$ required: C, 81·7; H, 11·0 per cent.)

2. *The Ether–Hexane* (1:1) *Eluate*

The solvent was evaporated and the residue crystallized from $CHCl_3$–MeOH. The first crop of solid showed a very faint Liebermann–Burchard reaction and was discarded, the mother liquor was evaporated and the residue recrystallized from acetone, needles, m.p. 136–137°, $[\alpha]_D$ −37° (*c.* 1·1) (acetate m.p. 127°), it was characterized as β-sitosterol and identified by underpressed mixture m.p. and identical i.r. and NMR spectra with an authentic sample.

[7] H. Ageta and K. Iwata, *Tetrahedron Letters* 6069 (1966).

The mother-liquor from the crystallization of β-sitosterol was evaporated to dryness and the residue acetylated with acetic anhydride and pyridine (24 hr at room temperature). The acetylated mixture was dissolved in acetone (1 g in 40 ml) and left at room temperature. The first crystalline deposit was recrystallized from the same solvent, and identified as 24-methylene-cycloartanyl acetate, m.p. 116–117°, $[\alpha]_D + 54°$ (c. 1·3); ν_{max} 1745, 1640, 1250, 880 cm^{-1}; u.v. end absorption. (Found: C, 82·0; H, 10·1; M^+, 482. $C_{33}H_{54}O_2$ required C, 82·1; H, 11·3 per cent; M wt., 482·76.) Hydrolysis of the acetate afforded 24-methylene-cycloartanol, m.p. 125–128°, $[\alpha]_D + 45°$ (c. 1·0).

For identification, the alcohol was epoxidized with perbenzoic acid to 24,31-epoxymethylene-cycloartanol which was compared with an authentic sample, identical i.r. and NMR spectra.[8]

The mother liquors from the crystallization of the 24-methylene-cycloartanyl acetate were combined and the residue, after evaporation of the solvent, was chromatographed on a column of silica H (E. Merck, 50 g for 1·8 g of residue) in benzene solution, and fractions of 20 ml each were collected.

Fractions 15–17 of this chromatography afforded a solid which after recrystallization had m.p. 209–211°, $[\alpha]_D + 40°$ (c. 1·8), and was identified as *lupenyl acetate* by comparison with an authentic sample (identical i.r. and NMR and undepressed mixture m.p.). Fractions 19–20 gave, after several recrystallizations, moretenyl acetate, m.p. 283–285°, $[\alpha]_D + 24°$ (c. 1·8), identified by undepressed mixture m.p. and identical i.r. and NMR spectra with an authentic sample.

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[8] G. PONSINET and G. OURISSON, *Phytochem.* 4, 799 (1965).