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ENT-KAURANES AND OLEANANES FROM *CROTON LACCIFERUS*

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Key Word Index—*Croton lacciferus*; Euphorbiaceae; *ent*-kauranes; oleananes; insecticidal activity.

Abstract—The roots of *Croton lacciferus* furnished three *ent*-kauranoids, two of which are new natural products, and two oleananes. Two of the kauranoids showed moderate insecticidal activity against *Aphis craccivora*.

INTRODUCTION

Croton lacciferus Linn. is a medicinally important plant commonly found in Sri Lanka and South India. In addition to an antifungal benzoquinone, four kauranoid alcohols have been previously reported from this plant [1]. This paper describes the structure elucidation as well as the examination of insecticidal properties of two oleanane derivatives and three kauranoids isolated from the root extracts of *C. lacciferus*.

RESULTS AND DISCUSSION

The hot petrol extracts of the roots of *C. lacciferus* showed significant insecticidal activity against *Aphis craccivora* Koch. The extract when chromatographed over silica gel and eluted with petrol–chloroform gave 16α-H-*ent*-kauran-17-oic acid (**1**), *ent*-15β,16-epoxykauran-17-ol (**10**) [2], 3β-acetoxy-D-friedoolean-14-en-28-oic acid (**11**) [3], oleanolic acid [4] and *ent*-kaur-15-en-3β,17-diol (**8**).

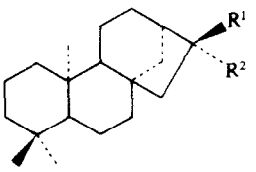
It appeared from spectral data and its molecular formula, $C_{20}H_{32}O_2$, that **1** was a kauranoid containing a CO_2H group. The lithium aluminium hydride reduction product of **1** and the hydrogenated (H_2,Pd/C, EtOH) product of **7** [1, 2] had the same R_f on TLC. The ^{1}H NMR spectra of the reduction products from both reactions were superimposable except that the hydrogenated product showed an additional doublet at δ3.70 corresponding to a hydroxymethylene group. The two types of CH_2OH groups in the 1H NMR spectrum (δ3.38, 3.70) of the hydrogenated product arise from the epimers **2** and **3**; the integration of the peaks at δ3.38 and 3.70 accounted for a 1:1 mixture of the epimers indicating lack of stereospecificity in the hydrogenation. With the sequential addition of Eu(fod)$_3$, both signals at δ3.38 and 3.70 shifted to lower fields at similar rates. However, the peaks at δ3.38 and 3.70 were assigned to the $-CH_2OH$ groups of **2** and **3**, respectively, by comparison of the ^{1}H NMR spectral data [2] of the epimers **4** and **5**. Thus **2** corresponds to the lithium aluminium hydride reduction product of **1**. The optical rotation of the ester prepared by methylating **1** with diazomethane showed a wavelength dependence similar to that of **6** which had been obtained from a methylated column fraction of the extracts from *Baccharis minutiflora* (Compositae) [5]. The assignment of structure was further supported by the ^{13}C NMR (APT) spectrum of **1** (Table 1). The compound **1** had been prepared from 16α-H-*ent*-kauran-17,19-dioic acid previously [6].

The ^{1}H NMR spectrum of **8** was similar to that of **9** [1] except that **8** lacked acetyl protons at δ2.05 and the hydroxymethine proton appeared at a higher field (δ3.20) than that of **9** (δ4.47). Physical data of **8** were identical to those of the hydrolysis product of **9**. The ^{13}C NMR (APT) data of **8** fitted well with the assigned structure (Table 1). The monoacetate (**9**) had been isolated from the chloroform extract of the roots of *C. lacciferus* [1]. The possibility that the diol (**8**) was an artefact of **9** in the extraction/purification procedure was eliminated on the following grounds; **9** survived heating in petrol (bp 60–80°) under reflux conditions for several days and remained unchanged during repeated TLC on silica gel. The hydrolysis of **9** to obtain **8** required rather strong alkaline conditions: 5% KOH in MeOH at room temperature for 24 hr [1].

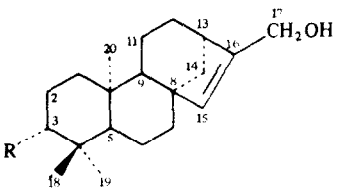
The presence of diastereotopic protons of a hydroxymethylene group in **10** was inferred from the AB quartet (J = 12 Hz) at δ4.07 and 3.75. This quartet and the singlet at δ2.92 which were present in the ^{1}H NMR spectrum of the epoxidation product of **9** [1] could be assigned to H-17 and H-15, respectively. However, the ^{1}H NMR spectrum of **10** lacked a signal corresponding to the H-3 hydroxymethine proton. The physical properties of **10** were identical to those of the epoxy product obtained from the reaction of **7** with *m*-CPBA. The oxirane **10** had been previously isolated from the bulbs of *Fritillaria thunbergii* (Liliaceae) [2].

Compound **11** appeared to be a triterpenoid (Liebermann–Burchard test, M^+ 498) having an olefinic hydrogen (δ5.53), a carboxy group (3600–2700, 1690 cm^{-1}) and a secondary acetocy group (1735 cm^{-1}, δ4.50 for 1H). The physical data of **11** and its hydrolysis (5% KOH–MeOH) product (**12**) were identical to those reported for the same compound and its derivative from *C. oblongifolius* [3]. Oleanolic acid was identified from its physical data [4, 7] and by comparison with an authentic sample.

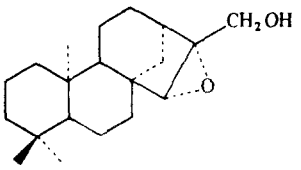
Compounds **1**, **8**, **10**, **11** and oleanolic acid were tested, using the microapplicator method [8], for insecticidal activity against *Aphis craccivora* which were maintained in the laboratory on one-week-old potted cowpea, *Vigna unguiculata* L. (Bandara, K. A. N. P., *et al.*, unpublished



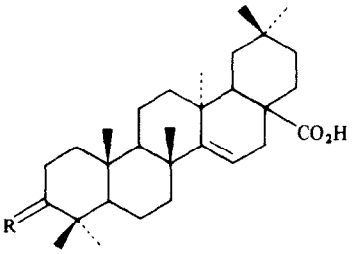
	R^1	R^2
1	H	CO_2H
2	H	CH_2OH
3	CH_2OH	H
4	CH_2OH	OH
5	OH	CH_2OH
6	H	CO_2Me



7 R = H
8 R = OH
9 R = OAc



10



11 R = H, β-OAc
12 R = H, β-OH

results). The kauranoids **8** and **10** at a dose of 5 ppm/insect caused 61% and 62% mortality, respectively, of adult female aphids after 24 hr.

EXPERIMENTAL

The dried roots (4.5 kg) of *C. lacciferus* collected near Peradeniya, Sri Lanka were extracted with hot petroleum (bp 60–80°). The usual work-up gave a brown semisolid (67 g). Silica gel column of the extract (30 g) eluted with petrol–$CHCl_3$ gave: **1** (67 mg) mp 215–217°, lit. [6] 217–219°; $[\alpha]_D$ −65.7° ($CHCl_3$; *c*0.7); IR ν_{max}^{KBr} cm^{-1}: 3420–2260, 2920, 1690, 1450, 1415, 1380, 1365, 1300, 1235, 950; ^1H NMR (60 MHz, $CDCl_3$); δ 10.13 (1H, *br s*, D_2O exchangeable), 2.65 (1H, *m*), 2.1–1.1 (21H, *m*), 1.0, 0.85, 0.8 (3H each, *s*, H-18, H-19 and H-20); ^{13}C NMR see Table 1; EIMS 70 eV *m/z* (rel. int.): 304 $[M]^+$ (32%), 289 $[M-Me]^+$ (100%), 248 (12), 231 (15). Found M_r (MS) 304.2400. $C_{20}H_{32}O_2$ requires: 304.2402; **10** (29 mg), mp 158–159°, lit. [2] 160°. The identity of **10** was confirmed by comparison of physical properties (mp, mmp, Co-TLC, IR, ^1H NMR) with those of a synthetic sample prepared from the reaction of **7** [1] with *m*-CPBA in CH_2Cl_2 at 0° for 15 min; **11** (103 mg), mp 302–204°, lit. [3] 304–305°; $[\alpha]_D$ +21.9° ($CHCl_3$; *c*0.73); the treatment of 25 mg of **11** with 5% KOH in MeOH at room temp. for 24 hr gave 20 mg **12**, mp 299–301°, lit. [3] 300–301°; $[\alpha]_D$ +14.3° ($CHCl_3$; *c*0.28); oleanolic acid (38 mg), mp 305–307°, lit. [7] 306–308°; **8** (50 mg), mp 174–175°; $[\alpha]_D$ −22.7° ($CHCl_3$; *c*0.44); IR ν_{max}^{KBr} cm^{-1}: 3540–3080, 1680; ^1H NMR ($CDCl_3$) δ 5.35 (1H, *s*, H-15), 4.16 (2H, *br s*, H-17), 3.20 (1H, *dd*, J = 10, 6 Hz, H-3), 2.55 (1H, *m*), 2.3–1.1 (18H, *m*), 1.06, 0.97, 0.8 (3H each, *s*, H-18, H-19 and H-20); ^{13}C NMR see Table 1; MS *m/z* (rel. int.): 304 $[M]^+$ (28%), 289 $[M-Me]^+$ (18), 286 $[M-H_2O]^+$ (28), 271 (38), 179 (44), 110 (100). Found M_r (MS) 304.2404. $C_{20}H_{32}O_2$ requires: 304.2402.

Table 1. ^{13}C NMR spectral data of **1** and **8** (75.5 MHz, $CDCl_3$, TMS as internal standard)

C	1	8
1	42.2	43.8
2	19.6	38.7
3	43.3	78.8
4	34.5	38.8
5	42.7	48.7
6	22.0	19.0
7	39.4	38.7
8	46.0	48.2
9	46.7	54.8
10	40.5	39.1
11	19.9	17.7
12	32.6	25.6
13	57.3	41.1
14	41.7	39.1
15	46.4	135.6
16	57.4	146.2
17	184.2	61.3
18	34.9	28.2
19	22.9	18.7
20	18.7	17.6

To a solution of 15 mg **1** in Et_2O, 20 mg $LiAlH_4$ was added portionwise, stirred at room temp. for 6 days and heated under reflux for 6 hr. Usual work-up gave **2** (12 mg), mp 130–131°; $[\alpha]_D$ −46.5° ($CHCl_3$; *c*0.5); IR ν_{max}^{KBr} cm^{-1}: 3300–3100; ^1H NMR

($CDCl_3$): δ3.72 (s, D_2O exchangeable), 3.38 (2H, *d*, $J = 8$ Hz, H-17), 2.4–1.1 (22H, *m*), 1.0, 0.83, 0.8 (3H each, *s*, H-18, H-19 and H-20); MS *m/z* (rel. int.): 290 $[M]^+$ (25%), 275 $[M - Me]^+$ (27), 257 $[M - Me - H_2O]^+$ (3), 231 (9), 123 (57), 109 (24), 94 (100). Found M_r (MS) 290.2610. $C_{20}H_{34}O$ requires 290.2610. A solution of 15 mg **1** in Et_2O was treated with excess CH_2N_2 and usual work-up gave **4** (12 mg), mp 72–74°; IR ν_{max}^{KBr} cm^{-1}: 1715; ^1H NMR ($CDCl_3$): δ3.60 (3H, *m*, CO_2Me), 2.57 (1H, *m*), 2.1–1.1 (21 H, *m*), 1.0, 0.86, 0.81 (3H each, *s*, H-18, H-19 and H-20)

$$[\alpha]_{23.5°}^{\lambda} = \frac{589 \quad 578 \quad 546 \quad 436}{-63 \quad -66 \quad -76 \quad -129} (CHCl_3;\ c0.38)$$

15 mg **7** [1, 2] in 2 ml EtOH was hydrogenated in the presence of Pd/C at room temp. for 1 hr, and usual work-up gave a mixture (12 mg) of **2** and **3**; ^1H NMR ($CDCl_3$): δ3.70 (1H, *d*, $J = 8$ Hz, H-17 of **3**), 3.38 (1H, d, $J = 8$ Hz, H-17 of **2**), 2.2–1.1 (22H, *m*), 1.0, 0.83, 0.8 (3H each, *s*, H-18, H-19 and H-20); MS *m/z* (rel. int.): 290 $[M]^+$ (56%), 275 (44), 257 (7), 231 (13), 123 (100), 109 (34), 94 (38). Found M_r (MS) 290.2611. A 0.2 M solution of Eu $(fod)_3$ in $CDCl_3$ was added to this mixture portion-wise (5 μl) and observed the chemical shifts of H-17.

0.2 M Eu $(fod)_3$: μl	0	5	10	15	20	25
H-17 of **3**: δ	3.70	4.28	5.17	6.17	7.10	8.03
H-17 of **2**: δ	3.38	3.92	4.77	5.72	6.60	7.57

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