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TERPENOIDS OF *PLEUROZIA ACINOSA*

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Key Word Index—*Pleurozia acinosa*; Jungermanniaceae; liverwort; terpenoids; (+)-aristol-9-ene; (−)-3,14-clerodadien-13-ol; 14-labdene-8,13-diol.

Abstract—The major terpenoid constituent of the liverwort, *Pleurozia acinosa*, was a diterpene, (−)-3,14-clerodadien-13-ol. Two other minor components were also isolated and identified as (+)-aristol-9-ene and 14-labdene-8,13-diol.

INTRODUCTION

Liverworts (Hepaticae) are known to be a rich source of terpenoids and/or lipophilic aromatic compounds. The genus *Pleurozia* belonging to the Jungermanniales is a rather small one with three species known in Taiwan. The only published report on the chemistry of this genus was about *P. purpurea* [1] in which the growth inhibiting hormone of liverworts, lunularic acid (**1**), was detected by TLC and GC after derivatization. Another *Pleurozia* sp. from Scotland has also been studied, but only fatty acids were found as the main constituents (Harrison, L. J., personal communication).

P. acinosa grows on tree logs in the swampy area of Yuenyang Lake, Ilan, a damp natural reserve of mainly red cypress forests. A GC/MS examination of the hexane extract of this species revealed a simple constitution of two major diterpenes which were later determined as (−)-3,14-clerodandien-13-ol (kolavelool) (**2**) and 14-labdene-8,13-diol (**3**). The present paper reports the spectral identification of these two components, along with another minor sesquiterpene, (+)-aristol-9-ene[(+)-α-ferulene] (**4**).

RESULTS AND DISCUSSION

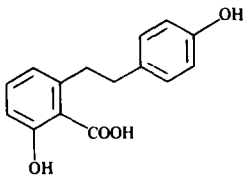
Column chromatography on silica gel of the *n*-hexane extract of the liverwort material resulted in the isolation of a pure sesquiterpene hydrocarbon from the second fraction. Its ^{1}H NMR displayed a cyclopropane ring with two protons on the ring (δ0.60 *d* and 0.75 *dd*), three singlet methyls (δ1.03, 1.05 and 1.08), one secondary methyl (δ0.94 *d*) and one trisubstituted double bond (δ5.09 *br s*). The above information together with the GC/MS data ($[M]^+$ *m/z* 204) suggested a possible structure of either aristol-9-ene (**4**) or aristol-1(10)-ene for this compound. Since these two isomers could be clearly distinguished by their ^{1}H NMR data and optical rotations, as reported in the literature [2], the isolated sesquiterpene was then identified as (+)-aristol-9-ene (**4**) after comparison with published data [2, 3].

Previously, (+)-aristolone (**5**) has been isolated from two Japanese liverworts *Porella caespitans* ssp. *setigera* and *P. fauriei* as a minor component [4].

The second pure component eluted by 5% ethyl acetate in hexane was the major component of the extract. It turned out to be a diterpene alcohol with a clerodane skeleton having the typical vinyl group at the carbinol carbon judging from the ^{1}H NMR (ABX pattern at δ5.06, 5.20 and 5.88; five methyls at δ0.72 *s*, 0.77 *d*, 0.98 *s*, 1.27 *s* and 1.58 *d* and one vinyl proton at 5.18 *br s*) and ^{13}C NMR (four sp^2 C's at δ145.2, 144.5, 120.5 and 111.8, one carbinol C, 73.5 and five methyls 19.9, 18.5, 18.2, 18.0 and 15.9). The ^{1}H NMR data agreed well with those of kolavelool (**2**) isolated from the higher plants *Hardwickia pinnata* [5] and *Solidago elongata* [6]. In the mass spectra of compounds in this series, a major peak of *m/z* 189 or 191 resulting from cleavage at the C9–C11 bond has always been recorded [6] and our component showed a base peak at *m/z* 189 and a second large peak at 191(72%). In a private communication with L. J. Harrison, we learned that the same compound had also been isolated from a Scottish liverwort species *Jungermannia paroica* (*Plectocolea paroica*) [7]. Since the rotational data from both species were consistent, the major component from *Pleurozia acinosa* could then be assigned as (−)-3,14-clerodadien-13-ol [(−)-kolavelool] (**2**).

The second largest peak (*ca* 30% of the major in area) in the GC of the *n*-hexane extract was only obtained in minute amounts due to insufficient amounts of plant material. However, its ^{1}H NMR, together with the ^{13}C NMR, still disclosed enough information to be recognized as a labdenediol (five singlet methyls at δ0.82, 0.86, 0.95, 1.12 and 1.30 and two carbinol carbons at δ73.6 and 73.4) with a terminal vinyl group again (ABX pattern). The isolated component was shown not to be the known stereoisomers, sclareol (**6**), 13-*epi*-sclareol, 8-*epi*-ent-sclareol or 8,9-di-*epi*-ent-sclareol after comparison of their spectral data (^{1}H NMR, ^{13}C NMR or mass spectrometry) [8–11]. Since this compound ought to have the same skeleton as sclareol (**6**) based on its ^{1}H NMR data, it is most likely the unknown 8,13-di-*epi*-sclareol (**7**) upon close comparison of their ^{13}C NMR data (Table 1). In spite of the stereochemistry, the structure of this minor diterpene could, evidently, be assigned as 14-labdene-8,13-diol (**3**). Although quite a number of labdane-type diterpenes have been identified in liverworts, none of the stereoisomers of sclareol (**6**) has ever been reported as a liverwort constituent.

In addition to the isolated components, one monoterpene and several sesquiterpenes were identified by GC/MS from *P. acinosa* [12]. They are: β-pinene, α-, β-



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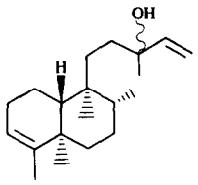

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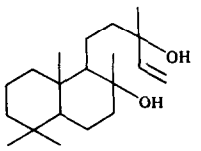

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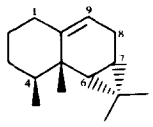

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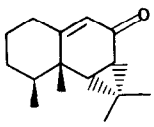

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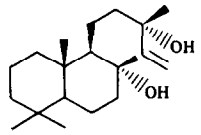

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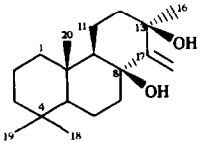

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Table 1. ^{13}C NMR data of sclareol, 13-*epi*-sclareol and the labdenediol from *P. acinosa*

C	Sclareol	(6)	13-*epi*-sclareol	3 from *P. acinosa*
14	146.7	146.2	145.1	144.9
15	111.0	110.3	111.9	112.0
8	74.9	74.3	74.9	73.6
13	73.6	73.0	74.1	73.4
9	61.8	61.4	62.1	59.0
5	56.1	55.9	56.1	56.0
12	45.1	44.8	45.1	46.1
7	44.0	43.9	44.2	42.2
3	42.1	41.9	42.1	42.1
1	39.7	39.5	39.7	39.2
10	39.3	39.0	39.3	39.1
18	33.5	33.3	33.5	33.4
4	33.3	33.1	33.3	33.2
16	26.4	26.2	29.5	30.6
17	24.1	24.0	24.4	27.6
19	21.5	21.4	21.5	21.7
6	20.5	20.4	20.5	19.3
2	19.0	18.8	19.0	18.3
11	18.4	18.4	18.4	18.2
20	15.4	15.3	15.4	15.1
ref.	[9]	[8]	[9]	Present work

and δ-elemene, bicyclogermacrene, β-chamigrene and elemol.

EXPERIMENTAL

TLC, GC and GC/MS were carried out as previously reported [13]. The solvents used for spectral determination were TMS-$CDCl_3$ for ^{1}H NMR (400 MHz) and ^{13}C NMR (100 MHz) and $CHCl_3$ for IR and $[\alpha]_D$.

Plant material. P. acinosa (Mitt.) Schiffn. collected at Yuen-yang Lake, Ilan, of Taiwan in June 1985 and identified by Dr M.-J. Lai, Dept. of Landscape Architecture, Tunghai University, was deposited in the Herbarium of the Institute of Pharmacognosy, Tokushima Bunri University.

Extraction and isolation. Plant material (13 g) was chopped in a blender and extracted with *n*-hexane twice. The hexane extract (0.25 g) was chromatographed on silica gel using a *n*-hexane–EtOAc gradient to give **4** (5 mg), **2** (26 mg) and **3** (12 mg); their structures were determined by spectral methods.

(+)-*Aristol-9-ene* (**4**). $[\alpha]_D$ +45° (*c* 0.31) (lit. [2] +80.9°, neat); IR ν_{max} cm^{-1}: 2950, 2852, 1458, 1370; ^{1}H NMR: δ0.60 (1H, *d*, *J* = 9 Hz, H-6), 0.75 (1H, *d*, *J* = 6 and 9 Hz, H-7), 0.94 (3H, *d*, *J* = 6.8 Hz, C-4 Me), 1.03, 1.05, 1.08 (each 3H, *s*, Me's), 1.2–1.7 (5H, *m*), 1.98 (1H, *br d*, *J* = 12.0 Hz, H-1), 2.03 (1H, *ddd*, *J* = 18.8, 4, 2 Hz, H-8), 2.18 (1H, *br t*, *J* = 12.0 Hz, H-1); 2.3 (1H, *dqn*, *J* = 18.8, 3, 4 Hz, H-8), 5.09 (1H, *br s*, H-9). Allylic-H assignments were based on double irradiation expts. ^{13}C NMR: δ141.0 (*s*, C-10), 118.2 (*d*, C-9), 37.7 (*d*, C-4), 36.5 (*s*, C-5), 32.9 (*t*, C-2), 32.0 (*d*, C-6), 31.3 (*t*, C-3), 30.0 (*d*, C-7), 27.0 (*t*, C-2), 21.6 (*t*, C-8), 21.3 (*q*), 19.2 (*q*), 17.9 (*s*, C_{11}), 16.0 (*q*), 15.8 (*q*). The carbon assignments were based on published data assigned for aristol-9-en-3-one and aristol-9-en-3-ol [4]. MS *m/z* (rel. int.): 204 ([M]$^+$, 45), 189 (30), 161 (40), 147 (30), 133 (40), 119 (40), 105 (75), 91 (100).

(−)-3,14-*Clerodadien*-13-*ol* (**2**). $[\alpha]_D$ −25.7° (*c* 0.7) (lit. [7] −40°); IR ν_{max} cm^{-1}: 3595, 2950, 1480, 1373, 1364, 988, 912; ^{1}H-NMR: δ0.72(*s*, Me), 0.77 (*d*, *J* = 6.1 Hz, Me), 0.98 (*s*, Me), 1.27 (*s*, Me), 1.58 (*d*, *J* = 1.5 Hz, Me), 5.18 (*br s*, 1H), 5.06 (*dd*, *J* = 10.8 Hz and ~1.5, 1H), 5.20 (*dd*, *J* = 17.34 and ~1.5 Hz, 1HO) and 5.88 (*dd*, *J* = 17.34 and 10.75 Hz, 1H); ^{13}C NMR: δ145.2 (*s*), 144.5 (*s*), 120.5 (*d*), 111.8 (*t*), 73.5 (*s*), 46.3 (*d*), 38.4 (*s*), 38.2 (*s*), 36.8 (*t*), 36.1 (*d*), 35.3 (*t*), 31.8 (*t*), 27.8 (*q*), 27.5 (*t*), 26.9 (*t*), 19.9 (*q*), 18.5 (*q*), 18.2 (*t*), 18.0 (*q*), 15.9 (*q*). GC/MS *m/z* (rel. int.): 272 ([M − 18]$^+$, 15), 257 (40), 191 (72), 190 (48), 189 (100), 175 (40), 121 (57), 107 (72), 95 (75).

14-*Labden*-8,13-*diol* (**3**). IR: ν_{max} cm^{-1}: 3600, 3460, 2940, 2855, 1460, 1380, 1366, 986, 915; ^{1}H NMR: δ0.82, 0.86, 0.95, 1.12, and 1.30 (each *s*, Me's × 5), 5.07 (*dd*, *J* = 10.7 Hz, 0.98), 5.23 (*dd*, *J* = 17.3, 1.2 Hz), 5.93 (*dd*, *J* = 17.3, 10.7 Hz); ^{13}C NMR (Table 1); GC/MS *m/z* (rel. int): 177 (100), 95 (65), 109 (54), 71 (50) and 81 (46).

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REFERENCES

1. Gorham, J. (1977) *Phytochemistry* **16**, 249.
2. Weinheimer, A. J., Washecheck, P. H., van der Helm, D. and Hossain, M. B. (1968) *Chem. Commun.* 1070.
3. Bowden, B. F., Coll, J. C. and Mitchell, S. J. (1980) *Aust. J. Chem.* **33**, 681.
4. Asakawa, Y., Yamamura, A., Waki, T. and Takemoto, T. (1980) *Phytochemistry* **19**, 603.
5. Misra, R., Pandey, R. C. and Dev, S. (1979) *Tetrahedron* **35**, 985.
6. Anthonsen, T. and McCrindle, R. (1969) *Acta Chem. Scan.* **23**, 1068.
7. Harrison, L. J. (1983) PhD Thesis, Univ. of Glasgow, Scotland.
8. Almqvist, S.-O., Enzell, C. R. and Wehrli, F. W. (1975) *Acta Chem. Scand.* **B29**, 695.
9. Forster, P. G., Ghisalberti, E. L. and Jefferies, P. R. (1985) *Phytochemistry* **24**, 2991.
10. Bohlmann, F. and Ziesche, J. (1980) *Phytochemistry* **19**, 71.
11. Fukuzawa, A., Miyamoto, M., Kumagai, Y., Abiko, A., Takaya, Y. and Masamune, T. (1985) *Chem. Letters* 1259.
12. Wu, C.-L. (1985) *Proc. 2nd ROK-ROC Symp. on Nat. Prod. Sci.* 17.
13. Asakawa, Y., Toyota, M. and Harrison, L. J. (1986) *Phytochemistry* **24**, 1505.

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5-DEHYDROORICIOPSIN, A RING-D CLEAVED TETRANORTRITERPENOID FROM *HARRISONIA ABYSSINICA*

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Key Word Index—*Harrisonia abyssinica*; Simaroubaceae; tetranortriterpenoid; furylketone limonoid; 5-dehydrooriciopsin.

Abstract—A new ring-D cleaved tetranortriterpenoid, 5-dehydrooriciopsin, has been isolated from the root-bark of Guinean samples of *Harrisonia abyssinica*. Its structure was established by spectral methods.

INTRODUCTION

Five limonoids have been isolated from the African shrub *Harrisonia abyssinica* Oliv. (Simaroubaceae). In addition to the common obacunone, harrisonin, acetoxyharrisonin and pedonin were isolated from Kenyan samples [1–3], whereas atalantolide was found in Nigerian samples [4]. The first four limonoids have been shown to exhibit insect antifeeding activity [2, 5, 6]. In the course of our study on the biologically active constituents of *H. abyssinica*, we report here the isolation of a new furylketone limonoid.

RESULT AND DISCUSSION

Isolation of the limonoids was performed by extraction of the root-bark with diethyl ether and submitting the extract to successive CC and prep. TLC on silica gel. This procedure afforded two limonoids, obacunone and **1**. Obacunone was identified by its spectral data [1, 4, 7, 8]. It was found to be the major limonoid in the samples under investigation (0.30%).

Although the purity of compound **1** was checked in several TLC systems, attempts to crystallize it were unsuccessful. Its EIMS exhibited a $[M]^+$ ion peak at *m/z* 482 corresponding to $C_{27}H_{30}O_8$ as confirmed by HRMS. Other spectral features of **1** such as a signal in the ^1H NMR spectrum for H-21 at δ 8.61 highly indicative of a 17-ketone showed clearly its relationship with obacunone [1], pedonin, **2** [3] and oriciopsin [9]. In comparison with this last limonoid, the conjugation of the C-7 carbonyl was supported in the ^{13}C NMR spectrum by the signal at δ 196.50 or δ 196.40 (C-7), by the signals attributed to C-5 and C-6 respectively at δ 166.35 and 124.62 and in the ^1H NMR by the proton singlet at δ5.73 corresponding to the vinylic proton H-6. Other ^1H NMR and ^{13}C NMR assignments of **1** were based on comparison of the spectral data of obacunone, **2** and oriciopsin [1, 3, 9] and confirmed by DEPT measurements; it was noticed that the attribution of the C-4, C-9 and C-11 signals of oriopsin [9] was not clearly defined. From the above data, **1** was identified as 5-dehydrooriciopsin. As this new limonoid was directly detectable by TLC of the crude $CHCl_3$ extract of the plant, it could not be considered as an artefact. **1** is the second ring-D cleaved tetranortriterpenoid isolated from *H. abyssinica*; the first one, **2**, exhibits a spirotetranortriterpenoid skeleton and the structure of **1** is more similar to that of oriociopsin