

TERPENOIDS FROM *LAURENCIA KARLAE*

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Key Word Index—*Laurencia karlae*; Zhang et Xia; Rhodomelaceae; diterpene; laukarlaol.

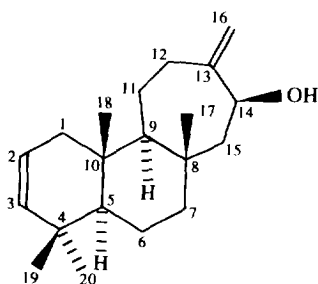
Abstract—A novel diterpene, laukarlaol, was isolated from the red alga *Laurencia karlae* Zhang et Xia, collected from the Nansha islands in the South China Sea. Its structure and relative stereochemistry was established by means of spectral data analysis. Complete ^1H NMR data and assignment of the ^{13}C NMR spectra are reported. Five known sesquiterpenoids were also isolated and identified as aplysistatin, palisadin A, palisadin B, 5-acetoxypalisadin B, and 12-hydroxypalisadin B.

INTRODUCTION

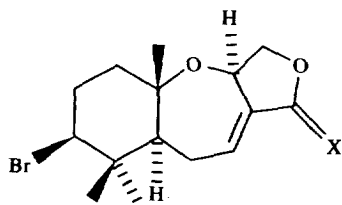
The red alga genus *Laurencia* is chemically characterized by its production of terpenes and C_{15} acetogenins, which are usually halogenated [1–3]. A new species of red alga *Laurencia karlae*, identified by Zhang et Xia (family Rhodomelaceae), was collected from the Nansha Islands in the South China Sea. From this species a novel labdane-derived diterpenoid, laukarlaol (1), and five known sesquiterpenoids (2–6) were obtained.

RESULTS AND DISCUSSION

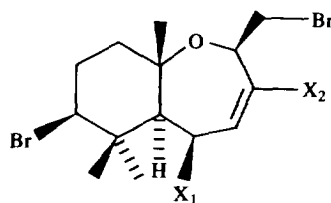
Laukarlaol (1) was assigned the molecular formula $\text{C}_{20}\text{H}_{32}\text{O}$ by HREI-mass spectrometry. The ^{13}C NMR spectrum exhibited 20 carbon resonances including four methyl, seven methylene, five methine, and four quaternary carbons. These data also showed the presence of two olefinic double bonds, one being a terminal methylene unit (δ_{H} 5.03, *br s*; 4.90, *br s*; δ_{C} 155.0, *s*; 111.0, *t*, the other *cis*-disubstituted δ_{H} 5.40, *ddd*, $J = 10.2, 5.4, 1.4$ Hz;



1



2 X=O

3 X=H₂4 X₁=H X₂=Me5 X₁=OAc X₂=Me6 X₁=H X₂=CH₂OH

5.37, *dd*, $J = 10.2, 2.3$ Hz; $\delta_{\text{C}} 121.2$, *d*, 138.0, *d*) (Table 1). The presence of a hydroxyl group was indicated by an O–H stretch in the IR spectrum (3416 cm^{-1}) and by a broad signal in the ^1H NMR spectrum ($\delta 2.17$, 1H), which was exchangeable. The oxygen-bearing carbon ($\delta_{\text{H}} 4.29$, *dd*, $J = 10.7, 4.2$ Hz; $\delta_{\text{C}} 71.0$, *d*) showed this group to be present as a secondary alcohol function. Therefore, according to its molecular formula, **1** must be tricyclic.

The ^1H – ^1H COSY spectral examination of **1** allowed us to establish the proton connections of the following four segments: C-1/C-3, C-5/C-6/C-7, C-9/C-12, and C-14/C-15. The assignment of carbon and proton resonances were achieved by ^{13}C – ^1H COSY (HMQC) spectroscopy of **1**. Furthermore, interpretation of the long-range ^{13}C – ^1H COSY spectrum (HMBC) linked these five segments together through the quaternary carbons C-4 ($\delta 34.9$), C-8 ($\delta 36.0$), C-10 ($\delta 59.4$), and C-13 ($\delta 155.0$). C-1 and C-9 were linked to H-18 (three-bonds), and C-10 correlated with H-18 by a two-bond interaction, which placed the Me-18 at the C-10 position. The connectivity of Me-20 with C-4 was confirmed by cross-peaks ob-

served between H-20/C-3, C-4, and C-5. The observed long-range correlations are summarized in Table 1.

The relative stereochemistry of **1** was determined by a 2D NOESY spectrum. NOE cross-peaks were observed between Me-18/Me-19 and Me-17, between H-5/Me-20 and H-9, and between H-9 and H-14. Therefore, laukarlaol is **1**.

Besides the novel compound **1**, five previously reported sesquiterpenes, namely aplystatin [3, 6], palisadin A [3], palisadin B [3], 5-acetoxypalisadin B [3], and 12-hydroxypalisadin B [3, 7], were isolated and characterized. It is noteworthy that **4** [3] and **5** [3] were obtained as crystalline materials for the first time. The ^1H and ^{13}C NMR data for **2**–**6** were consistent with those in the literature.

EXPERIMENTAL

General. Mp: uncorr. MS: MAT-731 or Finnigan MAT mass spectrometer. NMR spectra: Bruker AMX-

Table 1. NMR spectral data of laukarlaol (**1**)*

Position	δ_{H}	δ_{C}	HMBC (^1H) ($J = 8$ Hz)
1	2.05 1H, <i>dd</i> ($J = 16.7, 5.4$ Hz) 1.59 1H, <i>br d</i> ($J \approx 16.7$ Hz)	40.8 <i>t</i> †	H-18
2	5.40 1H, <i>ddd</i> ($J = 10.2, 5.4, 1.4$ Hz)	121.2 <i>d</i>	H-1
3	5.37 1H, <i>dd</i> ($J = 10.2, 2.3$ Hz)	138.0 <i>d</i>	H-19, H-20
4	—	34.9 <i>s</i>	H-19, H-20, H-5
5	1.15 1H, <i>dd</i> ($J = 9.1, 8.4$ Hz)	52.3 <i>d</i>	H-18, H-20, H-19
6	1.47 2H, <i>m</i>	19.4 <i>t</i>	
7	1.56 1H, <i>m</i> 1.26 1H, <i>m</i>	25.0 <i>t</i>	
8	—	36.0 <i>s</i>	H-6, H-15a, H-15b, H-17
9	0.94 1H, <i>m</i>	59.4 <i>d</i>	H-15a, H-17, H-18
10	—	37.5 <i>s</i>	H-18, H-5
11	1.80 1H, <i>m</i> 1.33 1H, <i>m</i>	44.3 <i>t</i>	
12	2.47 1H, <i>m</i> 2.21 1H, <i>td</i> ($J = 11.9, 2.7$ Hz)	33.2 <i>t</i>	
13	—	155.0 <i>s</i>	H-12a, H-15b
14	4.29 1H, <i>dd</i> ($J = 10.7, 4.2$ Hz)	71.0 <i>d</i>	
15	1.70 1H, <i>dd</i> ($J = 13.6, 4.2$ Hz) 1.44 1H, <i>m</i>	54.7 <i>t</i>	
16	5.03 1H, <i>br s</i> 4.90 1H, <i>br s</i>	111.0 <i>t</i>	
17	0.96 3H, <i>s</i>	19.4 <i>q</i>	
18	0.82 3H, <i>s</i>	16.1 <i>q</i>	
19	0.89 3H, <i>s</i>	22.7 <i>q</i>	
20	0.95 3H, <i>s</i>	31.8 <i>q</i>	
OH	2.17 1H, <i>br s</i> exch	—	

*Spectra were recorded at room temperature in CDCl_3 solution using a proton observation frequency of 600 MHz and a carbon observation frequency of 150 MHz.

†Multiplicity of ^{13}C signals established by DEPT.

600 (600 MHz for ^1H and 150 MHz for ^{13}C) spectrometer in CDCl_3 , using TMS as the int. standard.

Biological material. The red alga *L. karlae* was collected from the Nansha Islands in May 1990. A voucher specimen (90-8#) is deposited in the Research Centre of Organic Natural Products, Zhongshan University, Guangzhou, China.

Extraction and isolation. The sun-dried specimens (0.95 kg), were exhaustively extracted with EtOH at room temp. The concd extract was partitioned between EtOAc and H_2O . The EtOAc-soluble fraction (34.0 g) was subjected to vacuum liquid chromatography over silica gel H with solvent mixtures of increasing polarity from petrol to EtOAc. The fraction eluted with 7% EtOAc-petrol was chromatographed again with 5% Et_2O -petrol and then further purified by prep. TLC on silica gel with 10% Et_2O -petrol to give **1** (10 mg, 0.0011%), and **4** (56 mg, 0.0050%). Fraction 3, eluted with petrol, was further subjected to flash chromatography on silica gel H with 10% Et_2O -petrol to give **4** (505 mg, 0.053%). Fraction 2, eluted with 15% EtOAc-petrol was rechromatographed over silica gel eluting with Et_2O -petrol (1:5), and then was subjected to prep. TLC on silica gel with CH_2Cl_2 -EtOAc (30:1) to give **5** (238 mg, 0.030%), and **6** (253 mg, 0.027%). Fraction 2 eluted with 25% EtOAc-petrol was recrystallized from Me_2CO to give **2** (350 mg, 0.037%).

Laukarlaol (1). Needles, mp 128–129°C, $[\alpha]_D^{20} - 52.0$ (CHCl_3 ; c 0.025) IR $\nu_{\text{max}}^{\text{KBr}}$ cm^{-1} : 3416, 2955, 2928 2849, 1639, 1618, 1461, 1379, 1103, 873; ^1H , ^{13}C NMR: see Table 1. MS m/z (rel. int.): $[\text{M}]^+$ 288.2438 (57), (calc. for $\text{C}_{20}\text{H}_{32}\text{O}$ 288.2445), 273 (13), 270 (24), 255 (42), 245 (12), 188 (39), 175 (25), 173 (26), 159 (28), 145 (38) 134 (100), 119 (98), 107 (88), 105 (72), 95 (56), 91 (48), 81 (49), 79 (37), 67 (36), 55 (37).

Aplysistatin (2). Crystalline solid, mp 160–161°C, $[\alpha]_D - 29.0^\circ$ (MeOH; c 0.030).

Palisadin A (3). Oil, $[\alpha]_D 0 + 20.1^\circ$ (CHCl_3 ; c 0.036).

Palisadin B (4). Hexagons, mp 94–95°C (Me_2CO -petrol), $[\alpha]_D + 10.0^\circ$ (CHCl_3 ; c 0.28).

5-Acetoxypalisadin B (5). Solid, mp 58–59°C (CH_2Cl_2 -petrol), $[\alpha]_D - 133.5^\circ$ (CHCl_3 ; c 0.23).

12-Hydroxypalisadin B (6). Oil, $[\alpha]_D 20.5^\circ$ (CHCl_3 ; c 0.039).

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REFERENCES

1. Faulkner, D. J. (1984) *Nat. Prod. Rep.* **1**, 551.
2. Erickson, K. L. (1985) in *Marine Natural Products* (Scheuer, P. J., ed.) Vol. 5, pp. 131 Academic Press, New York.
3. Paul, V. J. and Fenical, W. (1980) *Tetrahedron Letters* 2787.
4. Fukuzawa, A., Miyamoto, M., Kumagai, Y., Abiko, A., Takaya, Y. and Masamune, T. (1985) *Chem. Letters* 1259.
5. Capon, R. Ghisalberti, E. L. Jefferies, P. R., Skelton, B. W. and White, A. H. (1981) *Tetrahedron* **37**, 1613.
6. Pettit, G. R., Herald, C. L., Allen, M. S., Dreele, R. B. V. Vanell, L. D., Kao, J. P. Y. and Blake, W. (1977) *J. Am. Chem. Soc.* **99**, 262.
7. de Nys, R., Wright, A. D., Konig, G. M., Sticher, O. and Alino, P. M. (1993) *J. Nat. Prod.* **56**, 877.