4.60 (1H, *s*, $J = 10.2$ Hz, C-4-CH_2OH), 5.0 (1H, *d*, $J = 7.0$ Hz, C-1'-H of methyl glucuronate unit), 5.25 (1H, *br s*, C-12-H).

As with BuOH extract (30.2 g) of *M. disperum* which was chromatographed separately over VLC on TLC (silica gel), (350 g) oleanolic acid (0.02 g) (**5**) and hederagenin (0.015 g) (**1**) were isolated and identified on the basis of their IR, ^{1}H NMR, ^{13}C NMR and mass spectra.

Isolation of MD-A (**6**). The $CHCl_3$–MeOH–H_2O (13:4–7:2) eluates afforded a colourless amorphous powder MD-A (100 mg) mp 244–246° (Me_2CO_3), R_f 0.42 (a) $[\alpha]_D$ +43.9 (MeOH; *c* 1). IR ν_{max}^{KBr} cm^{-1}, 3400, 2940, 1735, 1630, 1460, 1380, 1050, ^{13}C NMR: see Table 1.

Alkaline hydrolysis of saponin MD-A (**6**). Saponin MD-A (10 mg) was treated with 5% methanolic KOH and refluxed on a H_2O bath for 3 hr. After usual work-up, the soln was checked on PC which showed the presence of oleanolic acid and D-glucose.

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E-DIHYDRORHODOPHYTIN, A C_{15} ACETOGENIN FROM THE RED ALGA *LAURENCIA PINNATIFIDA*

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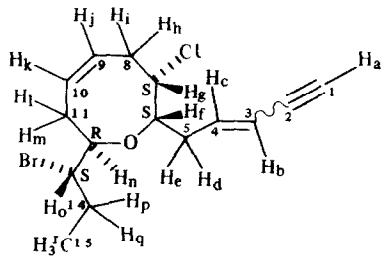
Key Word Index—*Laurencia pinnatifida*; Rhodomelaceae; Rhodophyta; marine natural product; acetogenin; dihydrorhodophytin.

Abstract—From the red alga *Laurencia pinnatifida* we have isolated *E* and *Z* dihydrorhodophytin. The structure and absolute configuration of the former have been established by spectroscopical and chemical methods.

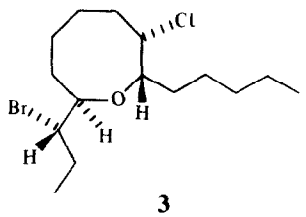
INTRODUCTION

Red alga of the genus *Laurencia* are known to produce C-15 acetogenins containing haloethers of different sizes [1]. In continuation of our studies on this type of compound we have reexamined an extract of *L. pinnatifida* collected at Callao Salvaje (Tenerife, Canary Islands). The isolation of *E*- and *Z*-dihydrorhodophytin was accomplished by combined silica gel (2% *n*-hexane–EtOAc), medium pressure and Sephadex LH-20 (*n*-hexane–$CHCl_3$–MeOH, 2:1:1) chromatography. The *Z* isomer **2** was identified by comparison of its physical and spectral properties with those reported [2]. This compound and the previously unpublished *E* isomer were chemically correlated as follows. Catalytic hydrogenation of each isomer over PtO_2 yielded the same compound, decahydrorhodophytin **3**. The ^{1}H and ^{13}C NMR chemical shift assignments of *E*-and *Z*-dihydrorhodophytin, which have not been published, have been obtained using two dimensional NMR experiments, COSY (^{1}H–^{1}H) and COSY (HETCOR) ^{1}H–^{13}C. (Table 1).

It is noteworthy that *E*-and *Z*-dihydrorhodophytin co-occur with *E*-and *Z*-pinnatifidenine in *L. pinnatifida* [3], in view of the biogenesis of the C_{15} halogenated cyclic ethers. The (6*R*, 7*R*)-3*Z*, 9*Z*, 12*Z*-6-acetoxy-7-chloropentadeca-3,9,12-trien-1-yne, isolated from the same alga [3], might be an intermediate in the biosynthetic pathway to a *Z*-pinnatifidenine. For dihydrorhodophytin the intermediate must be the (6*S*, 7*S*) isomer which has not been found previously.



1 E - isomer
2 Z - isomer



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EXPERIMENTAL

Collection, extraction and chromatographic separation. L. pinnatifida was collected in March 1986 in the intertidal zone at Callao Salvaje (Tenerife, Canary Islands). A voucher specimen is deposited in the Departamento de Biologia Vegetal (Universidad de La Laguna, Tenerife). The alga was dried and ground in a Wiley mill to 1 nm particle size. The dried alga (6 kg) was extracted with Et_2O (3 l) and EtOAc (3 l). The combined extracts were evapd and the crude extract chromatographed on a silica gel column using *n*-hexane–EtOAc mixts of increasing polarity. The *n*-hexane–EtOAc (19 : 1) eluate yielded a yellow oil which was chromatographed on a Sephadex LH-20 column using *n*-hexane–$CHCl_3$–MeOH (2 : 1 : 1) as eluent, yielding colourless crystals containing a mixture of *E*- and *Z*-dihydrorhodophytin **1** and **2** (100 mg). Medium pressure silica gel chromatography of this mixt (*n*-hexane–EtOAc, 49 : 1) gave 40 mg of pure *Z*-dihydrohodophytin and 29 mg of pure *E*-dihydrorhodophytin.

E-Dihydrorhodophytin **1**. Solid, mp 40–41°; $[\alpha] = +33.19°$ (*c*, 0.95, $CHCl_3$); UV $\lambda_{max} = 224$ nm ($\varepsilon = 16\,400$); IR ν^{CHCl_3} 3280, 3020, 2950, 2920, 2100, 1720, 1450, 1375, 1230, 1080 and 960 cm^{-1}. ^1H NMR and ^{13}C NMR in Table 1. HRMS $C_{15}H_{20}O\ ^{81}Br\ ^{35}Cl$ observed 332.0363, $\Delta = 0.2$, $[M]^+$ at *m/z* (rel. int.): 334, 332, 330 (2); 297, 295 (2), 269, 267, 265 (10).

Z-Dihydrorhodophytin **2**. Solid, mp 35–36°, $[\alpha] = +63.52°$ (*c*, 2.44, $CHCl_3$). The physical and spectroscopical data (TLC, GC, IR, NMR, MS) were identical with those reported previously [2].

Catalytic hydrogenation. Ca 10–20 mg of each compound to be hydrogenated was dissolved in 3 ml of dry Et_2O and added to a 10 ml Erlenmeyer suction flask containing a catalytic amount of PtO_2 and a magnetic stirring bar. The reaction vessel was fitted with a balloon and septum, purged with H_2 and the balloon filled. After stirring at 25°, H_2 was removed, the soln filtered and the Et_2O evapd to give, after silica gel chromatography, a purified reaction compound.

Table 1. NMR data for *E*- and *Z*-dihydrorhodophytin **1** and **2***†

		E-Dihydrorhodophytin			*Z*-Dihydrorhodophytin		
C	Proton(s) at carbon n°	δ	Multiplicity *J*(Hz)	C (ppm)	δ	Multiplicity *J*(Hz)	C (ppm)
1	H_1	2.90	*d*, $J_{1-3} = 2.5$	82.5	3.18	*d*, $J_{1-3} = 2.4$	82.92
2				77.13	—	—	80.50
3	H_3	5.65	*dd*, $J_{3-1} = 2.5$; $J_{3-4} = 16$	112.65	5.60	*dd*, $J_{3-1} = 2.4$; $J_{3-4} = 10.5$	111.62
4	H_4	6.23	*dt*, $J_{4-3} = 16$; $J_{4-5} = J_{4-5'} = 7.8$	141.24	6.05	*ddd*, $J_{4-3} = 10.5$; $J_{4-5}\ J_{4-5'} = 7.6$	140.53
5	H_5						
	$H_{5'}$	2.43	*m*	38.6	2.72	*m*	35.50
6	H_6		*ddd*, $J_{6-5} = J_{6-5'} = 7.3$; $J_{6-7} = 1$.	73.49	4.23	*ddd*, $J_{6-5} = J_{6-5'} = 7$; $J_{6-7} = 1$	73.56
7	H_7	4.10	*m*	64.24	4.09	*m*	64.46
8	H_8						
	$H_{8'}$	2.65	*m*	33.72	2.64	*m*	33.78
9	H_9	5.87	*ddd*, $J_{9-8} = 1.5$; $J_{9-8'} = 6.5$; $J_{9-10} = 10.4$	130.32	5.84	*ddd*, $J_{9-10} = 10.3$; $J_{9-8} = 1.2$; $J_{9-8'} = 6.5$	130.44
10	H_{10}	5.93	*ddd*, $J_{10-9} = 10.4$; $J_{10-11} = 6.3$; $J_{10-11'} = 0.5$	127.65	5.87	*ddd*, $J_{10-9} = 10.3$; $J_{10-11} = 6.5$; $J_{10-11'} = 0.3$	127.70
11	H_{11}						
	$H_{11'}$	2.58	*m*	30.28	2.58	*m*	30.41
12	H_{12}	3.96	*ddd*, $J = 4.2$; 5.6 and 9.8	79.33	3.92	*ddd*, $J = 3.1$; 7.3 and 10.6	79.65
13	H_{13}	4.13	*m*	61.69	4.11	*m*	60.58
14	H_{14}	2.08	*ddq*, $J_{14-14'} = 14.5$; $J_{14-15} = 7.2$; $J_{14-13} = 3.6$		2.21	*ddq*, $J_{14-14'} = 14.7$; $J_{14-15} = 7.2$; $J_{14-13} = 3.7$	
	$H_{14'}$	1.82	*ddq*, $J_{14'-14} = 14.5$; $J_{14'-15} = 7.2$; $J_{14'-13} = 2$	29.78	1.80	*ddq*, $J_{14'-14} = 14.7$; $J_{14'-15} = 7.2$; $J_{14'-13} = 2$	30.26
15	H_{15}	1.13	*t* $J = 7.2$	12.45	1.10	*t* $J_{15-14} = J_{15-14'} = 7.2$	12.29

* Chemical shifts are reported in ppm relative to TMS

† 2D NMR methods data support the proton and carbon assignments.

Decahydrorhodophytin **3**. (A) From *Z*-dihidrorodophytin **1**. Catalytic hydrogenation of **1** for 30 min gave the decahydro derivative **3** in 95% yield after chromatography (5% *n*-hexane–EtOAc), oil; $[\alpha] = +18°$ ($CHCl_3$; *c* 0.23). IR ν^{CHCl_3} 3000, 2965, 1460, 1340 and 1070 cm^{-1} ^1H NMR ($CDCl_3$) δ: 1.05 (3H, *t*, $J = 7.2$ Hz); 1.67 (*m*, 18H); 4.03 (*m*, 4H). MS: $[M]^+$ at *m/z* (rel. int.): 338, 340, 342 $[M]^+$ (2); 260, 262 $[M - Br]^+$ (7); 217, 219 $[M - C_3H_6Br]^+$ (4).

(B) From *trans*-dihydrorhodophytin **2**. Hydrogenation of **2** for 1 hr in the same manner as described above for **1** gave in 92% yield the octahydro derivative **3**: oil, $[\alpha] = +17.5°$ ($CHCl_3$; *c* 0.11). IR, NMR and MS were as described in (A) above.

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OXEPINE DERIVATIVES AND ANTHRAQUINONES FROM *ASPHODELINE TENUIOR* AND *A. TAURICA*

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Key Word Index—*Asphodeline tenuior*; *A. taurica*; Liliaceae; anthraquinones; eudesmanolide; oxepine derivatives.

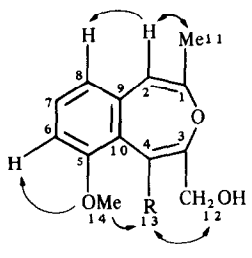
Abstract—From the chloroform extracts of *Asphodeline taurica* and *A. tenuior* anthraquinones chrysophanol, asphodeline and microcarpin as well as β-sitosterol, sitosteryl 3β-glucoside were isolated. In addition, *A. taurica* yielded an eudesmanolide 1β-acetoxy-8β-hydroxy-eudesman-4(15),7(11)-dien-8α,12-olide, while aleoemodin and two new oxepine derivatives, tenual and tenucarb, were obtained from *A. tenuior*.

In a continuation of our chemical studies with *Asphodeline* species [1, 2] we have now investigated *A. taurica* (Pallas) Kunth and *A. tenuior* (Fisher) Ledeb. subsp. *tenuiflora* (C. Koch) E. Tuzlacı var. *puberulenta* E. Tuzlacı. These two species are placed in different sections (Appendicigera E. Tuzlacı and Asphodeline respectively) which were recently described [3]. *A. tenuior* subsp. *tenuiflora* var. *puberulenta* is an endemic taxon in Turkey.

In our previous studies with *A. globifera*, *A. damascena* [1] and *A. anatolica* [2] (Sect. Appendicigera) we have obtained eudesmanolides in addition to anthraquinones. In the present study with *A. taurica* from the same section we have obtained a eudesmanolide 1β-acetoxy-8β-hydroxy-eudesman-4(15),7(11)-8α,12-olide in addition to anthraquinones chrysophanol, asphodeline and microcarpin as well as β-sitosterol and sitosteryl 3β-glucoside, while *A. tenuior* (Sect. Asphodeline) yielded aleoemodin and two new oxepine derivatives tenual and tenucarb in addition to the above compounds with the exception of the eudesmanolide.

The high resolution mass spectrum of tenual (**1**) gave a molecular ion peak at *m/z* 246.08921 indicating a molecular formula $C_{14}H_{14}O_4$. The UV spectrum of **1** at 336, 312, 298, 283, 259, 230 nm indicated a conjugated aromatic system which was correlated with the IR peaks at 3050, 1584, 1555, 1520 cm^{-1}. The presence of hydroxyl (3420 cm^{-1}) and aldehyde (1710 cm^{-1}) group were indicated by the same spectrum. ^1H NMR spectrum showed three adjacent aromatic proton peaks at δ7.38 (1H, *t*, $J = 8$ Hz, H-8), 6.79 (1H, *br d*, $J = 8$ Hz, H-6) and at δ7.12 (1H, *d*, $J = 8$ Hz, H-2). Other peaks showed the presence of an aldehyde group at δ9.96 (1H, s, CHO), a methoxyl group at δ4.09 (3H, *s*, OMe), an aromatic methyl at δ2.42 (3H, *d*, $J = 0.8$ Hz, Me) and a hydroxymethylene group at δ4.77 (2H, *br s*, CH_2OH) and at δ1.57 (1H, *br s*, OH) (D_2O exchange). The fourth oxygen function must be ether; this was correlated with its IR peaks at 1270, 1240, 1068 and 1035 cm^{-1}.

The positions of the functional groups were decided by NOE experiments and by the ^{13}C NMR spectrum. Since the ^1H NMR spectrum of **1** showed three adjacent aro-



R = CHO
R = COOMe[15]