New C Acetogenins and Sesquiterpenes from the Red Alga Laurencia sp. cf. L. gracilis

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NEW C_{15} ACETOGENINS AND SESQUITERPENES FROM THE RED ALGA *LAURENCIA* SP. CF. *L. GRACILIS*

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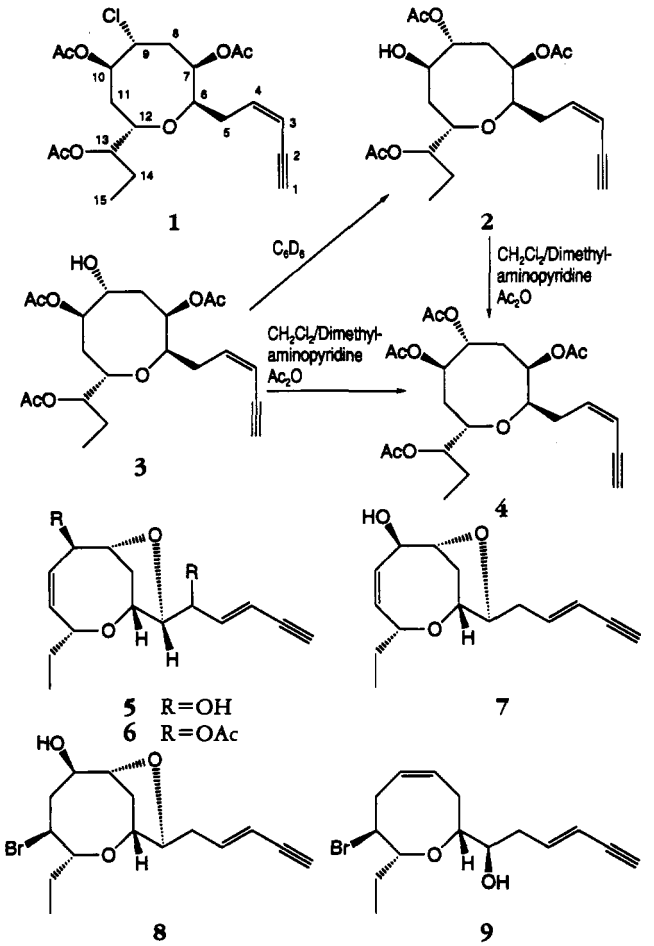
ABSTRACT.—From the red alga *Laurencia* sp. cf. *L. gracilis*, collected from New Zealand waters, five new natural products (**1–3**, **5**, **10**) were isolated and characterized. Together with these compounds, a further eleven metabolites, **7–9** and **11–18**, were isolated. Of these eleven compounds, **7** is reported as a pure substance for the first time, while for all others complete spectroscopic data are provided, where not reported previously. Compound **11** appears to be identical to preintricatol.

The genus *Laurencia* is one of the most intensively chemically investigated of all marine genera. The reasons for this are twofold. First, algae belonging to this genus are extremely widespread, being found in all oceans and seas as well as at almost all latitudes, and second, because plants belonging to this genus, almost without exception, have a high secondary metabolite content. Both of these features make *Laurencia* species attractive sources for new and potentially biologically active novel natural products, especially when plants can be collected from geographic locations where little or no research has been undertaken. In this respect, *Laurencia* species collected from New Zealand waters represent an untouched resource and as such inspired the current investigation into *Laurencia* sp. cf. *L. gracilis* Harvey, sensu V.W. Lindauer, "Algae Novae Zelandicae Exsiccatae," No. 94, collected from Matheson Bay, Leigh, New Zealand.

RESULTS AND DISCUSSION

Sixteen compounds were purified and characterized from the CH_2Cl_2 solubles extracted from the marine red alga *Laurencia* sp. cf. *L. gracilis*.

Compound **1**, a crystalline white solid, had the molecular formula $C_{21}H_{29}ClO_7$ by mass spectrometry and nmr spectroscopy. Of the seven degrees of unsaturation implied by the molecular formula of **1**, six were occupied by either sp^2 or by sp hybridized carbon atoms; **1** was thus a monocyclic molecule containing a single carbon-carbon triple bond, a carbon-carbon double bond and three carbon-oxygen double bonds, as part of three acetoxyl functions. From the ir data of **1** it was evident that this molecule contained no hydroxyl functions, indicating the remaining oxygen functionality to be present in the form of an ether. The results obtained from recording ^1H-^1H and ^1H-^{13}C (J=150 Hz) 2D nmr COSY spectra of **1** enabled a continuous chain of ^1H-^1H coupling to be traced from H-1 through to H_3-15. Thus, H-1 showed coupling to H-3 (J=2.2 Hz), which in turn coupled to H-4. From $J_{3,4}$=10.8 Hz it was evident that these latter two protons were cisoid. In turn, both H-3 and H-4 coupled to the protons at C-5, which both demonstrated coupling to H-6. The dihedral angle between H-6 and H-7 was evidently approaching 90° as the coupling between these two protons was less than 1 Hz. This was also the case for the relationship between H-7 and the more shielded of the two protons at C-8 (δ 1.56). The other proton at C-8 (δ 2.55) showed a 5.1 Hz coupling to H-7 but no coupling to H-9, indicating the dihedral angle between these two to be of the order of 90°. The less shielded of the two protons at C-8 (δ 1.56) did couple to H-9, which in turn coupled to H-10. This latter proton coupled to both protons at C-11, which inter-coupled and both demonstrated coupling to H-12, which coupled to H-13. This proton



further coupled to both protons at C-14, which also inter-coupled and coupled to the protons of the C-15 methyl group, giving rise to the following partial structure:

$$\overset{1}{HC}\equiv\overset{}{C}-\overset{3}{CH}=CH-\overset{5}{CH_2}\underset{X}{\overset{}{CH}}-\overset{7}{\underset{X}{\overset{}{CH}}}-CH_2\cdot\overset{9}{\underset{X}{\overset{}{CH}}}-\overset{11}{\underset{X}{\overset{}{CH}}}-CH_2\cdot\overset{13}{\underset{X}{\overset{}{CH}}}-\overset{15}{\underset{X}{\overset{}{CH}}}-CH_2\cdot CH_3$$

From the results of a long-range ^1H-^{13}C (HMBC, J=8.3 Hz) 2D nmr COSY measurement it was possible to position the three acetoxyl groups at C-7, C-10, and C-13, based on correlations between the carbonyl carbons (169.5, 169.5, and 169.9 ppm) and H-7 (δ 4.76), H-13 (δ 5.01), and H-10 (δ 5.58), respectively. The position of the single ether bridge between C-6 and C-12 was evident from a long-range correlation between H-6 (δ 3.61) and C-12 (72.8 ppm): the single chloro-function is at C-9. With the regiochemistry of **1**, and the stereochemistry of the $\Delta^{3,4}$ double bond established, the relative stereochemistry at six chiral centers required resolution. From the NOESY spectrum of **1**, diagnostic nOe cross-peaks were observed between H-9 and H-12, H-8 (δ 1.56) and H-10, H-6 and H-13, and from H-6 to H-11 (δ 1.71). These data together with the observations that the dihedral angles between H-6 (δ 3.61) and H-7 (δ 4.76), H-7 and H-8 (δ 1.56), and H-8 (δ 2.55) and H-9 (δ 5.21) are approaching 90° indicates **1** to be (3*Z*,6*R**,7*R**,9*R**,10*R**,12*S**,13*R**)-9-chloro-6:12-epoxy-7,10,13-triacetoxypentadec-3-en-1-yne.

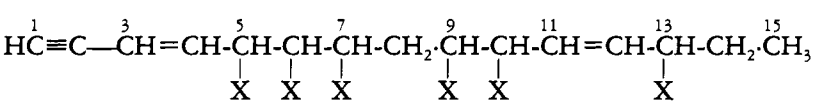
Compound **2** was shown to have the molecular formula $C_{21}H_{30}O_8$ by mass spectrometry. The seven degrees of unsaturation indicated by the molecular formula of **2** were comprised in an identical manner as determined for **1**. It was also evident from the results of ^{1}H-^{1}H and ^{1}H-^{13}C (J=150 Hz) 2D nmr measurements that **1** and **2** were virtually identical molecules, with the differences between the two being due to the presence of a hydroxy function in **2** instead of the chloro-function found in **1**, and also its location. The ^{1}H- and ^{13}C-nmr data (Tables 1 and 2) indicated two of the acetoxyl groups to be located at C-7 and C-13, as in **1**. From these data it was also clear that the third of these groups must reside at C-9 [75.3 (d) ppm, δ 5.74 (br dd, J=8.9 and 9.1 Hz)], leaving the hydroxyl function to be positioned at C-10. From the results of a NOESY measurement made on **2**, it was evident that this molecule was stereochemically identical with **1**. Compound **2** is thus (3*Z*,6*R**,7*R**,9*R**,10*R**,12*S**,13*R**)-6:12-epoxy-10-hydroxy-7,9,13-triacetoxypentadec-3-en-1-yne.

Compound **3** also analyzed for $C_{21}H_{30}O_8$ by mass spectrometry and ^{13}C-nmr spectroscopy. It was found to be a positional isomer of **2**. Close comparison of the ^{1}H- and ^{13}C-nmr data (Tables 1 and 2) of **3** with those of **2** indicated that **3** was substituted with a hydroxyl function at C-9 and an acetoxyl group at C-10. Compound **3** is thus (3*Z*,6*R**,7*R**,9*R**,10*R**,12*S**,13*R**)-6:12-epoxy-9-hydroxy-7,10,13-triacetoxypentadec-3-en-1-yne. Storage of compound **3** in C_6D_6 at −4° resulted in its quantitative conversion to **2**; compound **2** could thus be an artifact.

Acetylation of **2** and **3** yielded in each case the tetra-acetate **4**, confirming the stereochemical deductions made concerning these compounds. The ^{1}H-nmr spectrum of **4**, when recorded in C_6D_6, showed each individual proton resonance separately and thus allowed a clear interpretation of nOe effects in the NOESY spectrum of **4**. This was in contrast to the other three samples which all had some overlapping proton resonances.

Compounds **1–4** are evidently iso-structural with laurencienyne (1), isolated by Caccamese *et al.*, from a Sicilian species of *Laurencia obtusa*. Since the original submission of the present manuscript for publication we have become aware of the recent work by Ojika *et al.* (18). From the data presented in this publication (^{1}H-nmr and optical rotation) it is clear that the semi-synthetic tetra-acetate obtained by Ojika and co-workers is identical to compound **4** and hence the stereochemistry of doliculols A and B is identical to that shown for **4**. Compounds **2** and **3** are the first examples of non-halogenated C_{15} acetogenins, belonging to this class, to be isolated from any *Laurencia* species, with the only other examples being doliculols A and B recently isolated from sea hares and clearly of dietary origin (18).

Compound **5** was found to have the molecular formula $C_{15}H_{20}O_4$ by a combination of mass spectrometry and ^{13}C-nmr spectroscopy. These data also indicated **5** to be a bicyclic molecule containing two carbon-carbon double bonds and a carbon-carbon triple bond. Other functionalites within **5** were determined as two secondary hydroxyl functions [71.3 (d), 4.89 (ddd), 77.8 (d), 5.02 (ddd) ppm] and two ether rings. From the ^{1}H-^{1}H and ^{1}H-^{13}C (J=150 Hz) 2D nmr COSY spectra of **5** a continuous chain of ^{1}H-^{1}H coupling could be traced from the proton on C-1 through to the protons on C-15, giving rise to the following partial structure:



The ^{1}H-^{13}C (HMBC, J=8.3 Hz) 2D nmr COSY spectrum of **5** showed a correlation from C-6 (84.4 ppm) to H-9 (δ 4.26). It was thus apparent that one of the two ether bridges occurred between C-6 and C-9. The second ether bridge, between C-7 and C-

TABLE 1. ^{1}H Nmr Data (300 MHz) for Compounds **1–9**.

Position	Compound								
	1[a]	**2**[a]	**3**[a]	**4**[a]	**5**[b]	**6**[b]	**7**[a]	**8**[a]	**9**[a]
1	3.11 (d, J=2.2 Hz)	3.14 (dd, J=0.8, 2.2 Hz)	3.13 (dd, J=0.8, 2.3 Hz)	3.12 (dd, J=0.7, 2.3 Hz)	2.90 (d, J=2.2 Hz)	2.90 (d, J=2.4 Hz)	2.64 (d, J=2.2 Hz)	2.63 (d, J=2.3 Hz)	2.61 (dd, J=0.6, 2.3 Hz)
3	5.43 (br d, J=10.9 Hz)	5.45 (br d, J=10.8 Hz)	5.45 (dddd, J=1.0, 1.0, 2.3, 10.8 Hz)	5.44 (br d, J=10.8 Hz)	5.86 (ddd, J=2.1, 2.2, 16.1 Hz)	5.77 (ddd, J=1.3, 2.4, 16.1 Hz)	5.60 (dddd, J=1.7, 2.2, 3.8, 15.8 Hz)	5.57 (dddd, J=1.5, 2.3, 3.8, 15.8 Hz)	5.49 (dddd, J=1.6, 1.6, 2.3, 16.0 Hz)
4	5.73 (ddd, J=7.7, 7.7, 10.9 Hz)	5.75 (dddd, J=0.8, 7.4, 7.4, 10.8 Hz)	5.81 (dddd, J=1.0, 7.6, 7.6, 10.8 Hz)	5.72 (br ddd, J=7.5, 7.5, 10.8 Hz)	6.39 (dd, J=4.8, 16.1 Hz)	6.30 (ddd, J=0.7, 6.4, 16.1 Hz)	6.33 (ddd, J=7.4, 7.5, 15.8 Hz)	6.26 (dddd, J=7.3, 7.4, 15.8 Hz)	6.36 (ddd, J=0.6, 7.3, 7.4, 16.0 Hz)
5	2.61 (br dd, J=7.4, 7.7 Hz)	2.67 (m), 2.82 (m)	2.67 (ddd, J=1.0, 7.4, 7.6 Hz), 2.76 (ddd, J=1.0, 7.4, 7.6 Hz)	2.67 (m), 2.81 (ddd, J=7.5, 9.4, 13.8 Hz)	4.89 (ddd, J=2.1, 4.8, 6.7 Hz)	5.57 (m)	2.50 (m)	2.36 (m)	1.97 (m)
6	3.61 (br dd, J=7.2, 7.4 Hz)	3.73 (m)	3.65 (br dd, J=7.4, 7.6 Hz)	3.65 (dd, J=5.2, 9.4 Hz)	3.73 (dd, J=2.8, 6.7 Hz)	3.84 (dd, J=2.6, 9.1 Hz)	3.52 (ddd, J=2.6, 7.1, 7.4 Hz)	3.38 (ddd, J=2.4, 7.1, 7.3 Hz)	3.20 (ddd, J=4.2, 5.3, 7.8 Hz)
7	4.76 (br d, J=5.1 Hz)	4.79 (br d, J=4.8 Hz)	4.90 (br d, J=4.7 Hz)	4.76 (br d, J=5.0 Hz)	4.32 (ddd, J=1.4, 2.5, 2.8 Hz)	4.16 (ddd, J=1.4, 2.5, 2.6 Hz)	3.37 (br s)	3.27 (ddd, J=1.1, 2.3, 2.4 Hz)	2.78 (ddd, J=1.4, 5.3, 10.5 Hz)
8	1.56 (m), 2.55 (dd, J=5.1, 16.6 Hz)	1.24 (ddd, J=2.0, 9.1, 15.7 Hz), 2.20 (dd, J=4.8, 15.7 Hz)	1.38 (ddd, J=2.3, 8.8, 16.1 Hz), 2.15 (dd, J=4.7, 16.1 Hz)	1.27 (ddd, J=2.0, 9.5, 16.0 Hz), 2.21 (dd, J=5.0, 16.0 Hz)	1.77 (ddd, J=2.5, 7.1, 13.7 Hz), 2.70 (dd, J=1.4, 13.7 Hz)	1.81 (ddd, J=2.5, 7.2, 13.7 Hz), 2.73 (dd, J=1.4, 13.7 Hz)	1.27 (ddd, J=2.4, 7.1, 13.3 Hz), 2.32 (dd, J=1.3, 13.3 Hz)	1.16 (ddd, J=2.3, 8.0, 14.2 Hz), 1.88 (br d, J=14.2 Hz)	1.67 (ddd, J=1.4, 8.5, 14.2 Hz), 2.12 (m)
9	5.21 (dd, J=8.8, 9.9 Hz)	5.74 (br dd, J=8.9, 9.1 Hz)	4.78 (dd, J=8.8, 8.8 Hz)	6.02 (dd, J=9.3, 9.5 Hz)	4.26 (dd, J=3.5, 7.1 Hz)	4.35 (dd, J=3.9, 7.2 Hz)	4.25 (dd, J=3.4, 7.1 Hz)	4.09 (dd, J=3.6, 8.0 Hz)	5.67 (dddd, J=1.0, 6.9, 8.2, 10.6 Hz)
10	5.28 (ddd, J=2.3, 4.1, 9.9 Hz)	3.72 (m)	4.94 (ddd, J=2.7, 4.3, 8.8 Hz)	5.27 (ddd, J=2.3, 4.3, 9.3 Hz)	5.02 (ddd, J=2.4, 2.4, 3.5 Hz)	5.88 (m)	5.04 (ddd, J=2.1, 2.2, 3.4 Hz)	3.72 (ddd, J=1.3, 3.6, 9.8 Hz)	5.89 (dddd, J=2.0, 6.3, 9.4, 10.6 Hz)
11	1.71 (m), 1.98 (ddd, J=1.9, 4.1, 16.6 Hz)	1.70 (m), 1.82 (ddd, J=2.5, 3.8, 15.7 Hz)	1.73 (m), 1.98 (ddd, J=2.1, 4.3, 16.5 Hz)	1.73 (m), 1.92 (m)	5.41 (ddd, J=2.4, 3.5, 12.1 Hz)	5.15 (dddd, J=0.6, 2.2, 4.3, 12.1 Hz)	5.17 (ddd, J=2.2, 2.3, 11.9 Hz)	2.04 (m), 2.26 (ddd, J=1.3, 4.7, 14.7 Hz)	2.29 (ddd, J=3.3, 6.3, 14.0 Hz), 2.89 (dddd, J=1.0, 3.7, 9.4, 14.0 Hz)
12	4.07 (ddd, J=1.9, 8.4, 12.2 Hz)	4.33 (dddd, J=0.5, 2.5, 8.6, 11.4 Hz)	4.09 (ddd, J=2.1, 8.3, 12.5 Hz)	4.10 (br ddd, J=2.0, 8.3, 12.4 Hz)	5.50 (ddd, J=2.4, 4.0, 12.1 Hz)	5.56 (m)	5.55 (ddd, J=2.1, 4.3, 11.9 Hz)	3.50 (ddd, J=4.7, 10.4, 10.6 Hz)	3.80 (ddd, J=3.3, 3.7, 10.0 Hz)
13	5.01 (ddd, J=3.8, 8.3, 8.4 Hz)	5.10 (ddd, J=3.6, 8.3, 8.6 Hz)	5.04 (ddd, J=3.8, 8.3, 8.3 Hz)	5.07 (ddd, J=3.5, 8.2, 8.3 Hz)	3.87 (m)	3.81 (m)	3.30 (m)	3.09 (ddd, J=2.7, 8.9, 10.6 Hz)	3.35 (ddd, J=2.7, 6.9, 10.0 Hz)
14	1.54 (m), 1.84 (m)	1.63 (m), 1.96 (m)	1.58 (m), 1.89 (m)	1.58 (m), 1.92 (m)	1.52 (m), 1.60 (m)	1.53 (m)	1.33 (m)	1.32 (m), 2.01 (m)	1.46 (m), 1.84 (m)
15	0.83 (dd, J=7.5, 7.5 Hz)	0.90 (dd, J=7.5, 7.5 Hz)	0.86 (dd, J=7.5, 7.5 Hz)	0.87 (dd, J=7.5, 7.5 Hz)	0.95 (dd, J=7.5, 7.5 Hz)	0.90 (dd, J=7.5, 7.5 Hz)	0.85 (dd, J=7.3, 7.5 Hz)	0.81 (dd, J=7.4, 7.4 Hz)	0.82 (dd, J=7.4, 7.4 Hz)
Acetate	1.69 (s), 1.72 (s), 1.76 (s)	1.57 (s), 1.71 (s), 2.19 (s)	1.65 (s), 1.72 (s), 1.81 (s)			2.05 (s), 2.09 (s)			

[a] In C_6D_6.
[b] In $CDCl_3$.

TABLE 2. ^{13}C-Nmr Data (75.5 MHz) for Compounds **1–9**.

Position	Compound								
	1[a]	**2**[a]	**3**[a]	**4**[a]	**5**[b]	**6**[b]	**7**[a]	**8**[a]	**9**[a]
1	83.8 (d)[c]	83.7 (d)	83.7 (d)	83.7 (d)	78.0 (d)	78.7 (d)	76.8 (d)	76.9 (d)	77.0 (d)
2	80.2 (s)	80.3 (s)	80.3 (s)	80.2 (s)	81.8 (s)	81.4 (s)	82.7 (s)	82.5 (s)	82.4 (s)
3	111.9 (d)	111.7 (d)	111.6 (d)	111.8 (d)	109.6 (d)	112.1 (d)	111.5 (d)	111.5 (d)	111.4 (d)
4	139.7 (d)	140.1 (d)	140.2 (d)	139.9 (d)	144.3 (d)	141.1 (d)	142.7 (d)	142.5 (d)	142.7 (d)
5	34.5 (t)	34.6 (t)	34.7 (t)	34.6 (t)	71.3 (d)	71.3 (d)	34.5 (t)	33.8 (t)	37.1 (t)
6	72.2 (d)	72.2 (d)[d]	72.3 (d)	72.2 (d)	84.4 (d)	83.5 (d)	82.3 (d)	82.2 (d)	73.3 (d)
7	70.9 (d)	71.0 (d)	71.0 (d)	70.7 (d)	79.3 (d)	77.3 (d)	77.7 (d)	76.9 (d)	83.6 (d)
8	38.8 (t)	35.0 (t)	37.3 (t)	35.0 (t)	33.7 (t)	33.4 (t)	34.0 (t)	33.2 (t)	30.4 (t)
9	57.8 (d)	75.3 (d)	67.6 (d)	71.2 (d)	86.5 (d)	82.5 (d)	86.6 (d)	84.3 (d)	129.9 (d)
10	76.5 (d)	72.0 (d)[d]	78.0 (d)	73.5 (d)	77.8 (d)	81.1 (d)	78.7 (d)	78.0 (d)	128.6 (d)
11	28.3 (t)	29.4 (t)	27.6 (t)	27.4 (t)	132.3 (d)	128.8 (d)	133.4 (d)	43.7 (t)	32.5 (t)
12	72.8 (d)	72.0 (d)[d]	72.8 (d)	72.7 (d)	133.1 (d)	134.9 (d)	133.2 (d)	53.2 (d)	56.0 (d)
13	74.4 (d)	74.7 (d)	74.8 (d)	74.4 (d)	80.6 (d)	80.1 (d)	79.8 (d)	80.1 (d)	83.6 (d)
14	25.5 (t)	25.6 (t)	25.4 (t)	25.5 (t)	28.5 (t)	28.3 (t)	28.7 (t)	28.3 (t)	25.9 (t)
15	9.5 (q)	9.6 (q)	9.6 (q)	9.6 (q)	10.0 (q)	9.8 (q)	10.0 (q)	9.6 (q)	9.1 (q)
Acetates ...	20.4 (q)	20.6 (q)	20.5 (q)	20.4 (q)		20.9 (q)			
	20.4 (q)	20.8 (q)	20.6 (q)	20.5 (q)		21.4 (q)			
	20.5 (q)	21.2 (q)	20.7 (q)	20.7 (q)		168.9 (s)			
	169.5 (s)	170.1 (s)	169.7 (s)	21.1 (s)		170.1 (s)			
	169.5 (s)	170.2 (s)	169.9 (s)	169.5 (s)					
	169.9 (s)	170.5 (s)	170.4 (s)	169.7 (s)					
				169.9 (s)					
				170.4 (s)					

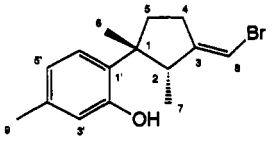
[a]In C_6D_6.
[b]In $CDCl_3$.
[c]Multiplicities by DEPT.
[d]Assignments interchangeable.

13, was positioned by comparing spectroscopic data of **5** with those of **7** and **8**. Thus, the two hydroxyl functions were at C-5 and C-10. The stereochemistry at five out of the six chiral centers was proposed from a NOESY nmr spectrum of **5**, with diagnostic nOes being observed from H-13 to H-8 (δ 2.70) and H-7, and H-10 to H-5. The two carbon-carbon double bonds, $\Delta^{3,4}$ and $\Delta^{11,12}$, were assigned the *E* and *Z* configurations, respectively, based on $J_{3,4}$=16.1 Hz and $J_{11,12}$=12.1 Hz. Compound **5** is thus (3*E*,6*R**,7*S**,9*S**,10*S**,11*Z*,13*S**)-6:9,7:13-diepoxy-5,10-dihydroxypentadec-3,11-dien-1-yne or (3*E*)-5-hydroxydehydrobromolaurefucin.

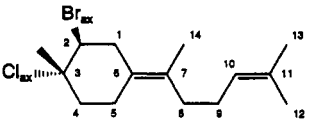
The mass spectral data of **5** were not considered adequate to obtain a suitably accurate mass measurement of any of the diagnostic high mass ions, due to their low intensities. Thus, a portion of **5** was acetylated to yield the diacetate **6**. The mass spectral characteristics of this compound enabled an accurate assessment of the mass of its molecular ion to be made.

Three further C_{15} acetogenins, **7** [(3*E*)-dehydrobromolaurefucin] (2),**8** (laurefucin) (3,4), and **9** (deacetyllaurencin) (5), were isolated and characterized. For **7**, this is its first report as a pure natural compound. Based on the results of a NOESY nmr measurement made with **7**, it was possible to deduce the C-10 hydroxyl function to be β, because of the presence of diagnostic cross-peaks between the resonances for H_2-5 and H-10, as well as between H-8 (δ 2.32) and H-10 and H-11. For compounds **7–9**, complete and unambiguous ^{1}H- and ^{13}C-nmr data are reported for the first time.

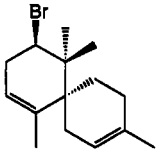
Compound **10** had the molecular formula $C_{15}H_{19}BrO$ by mass spectrometry. Evident in the ^{13}C-nmr spectrum of **10** were resonances for eight sp^2 hybridized, and seven sp^3 hybridized carbon atoms; **10** was thus bicyclic. One of the rings was a 1,2,4 trisubstituted aromatic system, while the other was present as a five-membered ring, as indicated by the ^{1}H-^{1}H COSY data of **10**. Cross-peaks in this latter spectrum were observed between H-2 and H_3-7, H-2 and H-8, H-8 and H_2-4, H_2-4 and H_2-5, and



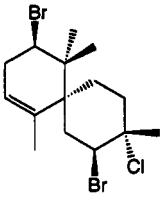
10



11



12



13

between H_α-5 and H_3-6. These data generated a basic structure which was essentially identical to that of laurenisol (6,7). The physical data for **10** were, however, different; **10** was an oil and had an optical rotation of +48.1°, as compared to laurenisol which was crystalline and had an optical rotation of +85.9°. These differences suggested **10** to be a structural isomer of laurenisol, and this deduction was confirmed from the results of a NOESY measurement made with **10**. It was clear from these data that the $\Delta^{3,8}$ double bond had the *E* configuration in **10** in contrast to the *Z* configuration described for laurenisol. Compound **10** is thus the $\Delta^{3,8}$ double-bond isomer of laurenisol and has been named *iso*-laurenisol.

Compound **11**, also a sesquiterpene, analyzed for $C_{15}H_{24}ClBr$ by mass spectrometry. From these data and its ^1H- and ^{13}C-nmr spectra **11** was deduced to be a monocyclic molecule containing a trisubstituted cyclohexane moiety, and an extended isoprene unit, C-12,13 to C-6. When the positions of the chloro-and bromo-functions had been determined it appeared that **11** contained all of the structural features found in preintricatol (8). The previous study describing the structure of preintricatol unfortunately contained a bare minimum of spectroscopic data, and lacked firm evidence for the clearly drawn stereochemistry. No optical rotation for this then new compound was reported, making comparison extremely difficult. As a consequence of being unable to make satisfactory spectroscopic and physical data comparisons between the data for **11** and those for preintricatol, extensive 2D nmr studies of **11** were undertaken. The results of these measurements, particularly NOESY spectra, indicated **11** to have the structure and relative stereochemistry as shown in the structural formula of **11**. Diagnostic nOes were observed from H_3-13 and H_3-14 to H-10, from H_3-14 to H_2-1, and from H_3-15 to H-2. In the case of the latter nOe, this interaction alone was not adequate to propose the relative configuration at C-3, however, these data combined with the ^{13}C-nmr data did permit such a deduction to be made (17). The nOe data also allowed the assignments for H_3-14 and H_3-15 to be made unambiguously. With these deduced stereochemistries, **11** would appear to be identical to preintricatol. The lack of a published optical rotation for this compound (8), however, make this deduction speculative.

Seven known sesquiterpenes with the indicated yields were also isolated from this *Laurencia* species: laurene (13.7 mg, 0.0075%) (9), dihydrolaurene (8.4 mg, 0.0046%) (10), **12** (6.2 mg, 0.0034%) (11), **13** (17.8 mg, 0.0097%) (13), deoxyprepacifenol (80.4 mg, 0.044%) (14), prepacifenol (438.0 mg, 0.24%) (12), and pacifenol (74.0 mg, 0.04%) (15).

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.—Details have been published previously (16).

PLANT MATERIAL.—The algal material was collected in March 1991 from Matheson Bay, Leigh, New Zealand. Plants growing at depths between 1–3 m were collected, deep frozen, and freeze-dried on return to the laboratory. A voucher specimen is deposited at the Museum of New Zealand, Wellington, New Zealand (voucher number WELT A20125).

EXTRACTION AND ISOLATION.—The dry algal tissue (183.6 g) was exhaustively extracted with 2 liters of CH_2Cl_2 and then with 1.5 liter of MeOH to afford 5.82 g (3.17%) of CH_2Cl_2-soluble material. Vacuum-liquid chromatography (vlc) of this material over Si gel, using hexane containing increasing proportions of EtOAc as eluent, afforded 16 fractions each of 90 ml. Tlc and ^{1}H nmr investigation of these fractions indicated fractions 1–5 and 10–13 to be of further interest. Hplc separation of fraction 1, over normal phase silica with hexane as eluent yielded four sesquiterpenes, **11**, laurene, dihydrolaurene, and **12**.

Compound **11**.—This compound was isolated as an oil (4.8 mg, 0.0026%); $[\alpha]^{25}D$ +28.5° ($CHCl_3$, c=0.48); ^{1}H nmr ($CDCl_3$, 300 MHz) δ 1.60 (3H, s, H-12), 1.68 (3H, s, H-13), 1.68 (3H, s, H-14), 1.75 (3H, s, H-15), 1.88 (1H, m, H-4), 2.05 (2H, m, H-8), 2.05 (2H, m, H-9), 2.16 (1H, m, H-4), 2.29 (1H, m, H-5), 2.43 (1H, m, H-5), 2.79 (1H, ddd, J=1.3, 5.2, and 15.0 Hz, H-1), 3.03 (1H, br dd, J=3.4 and 15.0 Hz, H-1), 4.44 (1H, ddd, J=1.6, 3.4, and 5.2 hz, H-2), 5.13 (1H, br t, J=6.7 Hz, H-10) ppm; ^{13}C nmr ($CDCl_3$, 75.5 MHz) 17.6 (q, C-12), 18.2 (q, C-14), 25.6 (t, C-5), 25.8 (q, C-13), 26.9 (t, C-9), 30.9 (q, C-15), 34.4 (t, C-8), 35.9 (t, C-1), 38.1 (t, C-4), 61.7 (d, C-2), 72.7 (s, C-3), 124.1 (d, C-10), 125.3 (s, C-7), 130.3 (s, C-6), 131.7 (s, C-11) ppm; with ir and ms data as previously reported form preintricatol (8).

Hplc separation of combined vlc fractions 2 and 3, over normal phase silica employing hexane containing 2% EtOAc as eluent afforded the two chamigrene derivatives **13** and deoxyprepacifenol.

Hplc separation of vlc fraction 4, over normal phase silica employing hexane containing 6% EtOAc as eluent yielded two further sesquiterpenes, **10** and prepacifenol.

iso-*Laurenisol* [**10**].—Oil (4.2 mg, 0.0023%); $[\alpha]^{25}D$ +48.1° (c=0.42, $CHCl_3$); uv λ max (EtOH) 275 (ε 2560), 281 nm (ε 2420); ir ν max (film) 3400, 2910, 1615, 1415, 805 cm^{-1}; ^{1}H nmr ($CDCl_3$, 300 MHz) δ 0.73 (3H, d, J=7.4 Hz, H-7), 1.22 (3H, br s, H-6), 1.87 (1H, m, H-5), 2.27 (3H, s, H-9), 2.32 (1H, br dd, J=10.6 and 10.8 Hz, H-5), 2.46 (2H, m, H-4), 3.16 (1H, br q, J=7.4 Hz, H-2), 4.61 (1H, s, 2′-*OH*), 6.05 (1H, br s, H-8), 6.50 (1H, br s, H-3′), 6.71 (1H, br d, J=7.8 Hz, H-5′), 7.02 (1H, d, J=7.8 Hz, H-6′) ppm; ^{13}C nmr ($CDCl_3$, 75.5 MHz) 19.0 (q, C-7), 20.7 (q, C-9), 25.7 (q, C-6), 28.7 (t, C-4), 33.6 (t, C-5), 48.9 (d, C-2), 49.6 (s, C-1), 98.3 (d, C-8), 116.5 (d, C-5′), 121.6 (d, C-3′), 128.2 (d, C-6′), 130.6 (s, C-1′), 137.2 (s, C-4′), 152.9 (s, C-2′), 155.0 (s, C-3) ppm; eims m/z [M]$^+$ 296 (7), 294 (7), 281 (6), 279 (6), 215 (100), 173 (10), 159 (15), 121 (40); hrms m/z 294.0558 (calcd for $C_{15}H_{19}{}^{79}Br$, 294.0620).

Hplc separation of vlc fraction 5, over normal phase silica employing hexane containing 7% EtOAc as eluent, yielded pacifenol.

Hplc separation of vlc fraction 6, over normal phase silica employing hexane containing 1.5% MeOH as eluent, yielded a single C_{15} acetogenin, **9**.

Deacetyllaurencin [**9**].—Oil (12.0 mg, 0.0065%); $[\alpha]^{25}D$ +35.5° (c=0.4, $CHCl_3$) cf. +46.1°; ^{1}H and ^{13}C nmr see Tables 1 and 2, respectively; identical uv, ir and ms data to data previously reported (5).

Vlc fractions 10 and 11 were combined and chromatographed over RP-18 material, using MeOH containing 16% H_2O as eluent, to remove chlorophylls. The eluted fraction was then separated by hplc using RP-18 silica and MeOH containing 28% H_2O as eluent, to yield compound **1**.

(3Z,6R,7R*,9R*,10R*,12S*,13R*)-9-Chloro-6:12-epoxy-7,10,13-triacetoxypentadec-3-en-1-yne* [**1**].—A white crystalline solid, mp 126.5–127.5° (3.4 mg, 0.0019%); $[\alpha]^{25}D$ +21.1° (c=0.19, $CHCl_3$); uv λ max (EtOH) 223 (ε 7290), 231 sh nm (ε 5860); ir ν max (film) 2920, 1735, 1730, 1365, 1225 cm^{-1}; ^{1}H and ^{13}C nmr see Tables 1 and 2, respectively; eims m/z [M+1]$^+$ 431 (<1), 429 (<1), 393 (<), 365 (1), 363 (3), 329 (1), 327 (2), 165 (50); hrms m/z 363.1110 (calcd for $C_{16}H_{24}{}^{35}ClO_7$, 363.1211).

Further purification of the eluted fraction, from vlc fractions 10 and 11, by normal-phase hplc with hexane containing 30% Me_2CO as eluent, yielded two further C_{15} acetogenins, **2** and **3**.

(3Z,6R,7R*,9R*,10R*,12S*,13R*)-6:12-Epoxy-10-hydroxy-7,9,13-triacetoxypentadec-3-en-1-yne* [**2**].—A white chrystalline solid, mp 132.0–133.0° (6.9 mg, 0.0038%); $[\alpha]^{25}D$ +45.0° (c=0.04, $CHCl_3$); uv λ max (EtOH) 223 nm (ε 13510); ir ν max (film) 3440, 3230, 2920, 1730, 1715, 1370, 1240 cm^{-1}; ^{1}H and ^{13}C nmr see Tables 1 and 2, respectively; eims m/z [M−H_2O]$^+$ 392 (<1), 345 (2), 285 (4), 249 (3), 225 (3), 183 (7), 165 (15); hrms m/z 345.1491 (calcd for $C_{16}H_{25}O_8$, 345.1549).

(3Z,6R,7R*,9R*,10R*,12S*,13R*)-6:12-Epoxy-9-hydroxy-7,10,13-triacetoxypentadec-3-en-1-yne* [**3**].—Oil (7.2 mg, 0.0039%); $[\alpha]^{25}D$ +21.0° (c=0.3, $CHCl_3$); uv λ max (EtOH) 224 nm (ε 13720); ir ν

max (film) 3440, 3230, 2920, 1735, 1715, 1370, 1240 cm^{-1}; ^1H and ^{13}C nmr see Tables 1 and 2, respectively; eims *m/z* $[M-H_2O]^+$ 392 (1), 345 (3), 285 (6), 249 (3), 225 (3), 183 (7), 165 (20); hrms *m/z* 345.1501 (calcd for $C_{16}H_{25}O_8$, 345.1549).

Conversion of **3** *to* **2**.—Compound **3** (2 mg) dissolved in C_6D_6 and allowed to stand at −4° for 2 weeks converted quantitatively to **2**.

Acetylation of **2** *and* **3**.—A quantity (3 mg) of each of compounds **2** and **3** was dissolved in CH_2Cl_2 (2 ml). To this solution a catalytic amount of DMAP (dimethylaminopyridine, 0.1 mg) was added. The solution was then cooled to 0° and 0.1 ml of Ac_2O added. After 15 min the solution was allowed to come to room temperature. The reaction was then quenched with 2 ml of H_2O and the mixture neutralized with Na_2CO_3. The aqueous portion of this solution was extracted three times with 2 ml of CH_2Cl_2. The combined CH_2Cl_2 solubles were then chromatographed (hplc normal phase with hexane containing 20% Me_2CO as eluent) to yield **4** (2.1 mg and 1.8 mg, respectively).

(3Z,6R,7R*,9R*,10R*,12S*,13R*)-6:12-Epoxy-7,9,10,13-tetraacetoxy-pentadec-3-en-1-yne* [**4**].—An amorphous white powder; $[\alpha]^{25}D$ +40.3° (*c*=0.36, $CHCl_3$); ir ν max (film) 2920, 1735, 1725, 1370, 1250, 1225 cm^{-1}; ^1H and ^{13}C nmr in C_6D_6 see Tables 1 and 2, respectively; ^1H nmr ($CDCl_3$, 300 MHz) identical to previously reported data (18); ^{13}C nmr ($CDCl_3$, 75.5 MHz) 9.6 (q), 21.0 (q), 21.1 (q), 21.2 (q), 21.3 (q), 25.2 (t), 27.0 (t), 34.1 (t), 34.8 (t), 70.5 (d), 70.8 (d), 72.2 (d), 72.6 (d), 73.0 (d), 75.0 (d), 79.8 (s), 83.1 (d), 111.8 (d), 139.1 (d), 169.8 (s), 170.3 (s), 170.6 (s), 171.0 (s) ppm; eims *m/z* $[M]^+$ 452 (<1), 387 (35), 351 (20), 183 (30), 165 (67).

Hplc separation of combined vlc fractions 12 and 13, over RP-18 material and using MeOH containing 16% H_2O as eluent yielded three further C_{15} acetogenins, **5**, **7**, and **8**.

(3E,6R,7S*,9S*,10S*,11Z,13S*)-6:9,7:13-Diepoxy-5,10-dihydroxypentadec-3,11-dien-1-yne=(3E)-5-hydroxydehydrobromolaurefucin* [**5**].—Oil; $[\alpha]^{25}D$ −22.0° (*c*=0.5, $CHCl_3$); uv λ max (EtOH) 222 nm (ε 6360); ir ν max (film) 3400, 3290, 2930, 1720, 1500 cm^{-1}; ^1H and ^{13}C nmr see Tables 1 and 2, respectively; eims *m/z* $[M-H_2O]^+$ 246 (2), 217 (2), 189 (5), 165 (12), 123 (70), 98 (75), 83 (100).

(3E)-Dehydrobromolaurefucin [**7**].—Oil (58.8 mg, 0.32%); $[\alpha]^{25}D$ −26.3° (*c*=0.75, $CHCl_3$); uv λ max (EtOH) 224 nm (ε 9830); ir ν max (film) 3400, 3300, 2910, 1045 cm^{-1}; ^1H and ^{13}C nmr see Tables 1 and 2, respectively; eims *m/z* $[M]^+$ 248 (1), 230 (2), 191 (5), 165 (9), 149 (40), 105 (50), 83 (100); hrms *m/z* 248.1116 (calcd for $C_{15}H_{20}O_3$ 248.1413).

Laurefucin [**8**].—Oil (54 mg, 0.03%); $[\alpha]^{25}D$ −71.0° (*c*=0.5, $CHCl_3$) cf. −80°; ^1H and ^{13}C nmr see Tables 1 and 2, respectively; with all other data as previously reported (3,4).

Acetylation of **5**.—Compound **5** (6 mg) was acetylated in an identical fashion to **2** and **3** to yield 3.8 mg of **6**.

(3E,6R,7S*,9S*,10S*,11Z,13S*)-5,10-Diacetoxy-6:9,7:13-diepoxypentadec-3,11-dien-1-yne* [**6**].—Oil; $[\alpha]^{25}D$ +7.9° (*c*=0.19, $CHCl_3$); ir ν max (film) 2910, 1735, 1365, 1230 cm^{-1}; ^1H and ^{13}C nmr see Tables 1 and 2, respectively; eims *m/z* $[M]^+$ 348 (1), 306 (3), 288 (1), 246 (2), 225 (3), 165 (5), 98 (37), 97 (32); hrms *m/z* 348.1540 (calcd for $C_{19}H_{24}O_6$ 348.1572).

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