Notes

Drimane Sesquiterpenes from the Sponge *Dysidea fusca*

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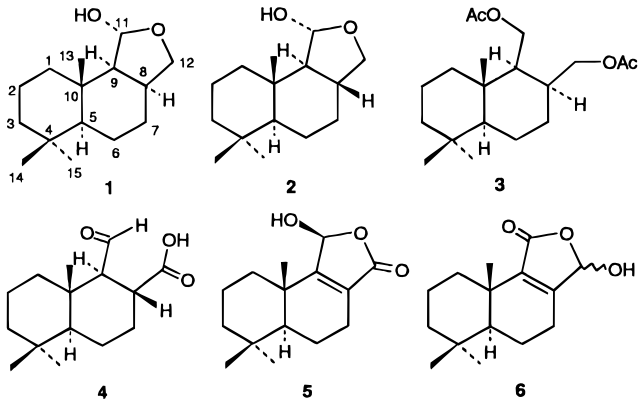
Received March 21, 1996[⊗]

One known drimane sesquiterpene (**1**) and five new ones (**2**−**6**) have been isolated from the sponge *Dysidea fusca*. Their structures were elucidated mainly by 2D NMR. The relative stereochemistry at C-11 of **1** has been corrected to H-11β.

Drimane sesquiterpenes are known as metabolites of plants as well as marine organisms;[1] in the marine environment they have been found especially in sponges of the genus *Dysidea*.[2−4] We report here the isolation of one known drimane (**1**)[2] and five new drimane sesquiterpenes (**2**−**6**) from the sponge *Dysidea fusca* Ridley (family Spongidae) collected in New Caledonia.[5] The structures of the new compounds were determined using 2D NMR.

The sponge was collected off the East Coast of New Caledonia, freeze dried, and extracted with EtOH. The EtOH extract was partitioned between H_2O and CH_2Cl_2. The organic extract was repeatedly chromatogaphed on a Si gel column, yielding compounds **1** and **4**. Compounds **2**, **3**, **5**, and **6** were further purified using reversed-phase HPLC.



The 1D NMR spectra for compound **1** were in agreement with the literature data.[2] However, the stereochemistry suggested for C-11 on the basis of chemical shifts obtained in the presence of the reagent Eu(fod)$_3$ has to be reversed. H-11 is β because in the NOESY experiment the correlations H-11/Me-13 and H-11/H-1β were observed. The normal chair conformation for ring B was secured by the correlation H-12β/Me-13.

Compound **2** gave a MH$^+$ peak in the HRCIMS at *m*/*z* 239.2016 corresponding to the molecular formula $C_{15}H_{26}O_2$ (Δ) −0.5 mmu), which is the same as that of **1**. The ^{1}H-NMR spectrum contained the signals of three methyl groups at δ 0.86, 0.87, and 0.94 (3s); of an oxymethine at δ 5.13 (d, J = 7 Hz); and of a CH_2O group at δ 3.40 (dd, J = 7, 11 Hz) and 3.70 (dd, J = 7, 7 Hz). The ^{13}C-NMR spectrum showed the characteristic peaks of Me-13, Me-14, and Me-15 of the drimane skeleton at δ 15.6, 21.8, and 34.1, respectively. The highfield shift of the oxymethine carbon (δ 99.2) indicated that this carbon bore two oxygens, whereas the oxymethylene appeared at δ 71.4. These data suggested that compound **2** was a diastereoisomer of **1.** The structure of **2** was further supported by COSY, HMQC, HMBC, and NOESY experiments (Table 1). The relative stereochemistry involving a trans BC junction and a H-11β configuration followed from the diagnostic NOE correlations H-9/H-5, H-8/Me-13, H-9/H12α, and H-11/Me-13. The NOESY spectrum was recorded in pyridine-d_5, inasmuch as H-5 and Me-13 were superimposed in Me_2CO-d_6 and $CDCl_3$.

The molecular formula for compound **3** ($C_{19}H_{32}O_4$) was established by accurate mass measurement of the MH$^+$ peak in the HRCIMS (*m*/*z* 325.2384, Δ 0.5 mmu). The IR spectrum showed the absorption band of an ester group at 1728 cm^{-1}. The ^{1}H-NMR spectrum exhibited five methyl singlets: two MeCO groups at δ 2.00 and 2.01 and three aliphatic methyls. The latter gave typical drimane resonances in the ^{13}C NMR (δ 16.5, 21.5, and 33.5). In addition, the ^{13}C spectrum showed two oxymethylene groups at δ 62.7 and 64.0, while the MeCO groups resonated at δ 21.3, 21.5, and 171.2. This suggested the structure depicted in **3**, which was confirmed by 2D experiments (Table 1). The NOESY spectrum indicated that H-8 and H-9 were cis, because a cross peak H-8/H-9 was observed. The correlations shown in Table 1 indicated further that both protons were α. The dialdehyde corresponding to **3** has been isolated recently from another *Dysidea* sponge.[2]

Compound **4** exhibited a MH$^+$ peak in the HRCIMS at *m*/*z* 253.1816, which matched the molecular formula of $C_{15}H_{26}O_4$ (Δ 1.2 mmu). The IR spectrum exhibited a carbonyl band at 1715 cm^{-1}. An aldehyde singlet was observed in the ^{1}H-NMR spectrum at δ 9.87 when the spectrum was run at −55 °C ($CDCl_3$); at room temperature, the signal was considerably broadened, probably due to hydrogen bonding with a nearby oxygen atom. The ^{13}C-NMR spectrum showed the aldehyde resonance

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[⊗] Abstract published in *Advance ACS Abstracts,* July 15, 1996.

S0163-3864(96)00373-4 CCC: $12.00

Table 1. ^{13}C NMR (75 MHz) and ^{1}H NMR[a] Data for Compounds **2**−**4**[b]

	2[c]				**3**[d]				**4**[d]			
position	δ_C	δ_H (*J* Hz)	HMBC	NOESY	δ_C	δ (*J* Hz)	HMBC	NOESY	δ_C	δ_H (*J* Hz)[e]	HMBC[e]	NOESY
1	40.0	α 1.20 m		1*β*	39.2	α 1.03 ddd (4,13,13)	2,5,9,10	1*β*,3α,5	40.2	α 1.56 m	3,5,10,13	1*β*,5,9
		β 1.62 m		2α*β*,11,13		*β* 1.58 m	3,5			*β* 1.88 br d (10)		13
2	19.3	α 1.40 m	1,3,10	2*β*	18.4	α 1.35 m		2*β*	19.0	α 1.40 m		
		β 1.66 m	1,3	13,14		*β* 1.55 m		13		*β* 1.58 m		
3	43.2	α 1.20 m		3*β*,5,15	41.8	α 1.12 m	1,14,15	6*β*, 15	42.5	α 1.35 m	5,14,15	3*β*, 5
		β 1.40 m			14,15		*β* 1.32 m	1,5		*β* 1.15 m		
4	33.9				33.2				34.0			
5	56.6	0.95 dd (12,2.5)		9	56.2	0.88dd (12,2.5)	4,6,7,10, 13,14,15	9	54.9	1.03 br d (11)	9,10,13,14,15	6α,9,15
6	22.3	α 1.68 m	5,7,8,10	6*β*,7α*β*,15	17.6	α 1.55 m	5,7	6*β*, 15	21.8	α 1.65 m		6*β*,7*β*,15
		β 1.30 m	5,7,8,10	7*β*,14		*β* 1.30 m	5,7			*β* 1.25		8,13
7	29.0	α 1.10 m	5,6,8,9,12	7*β*, 12α	29.1	α 1.40 m		7*β*	30.2	α 1.25 m		7*β*
		β 1.85 m	5,6,8,9			*β* 1.85 m	5,9			*β* 2.10 br d (12)	5	8
8	40.4	1.96 m	9,12	11,12α*β*,13	34.9	2.17 m		9	40.2	2.68 br t (8)	7,9,12	13
9	66.3	1.24 m	7,8,10,11,13	12α	51.5	1.65 m	1,8,10,11, 12,13	11ab	64.4	2.50 d (11)	5,7,11,12,13	
10	35.5				37.2				38.9			
11	99.2	5.13 d (7)	9,10,12	13	62.7	a 4.25dd (5,11)	8,9,10,CO	11b	210.9	9.84 s	8,9	
						b 4.05dd (9,11)	8,9,10,CO					
12	71.4	α 3.70 dd (7,7)	7,8		64.0	4.10 m	7,8,CO	13	182.9			
		β 3.40 dd (7,11)	8,9,11									
13	15.6	0.94 s	1,5,9,10		16.5	0.80 s	1,5,9,10		16.6	0.78 s	1,5,9,10	
14	21.8	0.87 s	3,4,5,15		21.5	0.78 s	3,4,5,15		22.2	0.72 s	3,4,5,15	
15	34.1	0.86 s	3,4,5,14		33.5	0.86 s	3,4,5,14		34.2	0.80 s	3,4,5,14	
*Me*CO					21.3	2.00 s	CO					
					21.5	2.01 s	CO					
CO					171.2							

[a] 400 MHz, except NOESY of compound **2**, which was 600 MHz. [b] Assignments are based on 2D experiments. [c] In acetone-d_6 except NOESY in pyridine-d_5. [d] In $CDCl_3$. [e] At −55 °C.

Table 2. ^{13}C NMR (75 MHz) and ^{1}H NMR (400 MHz) Data[a] for Compounds **5** and **6**

	5				6			
position	δ_C[b]	δ_H (*J* Hz)[b]	HMBC[b]	NOESY[c]	δ_C[b]	δ_H (*J* Hz)[c,d]	HMBC[c,e]	NOESY[c,d]
1	35.4	α 1.35 m		1*β*, 11	34.0	α 1.17 m	9	1*β*
		β 1.92 m		11		*β* 2.45 m	2, 5, 9	
2	18.3	α 1.54 m		2*β*	17.9	α 1.52 m		2*β*
		β 1.70 m				*β* 1.74 m		13, 14
3	41.7	α 1.19 m	1, 2, 4, 14	3*β*, 15	41.3	α 1.20 m		3*β*, 15
		β 1.48 m				*β* 1.52 m		14, 15
4	33.5				32.7			
5	51.5	1.15 d (13)	4,7,8,10,13,14		51.6	1.22 m	1,6,7,9,13,14,15	
6	18.0	α 1.86 m	4, 5, 7, 10	6*β*, 15	17.5	α 1.93 m	4, 5, 7, 8, 10	5, 6*β*, 15
		β 1.53 m	4, 5, 7, 10	7*β*		*β* 1.52 m	4, 5, 7, 10	13
7	21.5	α 2.12 m	6, 8, 9	7*β*	23.7	α 2.43 [2.48] m	8, 9	7*β*
		β 2.36 dd (18,6)	5, 6, 8, 9,12,13			*β* 2.25 [2.30] m	8, 9	
8	128.3				157.9			
9	167.7				138.3			
10	37.0				34.5			
11	99.1	6.08 s	8, 12		169.2			
12	172.0				96.2	5.844 [5.838] s	9, 11	
13	20.3	1.21 s	1, 5, 9, 10		19.7	1.14 [1.12] s	1, 5, 9, 10	
14	21.6	0.89 s	3, 4, 5, 15		20.9	0.92 s	3, 4, 5, 15	
15	33.5	0.91 s	3, 4, 5, 14		33.0	0.96 s	3, 4, 5, 14	

[a] Assignments based on 2D experiments. [b] In $CDCl_3$. [c] In CD_4O. [d] At −65 °C. [e] At −50 °C. δ_H values in brackets are for the one or other C_{11}-epimer of **6**.

at δ 210.9 and the signal of a carbonyl at δ 182.9, suggesting a carboxylic acid function. The latter was further confirmed by the yellow spot observed on TLC with bromocresol green. The remaining signals of the ^{13}C-NMR spectrum were consistent with a drimane skeleton. The HMBC correlations from C-9 to both H-5 and the aldehyde singlet, together with the correlation from H-8 to both C-7 and the acidic carbonyl, showed that the aldehyde and the carboxyl groups were located at C-11 and C-12, respectively. The relative stereochemistry as indicated by the NOESY experiment (Table 2) was similar to that of **2**, showing the typical drimane AB trans junction, whereas H-8 and H-9 were trans. The latter configuration was established from the cross peaks observed between H-9 and H-5 and between H-8 and H-13.

Compound **5** exhibited a MH^+ peak in the HRCIMS at *m*/*z* 251.1652 corresponding to the molecular formula of $C_{15}H_{22}O_3$ (Δ −0.5 mmu). The IR spectrum showed the absorptions of a conjugated carbonyl at 1762 and 1670 cm^{-1}. In the ^{13}C-NMR spectrum, the carbonyl group appeared at δ 172.0. The conjugated double bond

was tetrasubstituted and resonated at δ 128.3 and 167.7. In addition, a methine bearing two oxygen atoms was observed at δ_C 99.1 (δ_H 6.08, s). The other signals and the 2D spectra (Table 2) were in accordance with the structure depicted in **5.** In particular, the HMBC correlation between H-7 and the signal of the CO at 172.0 allowed location of the latter carbonyl group at C-12. Finally, the H-11α configuration was deduced from the NOESY relationships H-11/H-1α and H-11/H-1β. The NOESY spectrum was obtained in CD_3OD, because intermolecular association in $CDCl_3$ prevented spectral interpretation.

Compound **6** was an isomer of **5**, showing the same molecular ion peak at *m/z* 251 in the CIMS. Similar IR bands (1756 and 1670 cm^{-1}) and NMR spectra (Table 2) indicated the presence of a conjugated carbonyl and a dioxymethine. The HMBC spectrum (Table 2) displayed correlations from H-5 and H-1 to the olefinic carbon α to the carbonyl at δ 138.3, indicating that this carbon was located at C-9. Hence, the carbonyl was at position 11 and the dioxymethine at position 12. The C-11 position of the CO group was further supported by the downfield shift of H-1β, which was placed inside the shielding zone of the carbonyl. Finally, a NOESY experiment at −50 °C (in CD_3OD) confirmed the location of OH-12 by the correlation H-12/H-7$\alpha\beta$. At room temperature, H-7 appeared as a broad unresolved signal, and the latter correlation was absent. At a lower temperature (−65 °C) a doubling of the H-12, H-7, and Me-13 signals was observed, showing that compound **6** was a mixture of epimers at the hemiacetal carbon.

The absolute stereochemistry of **1** has been established previously as depicted. It is likely that compounds **2**−**6** possess the same stereochemistry, because they all probably derive from the same biosynthetic precursor, (+)-euryfuran.[2]

Experimental Section

General Experimental Procedures. Optical rotations at 20° were taken on a Perkin-Elmer 241 polarimeter. UV spectra were recorded in MeOH on a Shimadzu UV-161 UV-vis spectrophotometer; IR, on a Nicolet 205 FT-IR spectrometer; CIMS, on a Kratos MS 9; HRCIMS, on a Kratos MS 80; and NMR, on Bruker AC 250, AC 300, AM 400, or AM 600 spectrometers. The HMBC spectra were obtained using a INVDR2LP in Bruker program with an evolution delay for CH long-range coupling of 70 ms. The NOESY spectra (phase sensitive) were recorded using the NOESYPH Bruker program with a mixing time of 0.6 s. Column chromatography was performed using Si gel Merck H60. Preparative HPLC was achieved using Waters Delta Prep 3000 chromatography system (column Delta-Pak C-18, 15 μm, 100 Å, 180 × 20 mm, flow rate 8 mL/min, RI detection). Semi-preparative HPLC was performed on a Waters apparatus.

Animal Material. The sponge *Dysidea fusca* was collected off the East Coast of New Caledonia. A sample (ref. R1565) was identified by C. Lévi, (Muséum d'Histoire Naturelle, Paris, France), and is preserved at ORSTOM, Noumea, New Caledonia.

Extraction and Purification. The freeze-dried animal material (500 g) was extracted with 80% EtOH (3 × 4 L) at room temperature. After filtration, the solution was concentrated *in vacuo* to an aqueous suspension, which was extracted with CH_2Cl_2. The organic extract (11.5 g) was subjected to successive Si gel column chromatographies with heptane containing increasing concentrations of EtOAc, followed by HPLC on reversed phase, yielding **2**, 14 mg [(1) heptane−EtOAc 90:10, (2) heptane−EtOAc 90:10, (3) preparative HPLC MeOH−H_2O 80:20)]; **1**, 37 mg [(1) heptane−EtOAc 90:10, (2) heptane−EtOAc 90:10)], a mixture of **5** and **6**, 34 mg [(1) heptane−EtOAc 80:20, (2) heptane−EtOAc 85:15, (3) preparative HPLC MeOH−H_2O 70:30)]; **3**, 65 mg [(1) heptane−EtOAc 80:20, (2) heptane−EtOAc 85:15, (3) preparative HPLC MeOH−H_2O 70:30], and **4**, 98 mg [(1) heptane−EtOAc 70:30, (2) heptane−EtOAc 85:15]. The mixture of **5** and **6** (20 mg) was further purified by semi-preparative HPLC (column Nucleodex βPM 250 × 10 mm, eluent MeOH−H_2O 65:35, flow rate 2 mL/mn, UV detection at 230 nm) yielding **5** (4.7 mg) and **6** (4.1 mg).

Compound 1: colorless oil; $[\alpha]_D$ −38° ($CHCl_3$, *c* 1); lit.[2] −46.8°; 1D NMR, see lit.[2]; NOESY correlations 1α/1β, 1β/9, 1β/11β, 2α/2β, 3α/3β, 5/9, 6α/6β, 6α/15, 8/9, 11β/13, 12α/8, 12β/13.

Compound 2: colorless oil; $[\alpha]_D$ −6° ($CHCl_3$, *c* 0.6); HRCIMS *m/z* 239.2016 (MH^+, Δ −0.5); NMR, see Table 1.

Compound 3: colorless oil; $[\alpha]_D$ +37° ($CHCl_3$, *c* 1.4); IR ($CHCl_3$) ν max 1728 cm^{-1}; HRCIMS *m/z* 325.2384 (MH^+, Δ 0.5); NMR, see Table 1.

Compound 4: colorless oil; $[\alpha]_D$ +47° ($CHCl_3$, *c* 0.4); IR ($CHCl_3$) ν max 3400 and 1712 cm^{-1}; HRCIMS *m/z* 253.1816 (MH^+, Δ 1.2 mmu); NMR, see Table 1.

Compound 5 : colorless oil; $[\alpha]_D$ +87° ($CHCl_3$, *c* 0.5); IR ($CHCl_3$) ν max 3400, 1762, and 1670 cm^{-1}; HRCIMS *m/z* 251.1652 (MH^+, Δ −0.5 mmu); NMR, see Table 2.

Compound 6: colorless oil; $[\alpha]_D$ +74° ($CHCl_3$, *c* 0.4); IR ($CHCl_3$) ν max 3400, 1756, and 1670 cm^{-1}; NMR, see Table 2.

Acknowledgment. We thank Prof. C. Lévi for his kind identification of the sponge.

References and Notes

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(5) This work has been done in the frame of the CNRS-ORSTOM program "Substances Marines d'Intérêt Biologique" (SMIB).

NP9603737