Three New Scalarane-Based Sesterterpenes from the Tropical Marine Sponge *Strepsichordaia lendenfeldi*[1]

Thorsten Jahn,[†] Gabriele M. König,* and Anthony D. Wright

Institute for Pharmaceutical Biology, Technical University of Braunschweig, Mendelssohnstrasse 1, D-38106 Braunschweig, Germany

Received September 11, 1998

From the dichloromethane extract of the tropical marine sponge *Strepsichordaia lendenfeldi* collected from the Great Barrier Reef, Australia, three new (**1**, **2**, and **9**) and seven known (**3**−**8** and **10**) scalarane-based sesterterpenes were isolated. All molecular structures were secured by spectroscopic methods, particularly 1D and 2D NMR, and accurate mass measurement.

Sponges belonging to the family Dictyoceratida are often very good sources of scalarane-based sesterterpenes,[2−5] with those of the genera *Strepsichordaia* and *Carteriospongia* being particularly good examples.[2−5] It is likely that sponges from these genera require more chemical protection from predators than other sponges, as they are in general soft-bodied and lack a high degree of spiculation. A characteristic feature of the scalarane-based sesterterpenes isolated from such sponges is the presence of additional methyl groups at C-24 and C-19 (or C-20). At one point, it was thought that this characteristic was unique to sponges of the genus *Carteriospongia*,[3] but, in light of a later report, this appears no longer to be the case.[5] Many of these scalarane-based isolates have been reported to have interesting biological activities,[1,5] and it was on this basis that the current specimen was selected for study. The following report outlines the isolation and characterization of three new (**1**, **2**, and **9**) and seven known (**3**−**8**, **10**) scalarane-based sesterterpenes.

Chromatographic separation of the dichloromethane solubles of *Strepsichordaia lendenfeldi* Bergquist (Dictyoceratida, Irciniidae) yielded 10 scalarane-based sesterterpenes (**1**−**10**), of which three (**1**, **2**, and **9**) are new natural products.

The molecular formula of **1** was deduced as $C_{32}H_{52}O_6$ by accurate mass measurement. From its ^{13}C NMR and IR data, it was evident that the molecule must contain a secondary hydroxyl [64.3 (d) ppm, 3470 cm^{-1}], a keto [209.4 (s) ppm], and two ester [170.5 (s), 171.7 (s), 73.9 (d), 76.5 (d) ppm and 1730 cm^{-1}] groups as the oxygen-containing functionalities within the molecule. As these functional groups also accounted for all of the multiple bonds in the molecule, the remaining four elements of unsaturation indicated by the molecular formula of **1** must be present in the form of rings; **1** is thus tetracyclic. From both the ^1H and ^{13}C NMR data (see Tables 1 and 2), it was possible to identify the presence of a methyl ketone [209.4 (s), 2.11 (s), ppm], an acetate {170.5 (s), [73.9 (d), 4.75 (m)], [21.4 (q), 2.08 (s)] ppm}, and a 3-hydroxybutanoyloxy {171.7 (s), [43.1 (t), 2.32 (dd, J = 3.6, 16.4 Hz), 2.40 (dd, J = 8.7, 16.2 Hz)], [64.3 (d), 4.15 (dddq, J = 3.2, 3.6, 8.7, 6.6 Hz)], [22.4 (q), 1.20 (q, J = 6.6 Hz)], [76.5 (d), 4.99 (ddd, J = 5.1, 10.6, 10.7 Hz)] ppm} moiety, as well four further tertiary methyl groups. With these deductions in hand, a literature search was undertaken. The results of this search suggested **1** to be a sesterterpene similar to ones reported in 1992.[5] On closer examination, the NMR data of compounds **1** and **4** appeared to be almost identical. Where differences occurred, they were associated with the presence or absence of an aldehyde substituent at C-18. Compound **1** was deduced to have, in a relative sense, the same stereochemistry at comparable centers to those found in **4** on the basis of its analogous ^{13}C NMR data. Thus, **1** is best described as 25-nor-12α-acetoxy-16β-(3′-hydroxybutanoyloxy)-20,24-dimethyl-24-oxoscalarane.

Mass spectral analysis of **2** indicated it had the molecular formula $C_{33}H_{52}O_7$. Its ^{13}C NMR and IR data indicated the molecule to contain two secondary hydroxyl [63.6 (d), 64.3

Table 1. ^{13}C NMR Data for Compounds **1** (100 MHz, $CDCl_3$), **2** (125 MHz, $CDCl_3$), **4**, **8**, and **9** (100 MHz, $CDCl_3$)

carbon	**1**	**2**	**4**	**8**	**9**
1	40.2 t[a]	40.2 t	40.2 t	40.0 t	40.1 t
2	18.3 t	18.2 t	18.2 t	18.1 t	18.2 t
3	36.6 t	36.6 t	36.5 t	36.6 t	36.7 t
4	36.1 s	36.1 s	36.1 s	36.1 s	36.2 s
5	58.6 d	58.6 d	58.5 d	58.5 d	58.5 d
6	18.0 t	18.0 t	17.9 t	17.9 t	18.0 t
7	41.5 t	40.5 t	41.6 t	41.8 t	41.9 t
8	37.5 s	38.2 s	38.0 s	37.8 s	37.9 s
9	53.1 d	53.5 d	52.7 d	52.9 d	53.0 d
10	37.0 s	36.9 s	36.9 s	36.8 s	36.9 s
11	22.4 t	22.2 t	21.7 t	21.9 t	22.0 t
12	73.9 d	75.2 d	74.7 d	75.3 d	75.3 d
13	37.4 s	37.7 s	40.5 s	40.2 s	40.3 s
14	50.8 d	46.8 d	50.8 d	48.9 d	49.0 d
15	25.5 t	26.0 t	25.8 t	23.7 t	23.8 t
16	76.5 d	74.7 d	76.2 d	142.9 d	142.9 d
17	49.7 d	49.5 d	49.1 d	137.1 s	137.2 s
18	39.2 t	45.2 d	58.5 d	52.2 d	52.3 d
19	28.5 q	28.4 q	28.4 q	28.5 q	28.5 q
20	24.5 t	24.5 t	24.4 t	24.5 t	24.6 t
21	16.9 q	15.1 q	16.9 q	17.1 q	17.1 q
22	16.6 q	16.7 q	16.8 q	16.6 q	16.7 q
23	20.6 q	37.6 t	17.2 q	15.2 q	15.3 q
24	209.4 s	211.5 s	210.9 s	198.5 s	198.5 s
25		63.6 d	202.1 d	200.8 d	200.7 d
26	29.1 q	30.2 q	33.0 q	25.1 q	25.1 q
27	8.6 q	8.6 q	8.6 q	8.6 q	8.7 q
OAc-12	21.4 q	21.4 q	21.5 q		
	170.5 s	171.0 s	170.2 s		
OR 1′	171.7 s	171.9 s	171.5 s	169.1 s	169.1 s
2′	43.1 t	43.1 t	42.7 t	38.9 t	41.1 t
3′	64.3 d	64.3 d	64.1 d	21.1 q	9.1 q
4′	22.4 q	22.5 q	22.4 q	170.5 s	27.7 t
5′				71.6 d	173.7 s
6′				26.7 t	67.3 d
7′				9.4 q	19.9 q

[a] Multiplicity by DEPT (s = C, d = CH, t = CH_2, q = CH_3).

* To whom correspondence should be addressed. Tel.: +49 531 391 5680. Fax.: +49 531 391 8104. E-mail: g.koenig@tu-bs.de. Internet: http://www.tu-bs.de/institute/pharm.biol/GAWK.html.

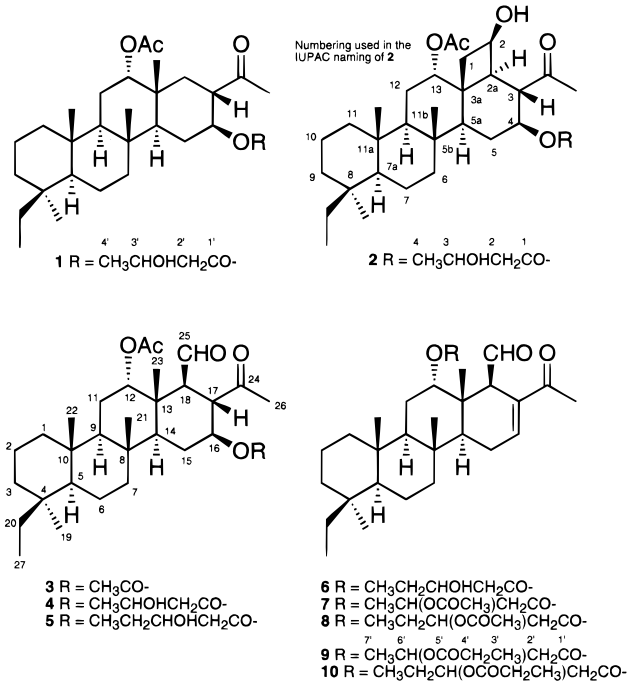
[†] Current address: Salutas Pharma GmbH, Otto von Guericke Allee 1, Barleben D 39179, Germany.

10.1021/np980389s CCC: $18.00
Published on Web 01/21/1999

Table 2. ^{1}H NMR Data for Compounds **1** (400 MHz, $CDCl_3$), **2** (600 MHz, $CDCl_3$), and **9** (400 MHz, $CDCl_3$)[a]

proton	**1**[b]	**2**	**9**
1	0.63 (1α, ddd, *J* = 4.2, 12.6, 12.6 Hz), 1*β**[c]	0.64 (m), 1.52 (m)	0.63 (ddd, *J* = 4.1, 13.1, 13.2 Hz), 1.72 (m)
2	*	1.32 (m), 1.42 (m)	1.30 (m), 1.52 (m)
3	*	0.81 (m), 1.63 (m)	0.86 (m), 1.67 (m)
5	*	0.86 (m)	0.90 (m),
6	*	1.36 (m), 1.63 (m)	1.32 (m), 1.52 (m)
7	*	1.05 (ddd, *J* = 3.6, 13.2, 13.2 Hz), 1.57 (m)	0.97 (m), 1.75 (m)
9	*	1.19 (m)	1.25 (m)
11	*	1.51 (m), 1.71 (br d, *J* = 15.8 Hz)	1.86 (br d, *J* = 14.3 Hz)
12	4.75 (m)	5.03 (m)	4.79 (br s)
14	*	1.51 (m)	1.46 (m)
15	1.35 (m), 2.03 (m)	1.22 (m), 1.95 (m)	2.29 (m)
16	4.99 (ddd, *J* = 5.1, 10.6, 10.7 Hz)	4.87 (ddd, *J* = 3.5, 11.4, 11.5 Hz)	7.09 (m)
17	2.84 (ddd, *J* = 3.5, 10.6, 14.2 Hz)	3.35 (dd, *J* = 9.3, 11.5 Hz)	
18	*	2.27 (m)	3.45 (br s)
19	0.80 (s)	0.78 (s)	0.83 (m)
20	*	1.13 (m), 1.47 (m)	1.16 (m), 1.53 (m)
21	0.81 (s)	0.69 (s)	0.81 (s)
22	0.81 (s)	0.78 (s)	0.96 (s)
23	1.09 (s)	1.99 (m), 2.25 (m)	0.94 (s)
25		4.41 (ddd, *J* = 7.0, 7.0, 8.4 Hz)	9.38 (d, *J* = 4.1 Hz)
26	2.11 (s)	2.18 (s)	2.32 (s)
27	0.73 (t, *J* = 7.5 Hz)	0.72 (t, *J* = 7.6 Hz)	0.74 (t, *J* = 7.6 Hz)
OAc-12 CH_3	2.08 (s)	2.11 (s)	
OR 2′	2.32 (dd, *J* = 3.6, 16.4 Hz)	2.38 (dd, *J* = 3.5, 16.2 Hz)	2.65 (dd, *J* = 5.2, 15.4 Hz)
	2.40 (dd, *J* = 8.7, 16.4 Hz)	2.42 (dd, *J* = 3.8, 16.2 Hz)	2.76 (dd, *J* = 7.1, 15.4 Hz)
3′	4.15 (dddq, *J* = 3.2, 3.6, 8.7, 6.6 Hz)	4.16 (ddq, *J* = 3.5, 3.8, 6.0 Hz)	1.10 (t, *J* = 6.0 Hz)
4′	1.20 (q, *J* = 6.6 Hz)	1.21 (q, *J* = 6.0 Hz)	2.27 (q, *J* = 6.0 Hz)
6′			5.34 (ddq, *J* = 5.2, 7.1, 6.0 Hz)
7′			1.36 (d, *J* = 6.0 Hz)
3′-O*H*	2.91 (d, *J* = 3.2 Hz)		

[a] All assignments are based on extensive 1D and 2D NMR experiments, including COSY90, HMQC, HMBC, and NOE difference. [b] As no HMQC spectrum was recorded for this sample assignments are based on COSY90 measurements only. [c] Protons not assigned a resonance as denoted by a *, all lie in the region *δ* 0.70−1.75.



(d) ppm, 3450 cm^{-1}], a keto [211.5 (s) ppm, 1670 cm^{-1}], and two ester [171.0 (s), 171.9 (s), 75.2 (d), 74.7 (d) ppm, 1730 cm^{-1}] functions as the oxygen-containing functionalities within the molecule. As was the case for **1**, these functionalities accounted for all of the multiple bonds in the molecule, so the remaining five elements of unsaturation indicated by the molecular formula of **2** must be present in the form of rings; **2** was thus pentacyclic. Comparison of the NMR data, particularly the ^{13}C NMR data, of **2** with those of **1** and **4** revealed the three molecules to be similar, except in the regions of C-14, C-18, C-23, and C-25. From the ^{1}H−^{1}H COSY spectrum of **2** a chain of coupling could be traced from H_2-23 (a methyl group in **1** and **4**) to H-14. Thus, H_2-23 [1.99 (m), 2.25 (m) ppm] demonstrated coupling with H-25 [4.41 (ddd, *J* = 7.0, 7.0, 8.4 Hz) ppm] (absent in **1**, and an aldehyde in **4**), which coupled to H-18 [2.27 (m) ppm], which in turn coupled with H-17 [3.35 (dd, *J* = 9.3, 11.5 Hz) ppm]; in turn, H-17 coupled to H-16 [4.87 (ddd, *J* = 3.5, 11.4, 11.5 Hz) ppm], which coupled to H_2-15 [1.22 (m), 1.95 (m) ppm]; and finally, H_2-15 coupled to H-14 [1.51 (m) ppm]. With this ^{1}H−^{1}H spin system delineated, it was evident that C-23 and C-25 were directly bonded, giving rise to the fifth ring in **2**, a cyclobutanol moiety between C-13 and C-18. From NOE difference measurements made with **2**, it was evident that the cyclobutanol moiety had, in a relative sense, a *β*-configuration, as did the hydroxyl function at C-25. Thus, low-power irradiation at the resonance frequency of H-12 caused enhancement of resonances associated with H_2-11, H_2-23, and H-25, while irradiation of H-25 resulted in the enhancement of signals associated with H-12 and H_2-23. All other centers had the same relative configurations as those found in **1** on the basis of comparable ^{13}C NMR chemical shifts for the same centers. Compound **2** is thus the new natural product 12α-acetoxy-16*β*-(3′-hydroxybutanoyloxy)-13*β*,18*β*-cyclobutan-20,24-dimethyl-24-oxoscalaran-25-ol. The IUPAC name for the configuration shown of this compound is (2a*S**,3*S**,4*S**,5a*S**,7a*S**,8*S**,11a*S**,13*S**,13a*S**,2*R**,5b*R**,11b*R**)-3-acetyl-8-ethyl-2-hydroxy-5b,8,11a-trimethyl-13-methylcarbonyloxyperhydrocyclobuta[*i*]chrysen-4-yl 3-hydroxybutanoate. Compound **2** is only the second example of this class of compound to contain a cyclobutanol moiety, with the other being a constituent of *Carteriospongia foliascens*.[2]

By mass spectral analysis **9** was deduced to have the molecular formula $C_{34}H_{52}O_6$, and thus have nine elements of unsaturation. From its ^{1}H and ^{13}C NMR, IR, and UV spectral data, the functionalities within the molecule were deduced as two esters {(169.1 (s), 173.7 (s), [75.3 (d), 4.79 (br s)], [67.3 (d), 5.34 (ddq, *J* = 5.2, 7.1, 6.0 Hz)] ppm, 1740 cm^{-1})}, an aldehyde [200.7 (d), 9.38 (d, *J* = 4.1 Hz) ppm, 1740 cm^{-1}], and a conjugated ketone {137.2 (s), [142.9 (d),

7.09 (m)], 198.5 (s) ppm, 1740 cm^{-1}, 232 nm (ϵ 18 650)}. These four functional groups accounted for all five multiple bonds within the molecule; **9** was tetracyclic. By comparing the deduced structural features and the ^{13}C NMR data of **9** with those for **6**−**8** and **10**, it was obvious that **9** had the same basic structure as these molecules, but differed in the composition of the ester function at C-12. From the NMR data of **9** the substituent at C-12 was identified as a 3-propanoyloxybutanoyloxy moiety. Thus, C*H*$_3$-7′ [1.36 (d, J = 6.0 Hz) ppm] coupled to C*H*-6′ [2.27 (q, J = 6.0 Hz) ppm], which in turn coupled to C*H*$_2$-2′ [2.65 (dd, J = 5.2, 15.4 Hz), (2.76 (dd, J = 7.1, 15.4 Hz) ppm}. Also, C*H*$_2$-2′ long-range coupled to C-1′ [169.1 (s) ppm], thus completing the 3-substituted butanoyloxy moiety. Further, C*H*$_3$-3′ [1.1 (t, J = 6.0 Hz) ppm] coupled to C*H*$_2$-4′ [2.27 (q, J = 6.0 Hz) ppm], which long-range coupled to C-5′ [173.7 (s) ppm], and finally C*H*-2′ also long-range coupled to C-5′, thus delineating the propanoyloxy moiety and its point of attachment to the butanoyloxy function. As the ^{13}C NMR data for **6**−**8** and **10** were virtually identical to those of **9**, it was concluded that the relative stereochemistry of **9** at all chiral centers was also the same. The molecule is best described as 12α-(3′-propanoyloxybutanoyloxy)-20,24-dimethyl-24-oxoscalar-16-en-25-al.

Together with **1**, **2**, and **9**, the seven previously reported scalarane-based sesterterpenes (**3**−**8** and **10**) were also isolated.[5] All of the isolates from this study are currently the subject of high throughput biological screening, the results of which will be published elsewhere.

Experimental Section

General Experimental Procedures. Procedures were as previously published.[6]

Animal Material. Sponges of the species *Strepsichordaia lendenfeldi* (originally assigned as *Carteriospongia calciformis* Carter, 1885) were collected in March 1993, by divers from depths of 6−9 m, at Kelso Reef, Great Barrier Reef, Australia. A voucher specimen representative of this collection is held at the Muséum d'Histoire Naturelle, Case Postale 6434, CH-1211, Geneva 6, Switzerland (voucher number CT293R). Animals were deep frozen and then freeze-dried prior to extraction.

Extraction and Isolation. Freeze-dried animals (181.3 g) were extracted first with CH_2Cl_2 (2.5 L) and then with MeOH (2 L). VLC of the resultant CH_2Cl_2 solubles (7.64 g, 4.2%) over Si gel employing a step gradient from hexane to EtOAc to MeOH yielded 15 fractions, each of 110 mL.

TLC and ^{1}H NMR inspection of all fractions indicated fractions 9−12 to be of further interest, mainly on the basis of the many low-field resonances observed in their proton spectra. HPLC purification [Si gel 60, Me_2CO−hexane (1:4) as eluent] of fraction 9 yielded compounds **4** and **7**−**10**. HPLC separation [Si gel 60, Me_2CO−hexane (1:3) as eluent, followed by Si gel 60, Me_2CO−hexane (1:4) as eluent] of fraction 10 yielded compounds **6**−**8**. HPLC [Si gel 60, Me_2CO−hexane (3:7) as eluent] of fraction 11 yielded compounds **1**−**3** and **5**. HPLC purification [Si gel 60, Me_2CO−hexane (3:7), followed by Me_2CO−CH_2Cl_2 (3:7), followed by RP18, H_2O−CH_3CN (1:9) as eluent] of fraction 12 yielded compounds **1**−**5**.

25-Nor-12α-acetoxy-16*β*-(3′-hydroxybutanoyloxy)-20,24-dimethyl-24-oxoscalarane (1): clear oil (71.0 mg, 0.039%); $[\alpha]^{25}_D$ +26.5° (*c* 1.62, $CHCl_3$); IR (film) ν_{max} 3460, 2960, 1730, 1245 cm^{-1}; ^{1}H NMR, see Table 2; ^{13}C NMR, see Table 1; EIMS *m*/*z* 533 ([M + H]$^+$, <1), 532 (M$^+$, <1), 473 (2), 472 (16), 444 (4), 443 (24), 397 (12), 396 (30), 369 (45), 368 (100), 353 (32), 339 (40), 325 (32), 219 (24), 205 (36); HRDCIMS (isobutane) *m*/*z* found 533.3837 ([M + H]$^+$), calcd for $C_{32}H_{53}O_6$ 533.3842.

12α-Acetoxy-16*β*-(3′-hydroxybutanoyloxy)-13*β*,18*β*-cyclobutan-20,24-dimethyl-24-oxoscalaran-25-ol (2): clear oil (31.5 mg, 0.017%); $[\alpha]^{25}_D$ +31.4° (*c* 2.07, $CHCl_3$); IR (film) ν_{max} 3460, 2960, 1730, 1250 cm^{-1}; ^{1}H NMR, see Table 2; ^{13}C NMR, see Table 1; CIMS (NH_3) *m*/*z* 578 ([M + NH_4]$^+$, 7), 475 (10), 474 (34), 398 (20), 397 (70), 381 (18), 379 (28), 122 (100); HRDCIMS (NH_3) *m*/*z* found 474.3590 ([M − $C_4H_8O_2$ + NH_4]$^+$), calcd for $C_{29}H_{48}NO_4$ 472.3583.

12α,16*β*-Diacetoxy-20,24-dimethyl-24-oxoscalaran-25-al (3): clear oil (8.6 mg, 0.0047%); $[\alpha]^{25}_D$ +57.1° (*c* 2.07, $CHCl_3$), lit. +88.1°,[5] and remaining spectroscopic data as previously reported.[5]

12α-Acetoxy-16*β*-(3*R*′-hydroxybutanoyloxy)-20,24-dimethyl-24-oxoscalaran-25-al (4): clear oil (22.8 mg, 0.013%); $[\alpha]^{25}_D$ +40.7° (*c* 0.15, $CHCl_3$), lit. +61.3°,[5] and remaining spectroscopic data as previously reported.[5]

12α-Acetoxy-16*β*-(3′-hydroxypentanoyloxy)-20,24-dimethyl-24-oxoscalaran-25-al (5): clear oil (14.1 mg, 0.0078%); $[\alpha]^{25}_D$ +53.6° (*c* 0.64, $CHCl_3$), cf +60.3°,[5] and remaining spectroscopic data as previously reported.[5]

12α-(3*R*′-Hydroxypentanoyloxy)-20,24-dimethyl-24-oxoscalara-16-en-25-al (6): clear oil (1.2 mg, 0.0007%); $[\alpha]^{25}_D$ +18.3° (*c* 0.12, $CHCl_3$), lit. +43.0°,[5] and remaining spectroscopic data as previously reported.[5]

12α-(3′-Acetoxybutanoyloxy)-20,24-dimethyl-24-oxoscalara-16-en-25-al (7): clear oil (21.1 mg, 0.012%); $[\alpha]^{25}_D$ +19.0° (*c* 0.10, $CHCl_3$), lit. +47.7°,[5] and remaining spectroscopic data as previously reported.[5]

12α-(3′-Acetoxypentanoyloxy)-20,24-dimethyl-24-oxoscalara-16-en-25-al (8): clear oil (57.8 mg, 0.032%); $[\alpha]^{25}_D$ +26.2° (*c* 0.81, $CHCl_3$), lit. +44.6°,[5] and remaining spectroscopic data as previously reported.[5]

12α-(3′-Propanoyloxybutanoyloxy)-20,24-dimethyl-24-oxoscalar-16-en-25-al (9): clear oil (12.4 mg, 0.0068%); $[\alpha]^{25}_D$ +56.5° (*c* 1.25, $CHCl_3$); UV λ_{max} (MeOH) 232 (ϵ 18 650) nm; IR (film) ν_{max} 2930, 1740, 1185 cm^{-1}; ^{1}H NMR, see Table 2; ^{13}C NMR see, Table 1; EIMS *m*/*z* 557 ([M + H]$^+$, <1), 556 (M$^+$, <1), 538 (<1), 529 (4), 528 (10), 397 (8), 396 (12), 369 (26), 368 (80), 353 (12), 339 (32), 325 (10), 219 (76), 205 (100); HREIMS *m*/*z* found 556.3760 (M$^+$), calcd for $C_{34}H_{52}O_6$ 556.3764.

12α-(3′-propanoyloxypentanoyloxy)-20,24-dimethyl-24-oxoscalara-16-en-2-5-al (10): clear oil (37.9 mg, 0.021%); $[\alpha]^{25}_D$ +50.0° (*c* 0.17, $CHCl_3$), lit. +37.4°,[5] and remaining spectroscopic data as previously reported.[5]

Acknowledgment. We thank Dr. V. Wray and his group (GBF, Braunschweig) for recording all NMR spectra, and Dr. R. Christ (GBF, Braunschweig), and H.-M. Schiebel, Mass Spectral Service, Department of Chemistry, TU-BS, for making all mass spectral measurements. Financial support from the DFG (Deutsche Forschungsgemeinschaft), grant number KO-902/2-1, and BMBF (Ministry of Education and Research), grant number 0339547, is gratefully acknowledged. We also thank Dr. R. Desqueyroux-Faundez, Muséum d'Histoire Naturelle, Case Postale 6434, CH-1211, Geneva 6, Switzerland, for original identification of the sponge sample, and Dr. B. F. Bowden, Department of Chemistry and Biochemistry, James Cook University of North Queensland, Queensland, Australia, and Dr. P. R. Bergquist, Department of Zoology, University of Auckland, Auckland, New Zealand, for correcting the original species assignment.

References and Notes

(1) Presented in part at the 5th International Sponge Symposium, 29 June−3 July, 1998, Brisbane, Queensland, Australia (poster 70), and at the 9th International Symposium on Marine Natural Products, 5−10 July 1998, Townsville, Queensland, Australia (poster 15).
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NP980389S