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STEROLS OF THE JELLYFISH PERIPHYLLA PERIPHYLLA. IDENTIFICATION OF ISOFUCOSTANOL.

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As part of our investigations into the sterols of marine organisms and their origin through the marine food web, we have examined the sterols present in the lipids from a deep sea jellyfish by GC-MS techniques, and wish to report the identification of seventeen different marine sterols, including no less than seven sterols with saturated ring systems. One of these, 24-ethyl-5α-cholest-24(28)Z-en-3β-ol (isofucostanol) is a new marine sterol.

The sample (3 specimens of Periphylla periphylla) was collected at a depth of 210-400 m by nets during a biological cruise of R.R.S. Discovery in the N.E. Atlantic and immediately deep frozen under nitrogen. The lipids were extracted in the usual way and the sterol TMS derivatives were examined by combined GC-MS techniques using 1% Dexsil 300GC and Silar 5CP columns as described previously.[1] The chromatograms are illustrated in Figs. 1 and 2 and the retention data are given in Table 1. The sterols were identified by comparison of GLC retention data with those of the TMS derivatives of standard sterols and by multiple MS scanning of each GLC peak to identify all overlapping constituents.[1]

The complete sterol analysis is presented in Table 1 and the jellyfish was found to contain mainly cholesterol and 22-dehydroderivatives but surprisingly was also found to contain seven pairs of sterols of which Δ 5 sterols were accompanied by the corresponding 5α-stanols: e.g. sterols 1/2, 3/4, 5/6, 7/9, 10/12, 14/15 and 16/17.

Dexsil 300GC is one of the very few GLC column systems with the capability of separating Δ 5 sterols from their corresponding 5α-stanols and it was found that there was a constant separation factor between these pairs of compounds (Table 2). This relationship enabled the identity of the unknown sterol 17 to be established.

In the case of the Silar 5CP column the Δ 5 sterols and the corresponding 5α-stanols could not be resolved but had identical retention times in all cases.

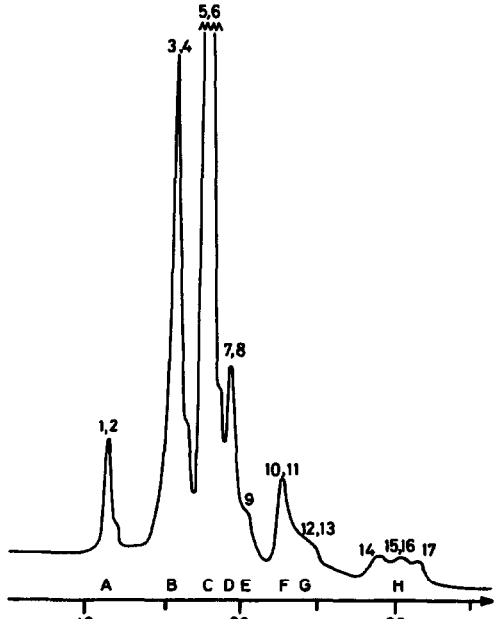

FIG.1 DEXSIL 300 GC

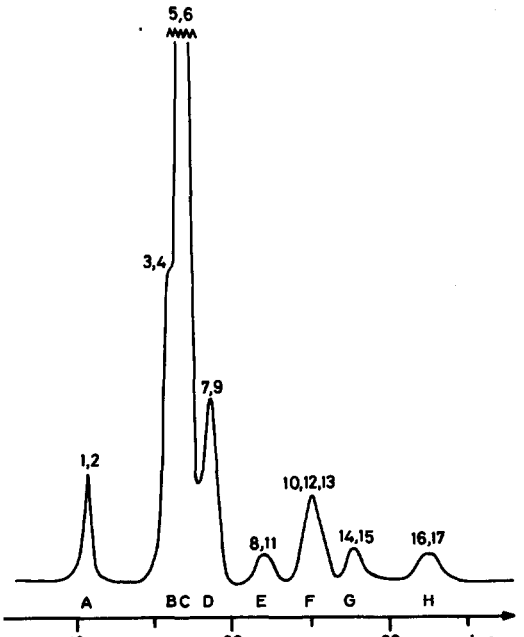

FIG.2 SILAR 5CP

Table 1. Data for Periphylla Sterols (TMS derivatives)

Sterol	Identity*		GLC-Dexsil		GLC-Silar		% of
			Peak (Fig 1)	RRT	Peak (Fig 2)	RRT	Sterol
1	26C	5, 22t	A	.625	A	.64	1.7
2	26C	22t	A	.89	A	.64	0.9
3	27C	5, 22t	B	.89	B	.93	17.3
4	27C	22t	B	.93	B	.93	
5	27C	5	C	1.0	C	1.0	62.6
6	27C		C	1.04	C	1.0	1.0
7	28C	5, 22	D	1.09	D	1.10	6.3
8	27C	5, 24(25)	D	1.09	E	1.30	0.7
9	28C	22	E	1.14	D	1.10	1.9
10	28C	5, 24(28)	F	1.28	F	1.45	2.5
11	28C	5	F	1.28	E	1.30	0.7
12	28C	24(28)	G	1.34	F	1.45	1.4
13	29C	5, 22	G	1.35	F	1.45	0.5
14	29C	5	H	1.62	G	1.58	1.0
15	29C		H	1.68	G	1.58	0.3
16	29C	5, 24(28)Z	H	1.68	H	1.88	0.8
17	29C	24(28)Z	H	1.74	H	1.88	1.0

* Sterol 7 is either crinosterol or brassicasterol, 9 is either spongesterol or neospongesterol, 11 is either 22, 23-dihydrobrassicasterol or campesterol, 13 is either stigmasterol or poriferasterol, 14 is either clionasterol or β-sitosterol, and 15 is either clionastanol or β-sitostanol.

Table 2.
TMS Derivatives on Dexsil 300 GC

5α-Stanol	Δ5 Sterol	Ratio of RRT	Separation Factor
26C 22t	26C 5, 22t	.65 / .625	1.040
27C 22t	27C 5, 22t	.93 / .89	1.045
27C	27C 5	1.04 / 1.00	1.040
28C 22	28C 5, 22	1.14 / 1.09	1.046
28C 24(28)	28C 5, 24(28)	1.34 / 1.28	1.046
29C	29C 5	1.68 / 1.62	1.037
29C 24(28)Z	29C 5, 24(28)Z	1.74 / 1.68	1.036

Sterol 17 was found to co-occur with iso-fucosterol (16) in peak H in the Silar chromatogram and to occur in the tail of peak H in the Dexsil chromatogram. The relative retention time of sterol 17 on Dexsil was established as 1.74 by fixing the position of the centroid for the large m/e 388 ion by mass scanning of peak H and corresponds to the calculated retention time for the TMS derivative of 24-ethyl-5α-cholest-24(28)Z-en-3β-ol (Table 2). The mass spectrum of sterol 17 TMS had significant peaks at m/e (%): 486 (6) $M^{+\cdot}$; 471 (8) M-15; 388 (65) M-98; 229 (9); 217 (9) M-90-$C_{10}H_{19}$-40; 216 (15) M-90-$C_{10}H_{19}$-41; 215 (30) M-90-$C_{10}H_{19}$-42; 95 (68); 75 (73); 73 (82); 69 (91); 57 (94); 55 (100); 43 (98%). The molecular ion at m/e 486 corresponds to a $C_{29:1}$ sterol and the M-98 ion resulting from a McLafferty rearrangement clearly establishes the presence of a 24-ethyl-24(28)-ene system.[2] The abundant m/e 215 ion establishes the presence of a saturated 5α-ring system[3,4] and sterol 17 is therefore formulated as 24-ethyl-5α-cholest-24(28)Z-en-3β-ol (iso-fucostanol). The 24(28)E isomer can be discounted because of the large retention time difference of E and Z isomers on Silar 5CP[1].

A 24-ethylidenecholestanol of unspecified stereochemistry has been briefly mentioned by Goad et al.[5] in unpublished investigations into the sterols of the echinoderm Henricia sanguinolenta but Periphylla periphylla is the first marine organism in which iso-fucostanol has been identified.

Previous investigations of the sterols present in other members of the Medusae species[6] have revealed cholestanol in only one member and no stanols whatsoever in any other members and the presence of seven stanols in periphylla is a most unexpected result.

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