SHORT PAPER

J. Chem. Research (S), 2000, 182–183

A new polyoxygenated steroid from the gorgonian *Gorgonella umbraculum*†

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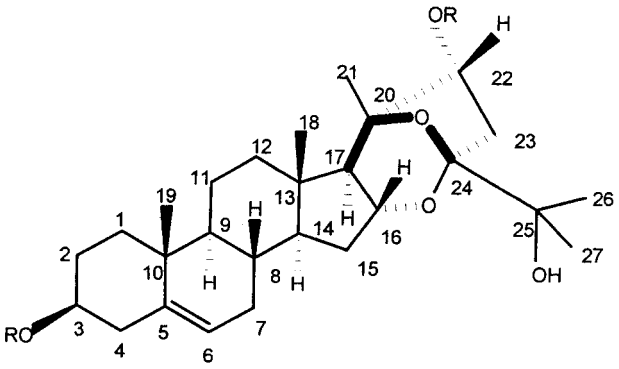
A new polyoxygenated steroid, **1**, has been isolated from the Indian ocean gorgonian *Gorgonella umbraculum* and its structure established from its spectral data.

The Indian ocean gorgonian *Gorgonella umbraculum* (Ell & Sol) (family Ellesellidae) has yielded six briarane diterpenoids.[1,2] Further purification of the more polar chromatographic fractions of its EtOAc solubles resulted in the isolation of a new polyoxygenated steroid, **1**. We present its structural elucidation in this report.

The steroid, **1**, analysed for $C_{27}H_{42}O_5$ and readily formed a diacetate (**2**). The ^{1}HNMR spectrum showed one olefinic proton, two secondary hydroxyl groups [δ 3.53 br m, $W^1/_2$ 20.0Hz, (1H) – shifted to 4.60 br m in **2** and 3.87 dd (1H) shifted to 4.70 dd in **2**], an axial oxymethine proton (4.51 br m, $W^1/_2$ 17.0 Hz) and five tertiary methyl groups. The ^{13}C NMR spectrum showed that **1** was a 3 β-hydroxy-Δ^5-cholestene derivative (δ 71.7d, C-3; 140.5s, C-5; 121.6d, C-6)[3], possessing a ketal group (107.7s), a quaternary oxygen bearing carbon (83.3s) a tertiary hydroxyl group (72.4s), a secondary hydroxyl group (79.3d) and an oxymethine carbon (72.7d). The chemical shifts of C-1 to C-14 and C-19 of **1** are almost identical with the chemical shifts of the same carbons in cholesterol,[3] suggesting that the additional features in **1** are incorporated in its ring D and the side chain.

Analysis of the ^{13}C NMR data of **1** and comparison of these data with that reported for the steroidal C-22 ketals isolated from marine organisms (the hippurins,[4] which are C_{16} β–O–C_{22}–O_C_{25} ketals) and terrestrial plants (diosgenins,[5] which are C_{16} β–O-C_{22}-O-C_{26} ketals) established that **1** was a C-24 ketal connected to the C-16 and C-20 carbons (C_{16}–O-C_{24}–O–C_{20}) and with hydroxyl groups present at C-22 and C-25. The stereochemistry of the ketal and the C-22 secondary hydroxyl groups were inferred from the chemical shifts of the H-16 and C-17 signals. The H-16 was observed as an axial proton (δ 4.51 br m, $W^1/_2$ = 17.0 Hz) and was thus β-oriented. Consequently, the C–O linkage at C-16 was α-oriented. The C-17 signal in **1** was observed at δ 49.5 d, which is a shielded position compared to the value of ca. 60 ppm found in hippurins.[4] This shielding of the C-17 signal indicated the stereochemistry of the ketal group and fixed the location of the additional secondary hydroxyl group at C-22.

Examination of molecular models showed that for the shielding of the C-17 to occur, C-24 should have the (S)-configuration, *i.e.*. the oxygen connecting the C-20 – C-24 carbons lies above the plane in the direction of the C-18 methyl group, and the secondary hydroxyl group is located in the resulting α-oriented C22–C23 part of the 5-membered ether ring at C-22 and in the (R)-configuration, *i.e.*. α-oriented and lying in the same direction as the H-17. In this orientation, the C-17 would experience large shielding due to the γ *gauche* effect[6] of the C-22_α hydroxyl, as found in similar cases, *e.g.* the C14_α–OH – C-17 interaction in ecydysterone,[7] (2β, 3β, 14_α, 20β, 22_α, 25-hexahydroxy-5β-cholest-7-en-6-one) (δ C_{17}, 50.08d). Thus **1** is assigned the (20S), (22 R) and (24 S) configurations. The carbon chemical shifts of C-16 (72.7 d) and C-22 (79.3 d) in **1** were assigned on the basis of their ^{13}C-^{1}H correlations. The C-22 signal is as observed for the C-22 hydroxyl flanked by an oxygen bearing carbon, *e.g.* the C-22 of ecdysterone[7] (δ 77.52d). The C-16 signal, however, does not seem to reflect its ether nature. The value is nearer the value of a free hydroxyl bearing carbon (*e.g.*. 16_α-OH in andostrane, δ 71.8)[8] than an ether bearing carbon. This may be due to the shielding effect on C-16 by C-20–O–C-24 oxygen atom which is located opposite to it. This shielding effect may be as much as the deshielding effect expected from the ether linkage, thus causing little shift in the signal.



1. R = H

2. R = COCH$_3$

Scheme 1

From the foregoing **1** was assigned the structure 3, 22, 25-trihydroxy-16–24, 20–24-bisepoxy-(3β, 16α, 20S, 22R, 24S)-cholest-5-ene. Compound **1** is the first steroidal C-24 ketal to be isolated from a marine organism.

Experimental

Extraction of the gorgonian and isolation of compound **1**: The gorgonian (3.4 kg) collected from the Vallinukum coast, Tamil Nadu, India, was extracted with ethanol (7 × 15 l) and the residue from the EtOH extract was taken up in EtOAc. Solvent was removed from the EtOAc extract and the resulting dark gum (15 g) was chromatographed on a column (60 mm × 1 m) of silica gel (300 g). The column was eluted with increasing polarity from n-hexane through EtOAc. Compound **1** (30 mg) was obtained from the 3:2 hexane : EtOAc eluants.

Compound 1. Colourless plates from EtOAc. mp 237–239°, $[\alpha]_D^{30}$ -23 (C, 0.46, MeOH). Pink spot changing to violet. R_f 0.30 (CH_2 $C1_2$: MeOH 9.9 : 0.1). ν_{max}/cm^{-1} 3393 (br). EIMS : (2.26 eV) *m/z* 429 (35), 428 (M-H_2O, 100), 315 (98%). CIMS (ammonia) : *m/z* 464 (M

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† This is a Short Paper, there is therefore no corresponding material in *J Chem. Research (M)*.

+ NH_4 of C_{27} H_{42} O_5, 100%), 447 (M+1, 68%) and 429 (100%). Found : *m/z* 447.3118 Calc. for M+1 of C_{27} H_{42} O_5 : *m/z* 447.3112. δ_H : (400 MHz, $CDCl_3$, TMS) 1.03s (H-21), 1.07s (H-19), 1.16s (H-18), 1.25s (H-27), 1.37s (H-26), 2.74 dd, *J*=14.0; 5.0 Hz (1H, H-23); 3.50 brm ($w_{1/2}$ 20Hz, H-3α); 3.87dd, *J*=11.0; 5.0 Hz (H-22); 4.51 brm ($w_{1/2}$ 17Hz, H-16β); 5.36 brd (H-6). δ_c : (100 MHz, $CDCl_3$, TMS) (C-1 to C-27) : 37.3, 31.7, 71.7, 42.2, 140.5, 121.6, 31.6, 31.1, 50.2, 36.5, 20.5, 39.6, 41.7, 54.9, 33.4, 72.7, 49.5, 23.4, 14.4, 83.3, 19.4, 79.3, 29.7, 107.7, 72.4, 23.5, 24.0

Compound 2. Compound 1 (5 mg) was acetylated with 0.5 ml each of Ac_2O and pyridine at room temperature (14 hours). The acetate, **2**, was obtained as a colourless gum : R_f 0.68 (CH_2Cl_2 : 9.9 : 0.1). ν_{max}/cm^{-1} 3462 br, 1726, 1234 and 720. δ_H (90 MHz, $CDC1_3$) : δ 5.40 brd (H-6). 4.70 dd, *J*=11.0, 5.0 Hz (1H, H-22), 4.60 m (2H, H-3α and H-16), 2.90 dd, *J*=14.0, 11.0 Hz (1H, H-23), 2.05s (3H), 2.12s (3H), 0.90s (3H), 1.08s (3H), 1.10s (3H), 1.25s (3H), 1.38 (3H).

We are extremely grateful to Dr R.S. Ward, Department of Chemistry, University of Wales Swansea, Swansea, UK for providing the spectral data. We thank Dr P.A. Thomas, Scientist, CMFRI, Vizhinjam, Kerala, India, for the identification of the gorgonian and the UGC, New Delhi for the COSIST and DRS facilities at our department.

Received 1 March 2000; accepted 30 March 2000
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