

Sterols from the soft coral *Lobophytum strictum* (Alcyonarian) from Lakshadweep Sea

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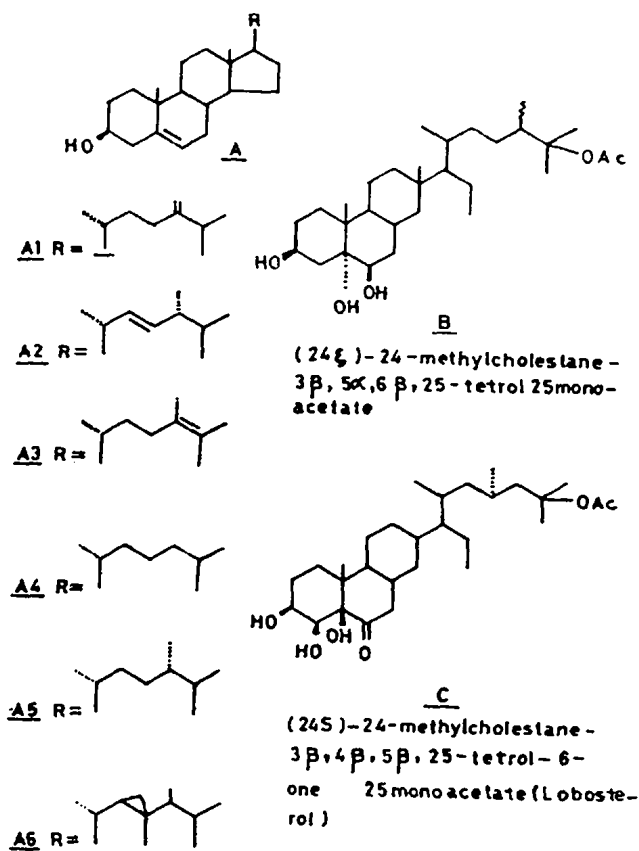
Two polyhydroxy sterols 24 ζ -methylcholestane-3 β , 5 α , 6 β , 25-tetrol, 25-monoacetate, and 24S-methylcholestane-3 β , 4 β , 5 β , 25-tetrol-6-one, 25-monoacetate (lobosterol), six monohydroxy sterols as well as batyl alcohol and its C-16 homologue have been isolated from the chloroform fraction of methanolic extract of *Lobophytum strictum*, the soft coral from Lakshadweep islands. These have been characterised based on their physico-chemical evidences.

Alcyonarians especially those belonging to *Lobophytum* genera are rich sources for many novel compounds belonging to the classes of prostanoids, polyhydroxysterols¹ and terpenoids². The methanol extract of *L. strictum* from Lakshadweep islands exhibited promising *in vivo* hypotensive activity³. In our continuing investigation of this species for possible bioactive compounds, we report herein the isolation and characterisation of two polyhydroxysterols (B) and (C) together with six monohydroxy sterols (A1-A6), batyl alcohol and its C-16 homologue.

The chloroform fraction was broadly separated into low polar (5-20% EtOAc in pet. ether) and more polar (50-100% ethyl acetate in pet. ether) fractions by conventional column chromatography. The former (~ 50 mg) was further purified on reverse phase HPLC (ODS-5 u, MeOH, RI detector) to yield six monohydroxy sterols, viz. 24-methylenecholesterol (A1, Rt 10.4 min), brassica sterol (A2, Rt 11.6 min), 24-methylcholesterol (A3, Rt 12.0 min), cholesterol (A4, Rt 12.4 min), campesterol (A5, Rt 14.0 min) and gorgosterol (A6, 16.4 min). They were characterised by the comparison of their PMR and mass spectral data with those reported in literature.

The more polar fraction was further purified on silica gel columns to yield three fractions, viz. fr. 1, mixture of batyl alcohol and its C-16 homologue, fr. 2 compound (B) and fr. 3 compound (C).

Compound (B) had two secondary hydroxyl and an acetate groups as indicated by its PMR spectra (see Experimental). Its CMR spectrum also indicated the presence of four oxygenated carbons in the molecule (see Experimental). This compound was found to be



identical with 24 ζ - methylcholestane-3 β , 5 α , 6 β , 25-tetrol, 25-monoacetate, previously reported from *Sarcophyton elegans*⁴.

Presence of hydroxyl acetate and carbonyl groups in compound (C) were revealed by its IR (nujol), PMR and CMR spectra. This compound was found to be identical with lobosterol [(24S)-24-methylcholestane-3 β , 4 β , 5 β , 25-tetrol-6-one, 25-monoacetate], previously reported from *L. paciflorum*⁵.

Compounds (B) and (C) have so far been isolated previously from only two species : (B) from *Sarcophyton elegans* and (C) from *L. paciflorum*. This is the first time that both (B) and (C) have been found to co-occur in the same species. It is thus conceivable that probably the former is the biogenetic precursor of the latter.

Experimental

All m.ps are uncorrected. CIMS was recorded on a Biospec instrument with ammonia or methane as reagent gas. Positive CIMS were recorded as their pseudoions, (M + NH₄⁺). Ammonium chloride was used in NCIMS to get (M + Cl⁻) adduct ion. PMR spectra

were obtained on a Bruker WM-200 spectrometer in CDCl_3 using TMS as internal reference. HPLC was carried on a Varian 5010 instrument using a dual cell refractometer detector.

The soft coral was hand picked using SCUBA diving from the coral reefs of Lakshadweep Sea during December, 1985. A reference specimen is on file at our laboratory. Freshly collected specimen (5 kg, wet weight) was soaked in MeOH in carboys and transported to the laboratory. After about one month, the solvent was drained off and concentrated under reduced pressure. The residue (50 g) was partitioned between chloroform and 10% aq. methanol repeatedly. The combined chloroform fractions were dried (Na_2SO_4) and concentrated under reduced pressure. The chloroform fraction was chromatographed over silica gel using increasing amounts of EtOAc in pet. ether to yield low polar (5-20% EtOAc in pet. ether) and more polar (50-100% EtOAc in pet. ether) fractions. The former fraction was further purified on HPLC (ODS-5 u, MeOH, 1.5 ml/min) to yield 6 monohydroxy sterols A1-A6. The more polar fraction was repeatedly purified over silica gel columns to yield baryl alcohol and its lower, C-16 homologue, compounds B and C.

24 ζ -methylcholestane, 3 β , 5 α , 6 β , 25-tetrol, 25-monoacetate (B), m.p. 233°C; IR (nujol): 3450, 1730, 1380, 1250 cm^{-1} ; PMR: δ 4.09 (1H, m, 3-H), 3.54 (1H, br.d, 6-H), 2.0 (3H, s, acetate), 1.3863 (6H, s, 26 and 27-H), 1.1833 (3H, s, 19-H), 0.9157 (3H, d, J = 6.48 Hz, 21-H), 0.8612 (3H, d, J = 6.8 Hz, 28-H), 0.68 (3H, s, 18-H). PCIMS: 526 ($\text{M} + \text{NH}_4^+$), 509 ($\text{M} + \text{H}^+$), 448 ($\text{M} - \text{AcOH}^+$); CMR: δ 32.6 (t, C-1), 34.51 (t, C-2), 67.63 (d, C-3), 41.89 (t, C-4), 76.06 (s, C-5), 76.06 (d, C-6), 34.63 (t, C-7), 30.83 (d, C-8), 45.84 (d, C-9), 39.89 (s, C-10), 22.55 (t, C-11), 40.72 (t, C-12), 42.72 (s, C-13), 55.89 (d, C-14), 24.11 (t, C-15), 28.11 (t, C-16), 55.89 (d, C-17), 12.15 (q, C-18), 16.87 (q, C-19), 36.2 (d, C-20), 18.92 (q, C-21), 30.19 (t, C-22), 27.7 (t, C-23), 38.28 (d, C-24), 85.93 (s, C-25), 23.31 and 22.89 (q's, C-26 & 27), 14.48 (q, C-28), 170.61 (s, acetate carbonyl) and 21.15 (q, acetate methyl).

(24S)-24-methylcholestane-3 β , 4 β , 5 β , 25-tetrol-6-one, 25-monoacetate (lobosterol, C); IR (nujol): 3450, 1730, 1700, 1450, 1380, 1250, 1050 cm^{-1} ; PMR: δ 4.52 (1H, br.d, 3-H), 4.17 (1H, d, 4-H), 2.0 (3H, s, acetate), 1.40 (6H, s, 26 and 27-H), 0.92 (3H, d, J = 6.8 Hz, 21-H), 0.86 (3H, d, J = 6.8 Hz, 28-H), 0.74 (3H, s, 19-H), 0.66 (3H, s, 18-H); CMR: δ 37.86 (t, C-1), 29.66 (t, C-2), 70.22 (d, C-3), 69.79 (d, C-4), 85.61 (s, C-5), 41.83 (t, C-7), 41.15 (d, C-8), 46.48 (d, C-9), 42.79 (s, C-10), 22.52 (t, C-11), 37.86 (t, C-12), 43.16 (s, C-13), 56.81 (d, C-14), 24.15 (t, C-15), 27.86 (t, C-16), 55.74 (d, C-17), 11.95 (q, C-18), 16.84 (q, C-19), 36.06 (d, C-20), 18.85 (q, C-21), 29.47 (t, C-22), 27.67 (t, C-23), 39.3 (d, C-24), 85.88 (s, C-25), 23.32 & 23.94 (q's, C-26 & 27), 170.0 (s, acetate carbonyl) and 21.62 (q, acetate methyl).

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