

STUDIES ON THE STEROLS OF A BROWN SEAWEED *IYENGARIA STELLATA* FROM PAKISTAN

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Abstract

The marine benthic brown alga *Iyengaria stellata* (Børg.) Børgesen, collected from the coast of Karachi was studied for its sterol composition. Analysis of ethyl acetate fraction of the methanolic extract of the seaweed by gas liquid chromatography – mass spectrometry revealed the presence of cholesterol, 24-methyl cholesterol and ergosterol. The last two sterols are being reported for the first time from a brown seaweed.

Introduction

Iyengaria stellata (Scytosiphonales, Phaeophyta) was initially reported from the coast of Karachi as *Colpomenia sinuosa* f. *tuberculata* by Anand (1940) and later by Nizamuddin & Gessner (1970) as *I. stellata*. The morphology, anatomy and ecology of *I. stellata* has also been described (Nizamuddin & Begum, 1978). The chemical constituents of closely related seaweeds like *Colpomenia sinuosa* (Hussein, 1975, Hussein *et al.*, 1983 Parekh *et al.*, 1984), and *Endarachne binghamiae* (Bano *et al.*, 1987) have been studied. *I. stellata* contains 1.27% crude oil, 15.15% of its total fat are sterols (Qasim, 1986). The sterol composition of *I. stellata* is presented here.

Materials and Methods

I. stellata collected from Buleji near Karachi was air dried, (970 g), ground and extracted with ethyl alcohol (EtOH) at room temperature. The extract (9.3 g) in methanol (MeOH) was refluxed with 100 ml 1 N KOH in water for 3 h and the reaction mixture evaporated to remove MeOH, diluted with water and then extracted with diethyl ether (Et₂O). The unsaponifiable lipid (0.5 g) was chromatographed on silica gel in ethyl acetate (EtOAc): *n*-hexane (C₆H₁₄) yielding a fraction of sterols (0.5 mg). The GLC-mass spectrometry of sterol fraction was performed with FINNINGAN-MAT 112 S mass spectrometer coupled with a Varian gas chromatograph equipped with a Sulpeco Sp-2100 glass capillary column (30 m x 0.25 mm i.d.) with carrier gas (N₂) flow of 2 ml/min., at 260°C.

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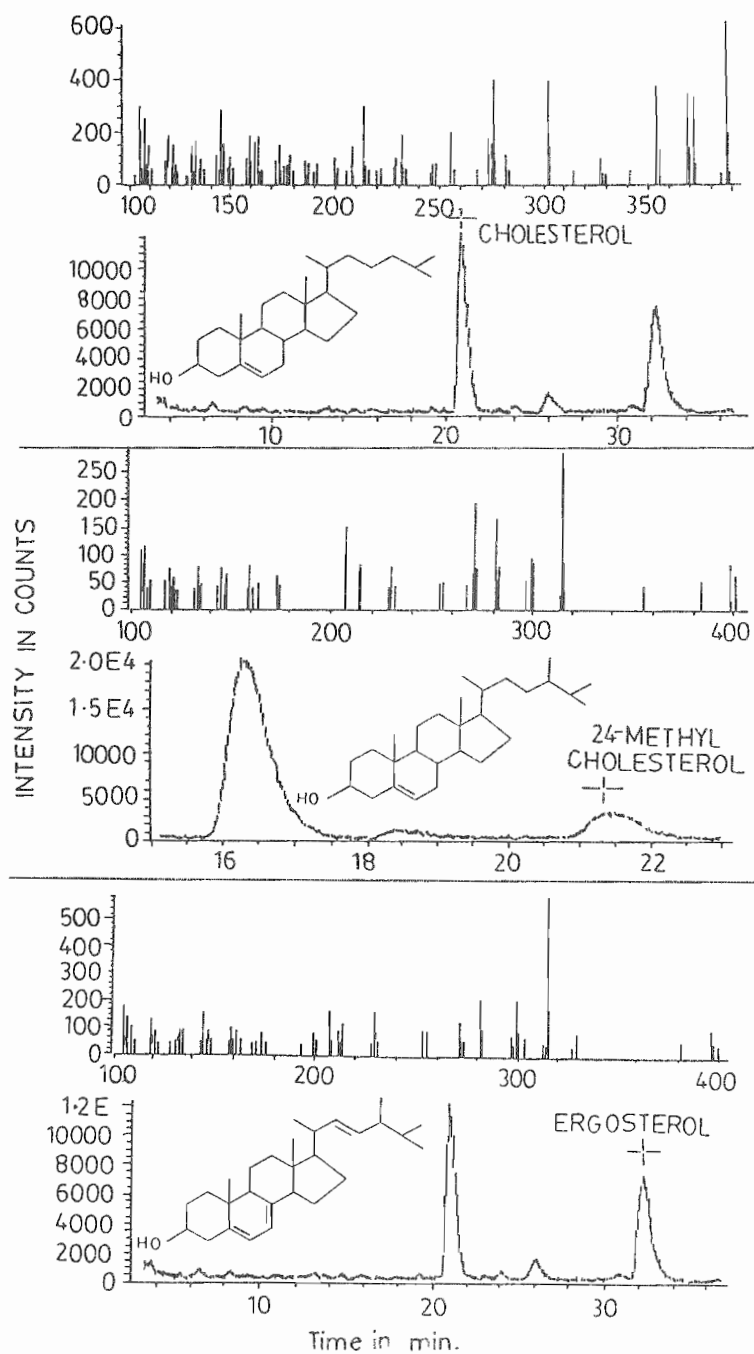


Fig. 1: Gas chromatograms and mass spectra of the sterol fraction of *Iyengaria stellata*.

Results and Discussion

In *I. stellata* the GLC analysis, mass and fragmentation pattern of the sterol fraction indicated the presence of 3 constituents: predominantly a C₂₇ sterol (identified as cholesterol), together with minute quantity of C₂₈ sterol (identified as 24-methylcholesterol) and a small amount of a C₂₈ sterol (identified as ergosterol, Fig. 1). The relative retention time is given in Table 1. The respective identification of sterols through MS is given below:

Cholesterol: MS at m/z (assignments, abundance) 386.45 M⁺ (C₂₇H₄₆O, 622), 371 (M⁺-CH₃, 332), 368 (M⁺-H₂O, 362), 353 (M⁺-CH₃-H₂O, 373), 273 (M⁺-side chain, 168), 255 (M⁺-side chain-C₁₆-C₁₇, 77), 229 (M⁺-side chain-C₁₆-C₁₇-OH, 87), 231 (M⁺-side chain-ring D cleavage, 184), 213 (M⁺-side chain-H₂O-ring D cleavage), 121 (156), 107 (251).

24-methylcholesterol: MS at m/z (assignments, abundance) 400.40 M⁺ (C₂₈H₄₈O, 60), 385 (M⁺-CH₃, 4), 382 (M⁺-H₂O, 19), 367 (M⁺-CH₂-H₂O, 2), 315 (86), 300 (86), 273 (M⁺-side chain, 7), 271 (134), 255 (48), 231 (43), 207 (156), 107 (110).

Ergosterol: MS at m/z (assignments, abundance) 396.25 (C₂₈H₄₄O, 91), 381 (M⁺-CH₃, 46), 325 (M⁺-C₅H₁₁, 61), 314 (M⁺-C₆H₁₀, 583), 300 (M⁺-C₇H₁₂, 83) 271 (M⁺-side chain, 112), 253 (M⁺-side chain-H₂O, 84), 213 (109), 207 (154), 107 (128).

Cholesterol, a major component of Rhodophyta often been reported from brown seaweeds (Goad, 1978), was found as a major component in *I. stellata*. Although, 24-methylene cholesterol has been detected in the thalli of *Laminaria digitata* (Patterson, 1971) and *Enderachne binghamiae* (Bano *et al.*, 1987), but 24-methyl cholesterol has not been reported from any brown alga. In *Pelvetia canaliculata* 24-oxocholesterol has been reported as an artefact (Motzfeldt, 1970), hence methylation at 24th carbon is highly probable.

Table 1. Gas-liquid chromatographic retention time of the sterols of *Iyengaria stellata*.

Trivial Name	Systematic Name	Mol. Formula	Mol. Wt.	Retention Time (min.)
Cholesterol	Cholest-5-en-3 β -ol	C ₂₇ H ₄₆ O	386	20.89
24-Methyl Cholesterol	24-R-Methyl-cholest-5-en-3 β -ol	C ₂₈ H ₄₈ O	400	21.43
Ergosterol	24-R-Methyl-cholesta-5, 7-22-trien-3 β -ol	C ₂₈ H ₄₄ O	396	32.01

Ergosterol is the major component of fungi and higher plants. Although found in certain green and red seaweeds (Goodwin, 1974), it has not so far been reported from any brown alga. *I. stellata* appears peculiar in this regard as it contains small quantity of ergosterol. This would also indicate that *I. stellata* is a distinct taxon from *Colpomenia sinuosa*, with which it has been confused by certain taxonomists.

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(Received for publication 26 March 1987)