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Research Article

STIGMASTEROL, CAMPESTEROL AND BRASSICOSTEROL FROM *HIBISCUS SABDARIFFA* SEED

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(Malvaceae).

ABSTRACT

The *Hibiscus sabdariffa Linn* (Malvaceae) has been used in folk medicine as a diuretic mild laxative and treatment for cardiac and nerve diseases. The seed of *Hibiscus sabdariffa* and a conventional extraction and a sequence of chromatographic methods afforded stigmasterol, campesterol and brassicosterol was isolated from *Hibiscus sabdariffa* seed.

INTRODUCTION

The main aim of the work done by the author was to carry out the phytochemical and pharmacological screening of anti-inflammatory activity of the plant *Hibiscus sabdariffa* (Malvaceae), claimed to be used traditionally in the treatment of various ailments including rheumatoid arthritis, anti-depressent, parkinson's disease, infertility, stomachic and emollient antiseptic, purgative, refrigerant, resolvent, sedative, stomachic, and tonic, roselle is a folk remedy for abscesses, bilious conditions, cancer, cough, debility, dyspepsia, dysuria, fever, hangover, heart ailments, hypertension, neurosis, scurvy, and strangury [1-5].

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However, Literature survey indicated hepatoprotective activity, anti-inflammatory and anti-ulcer activity [6-8]of leaves and anti-fertility activity on male rats. In the view of this the author aimed to study the isolation of pure molecules from methanolic *Hibiscus sabdariffa* seed extract by using column chromatography.

MATERIAL AND METHODS

1.1. Plant material collection and authentication

The plant material of *Hibiscus sabdariffa* was collected Annapurna agro agencies, sabbavaram, Andhra Pradesh, India in December 2014. The plant species was authenticated by Dr. Bodaih Padal, taxonomist, department of botany, Andhra university, Visakhapatnam. The voucher specimens (21921) were deposited in the herbarium, college of pharmaceutical sciences, Andhra university.

1.1.1 Soxhlet extraction Method:

The seeds of *Hibiscus sabdariffa* (1.5 kg) were air dried and coarsely powdered in a wiley mill and successively extracted with methanol for 19hrs in a soxhlet extraction method. And concentrated under reduced pressure at 70° C. The methanolic extract showed positive Liebermann- burchard test for triterpenoids. On column chromatographed over silica gel (100-200 mesh) the methanolic extract showed three compounds namely stigmasterol, campesterol, brassicosterol .

RESULTS AND DISCUSSION

stigmasterol (Compound HSB-01):

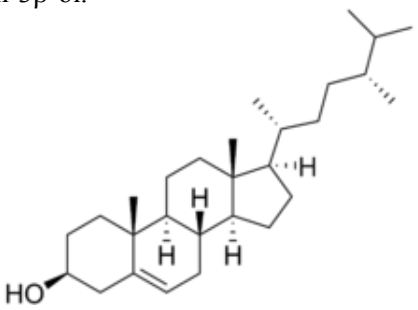
The compound was obtained from eluates 8-13 of column chromatogram of crude extract residue. These eluates were combined as they showed single spots in TLC and crystallized as feathery needles from 99% hexane: ethyl acetate. Melting point : 160-164^{0}C. It gave positive with Liebermann-burchard test. Its identity was further confirmed by comparison with an authentic sample through Co-TLC, IR and mass spectrum. The IR absorption spectrum showed absorption peaks at 3373.6cm-1 (O-H stretching.); 2940.7 cm-1and 2867.9cm-1 (aliphatic C-H : stretching); 1641.6cm-1 (C=C absorption peak); other absorption. peaks includes 1457.3cm-1 (CH2); 1381.6cm-1 (OH def), 1038.7cm-1 (cycloalkane) 881.6 cm^{-1} IUPAC NAME (3*S*,8*S*,9*S*,10*R*,13*R*,14*S*,17*R*)-17-[(*E*,2*R*,5*S*)-5-ethyl-6-methylhept-3-en-2-yl]-10,13-dimethyl-2,3,4,7,8,9,11,12,14,15,16,17-dodecahydro-1*H*-cyclopenta[*a*]phenanthren-3-ol.

Campesterol (Compound HSB-02)

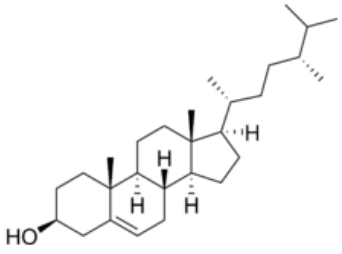
The compound was obtained from eluates 27-31 of column chromatography of crude extract residue. These eluates were combined as they showed single spots in TLC and crystallized as needle shaped crystals from 97% hexane: ethyl acetate. Its melting point was found to be 156-160^{0}C. It showed positive reaction for Liebermann burchard test. Its identity was further confirmed by comparison with an authentic sample through Co-TLC, IR and mass spectrum. IUPAC NAME: (3*S*,8*S*,9*S*,10*R*,13*R*,14*S*,17*R*)-17-[(2*R*,5*R*)-5,6-dimethylheptan-2-yl]-10,13-dimethyl-2,3,4,7,8,9,11,12,14,15,16,17-dodecahydro-1*H*-cyclopenta[*a*]phenanthren-3-ol.

brassicosterol (Compound HSB-03)

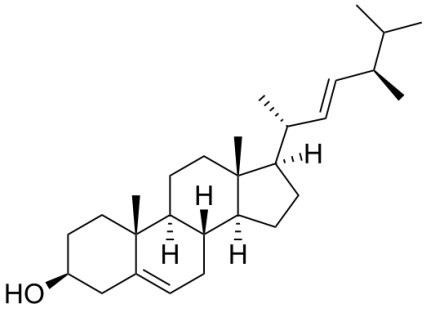
The compound was obtained from eluates 38-39 of column chromatography of crude extract residue. These eluates were combined as they showed single spots in TLC and crystallized as needle shaped crystals from 96% hexane: ethyl acetate. Melting point : 150^{0}C; LB reaction: violet-blue-green; Salkowski reaction: reddish brown; R_f value: 0.47. The compound was crystallized from 96% hexane: ethyl acetate as white crystalline powder. Its melting point was found to be 150^{0}C. It gave play of colors with Liebermann- burchard test. Its identity was further confirmed by comparison with an authentic sample through Co-TLC, IR and mass spectrum. IUPAC NAME: (3β,22E)-ergosta-5,22-dien-3-ol 24β-methylcholesta-5,22-dien-3beta-ol-ergosta-5,22-dien-3β-ol.



Stigmasterol

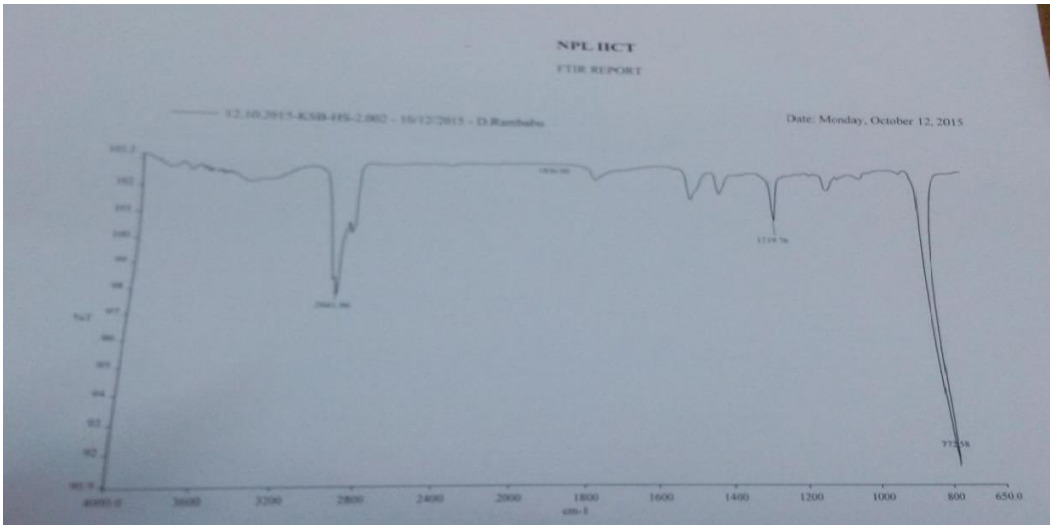


Campesterol

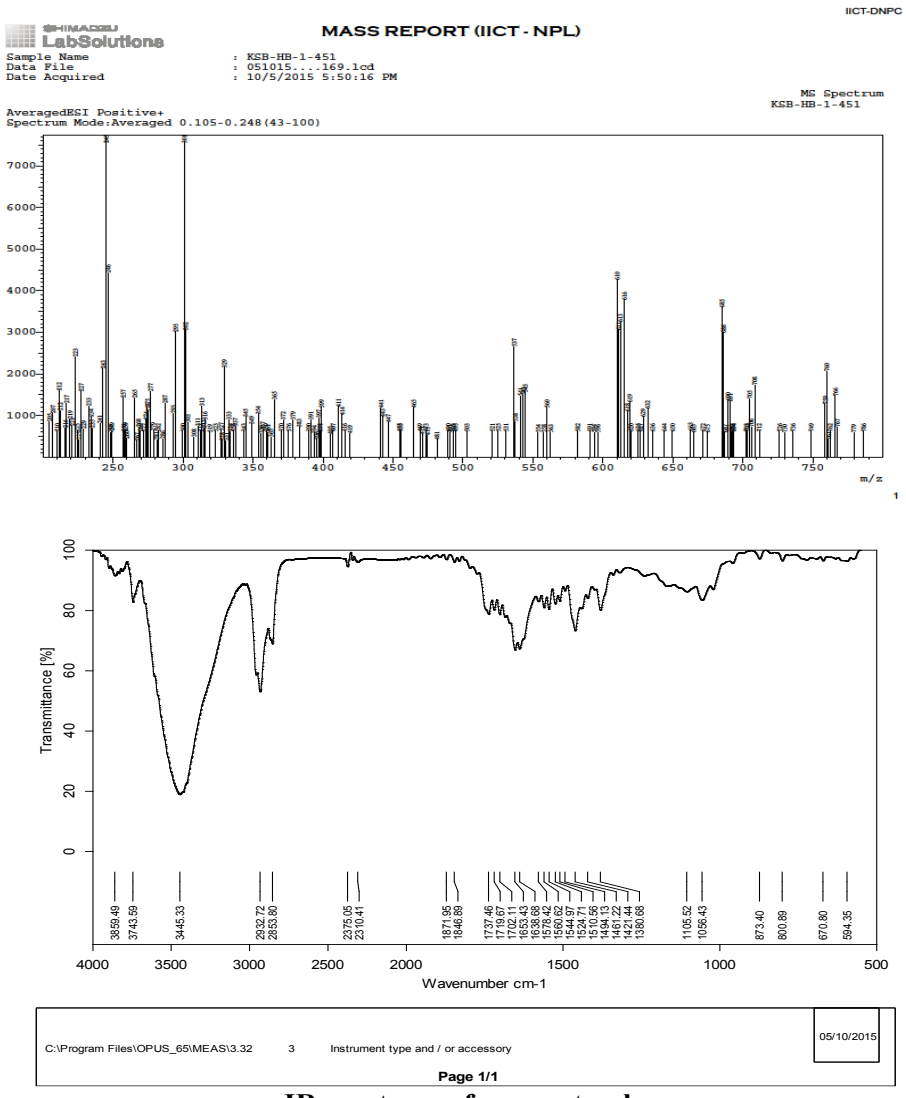


Brassicosterol

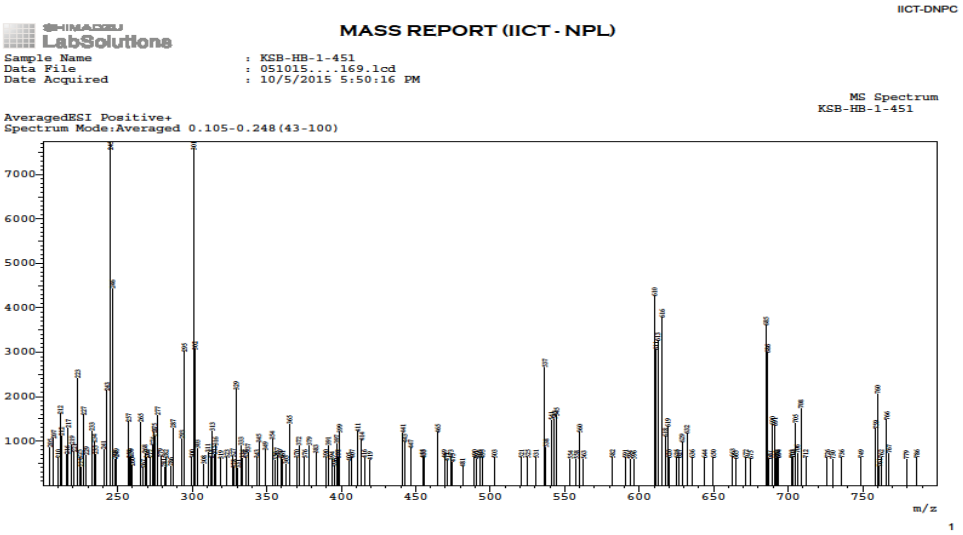
IR spectrum of stigmasterol



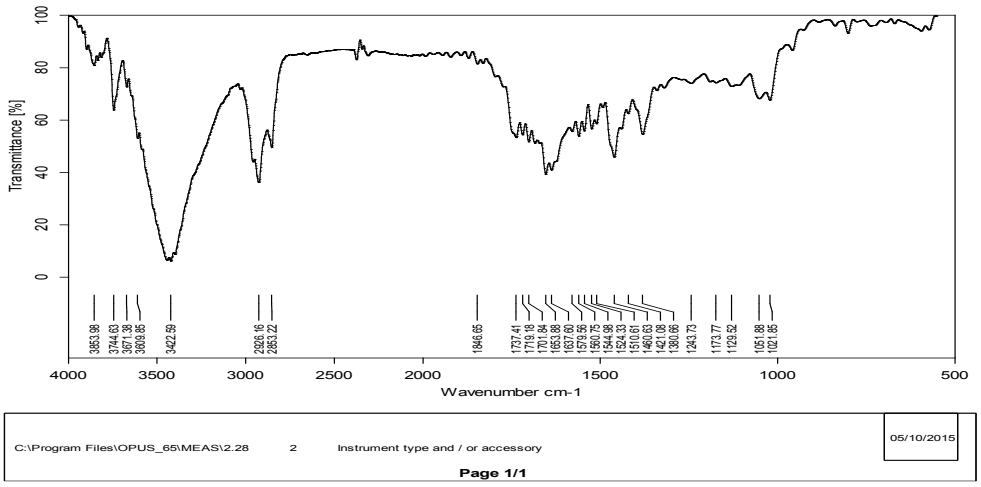
Mass spectrum of Stigmasterol



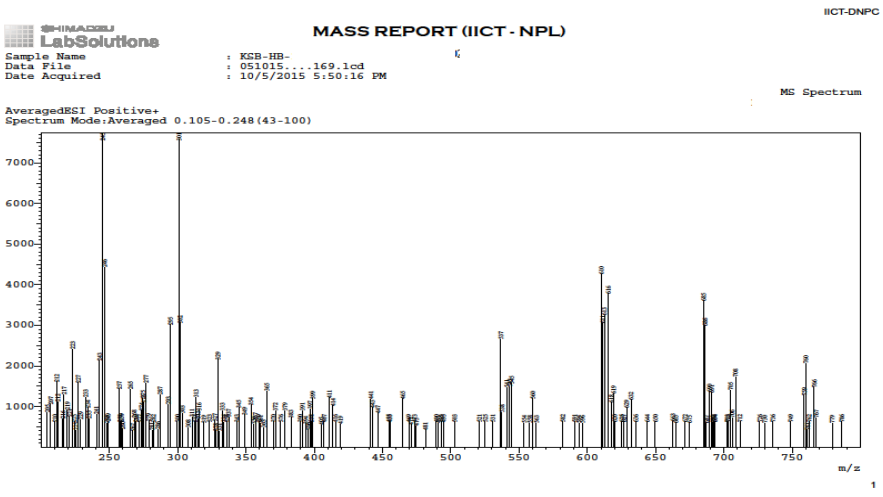
IR spectrum of campesterol



Mass spectrum of campesterol



IR spectrum of brassicosterol



Mass spectrum of Brassicosterol

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