Steroid; β- Sitosterol from Corydalis govaniana (Wall) growing Himalayan region of Nepal

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Abstract

The Himalayan plant Corydalis govaniana (Wall) commonly known as Bhutkesi in Hindi on detail chemical investigation led to the isolation of β -sitosterol and its structure was determined by detail spectroscopic analysis.

Keywords: Corydalis.Fumariaceae,Steroid.

Introduction

The genius Corydalis plant species, belonging to the family Fumariaceae, are represented by more than 20 species in Nepal¹. They occur mostly in the northern hemisphere, particularly in the temperate, sub-alpine and alpine zones between 2500-4500m of Asia.

Among these species, it has been studied to some extent, and all have been found to produce isoquinoline alkaloids ². *Corydalis govaniava* Wall (Fumariaceae) is a stout herb native to the western Himalayas at altitudes in the range of 2500 to 3500m. Ethno medically, the roots have been found use by the people in the treatment of syphilitic, scrofulous, and cutaneous infections^{3,4}. The alkaloidal fraction acts as a uterine stimulant and stomach muscle depressant in Vitro and vivo injection of the alkaloid fraction produces a hypertensive effect in anesthetized cats⁵.

Previous phyto-chemical investigation of *Corydalis govaniana* led to the isolation of number of isoquinoline alkaloids^{6,7,8}.

Experimental

The melting point was determined on a Toshniwal apparatus and was unconverted UV spectrum was recorded with Perkin Elmer Lambde spectrometer using spectral method. An IR spectrum was recorded in K Br Pellets, ¹HNMR spectra were recorded in 500 MHz in deuterated Acetone using tetramethyisilane(TMS) as internal reference. Mars spectrometer was performed on JEOL MS Route spectrometer operating at 70 ev. The purity of substance was checked on TLC plates.

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J. Nepal Chem. Soc., vol. 29, 2012

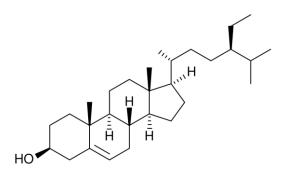


Figure 1: β -sitosterol

dodecahydro-1H-cyclopenta[a]phenanthren-3-ol

Plant material

The plant material used in this study was collected from Langtang, Himalayas of Nepal and identified by comparison with the authentic herbarium specimen at the National Herbarium Laboratory, Kathmandu, Nepal.

Extraction and Fractionation

Air-dried arial part of *Corydalis govaniana* (5kg) was extracted with methanol for seven days in cold. Percolator.

After removal of methanol under reduced pressure, the residue (500 g) was treated with 7% citric acid and separated to alkaloidal fraction using the procedure devised by R.N.Jha et. al⁹. The fraction obtained using above procedure was analyzed by TLC for alkaoid by spraying with

Dragendorff's reagent. The chloroform extract (62g) was chromatographed over silica-gel column using solvent of increasing polarity. The eluants from 5% acetone in Hexane on crystallization from methanol yield 15mg of steroid; β - sitosterol.

B-Sitosterol; white crystalline; M.P. $137-13^{\circ}$ C, λ max in MeOH; 206 nm. IR absorptions bands appeared at 3549 cm⁻¹. 2(OH), 2935 cm⁻¹ (CH); 1637cm⁻¹ (C=C), 1663 cm⁻¹ (C=O), ¹HNMR (in Table 1 and fig.3), ms (m/z), 414.3, 460, 381.5, 329, 303, 273, 255, 231, 213, 161, 145, 107, 95, 81, 55, 43 (Fig.2).

Results and Discussion

Steroid; β –sitosterol was isolated from the chloroform extract of *Corydalis govaniana* using usual method of isolation. The molecular formula of compound based on the high resolution mass spectrum was found to be C₂₀H₅₀O; m/z 4143(m+), 400, 381, 324, 303, 289, 273, 255, 231, 213, 199, 173, 161, 145, 121, 107, 95, 81, 69, 55, 43. A fragmentation pattern indicative of β –sitosterol could be easily rationalized from reported data¹⁰.

J. Nepal Chem. Soc., vol. 29, 2012

The ultraviolet spectrum in MeOH showed absorption maxima at 206nm like that of steroid (11). The IR spectrum contained at absorbance at 2349 cm-1(OH), 2935cm-1 (CH₂), 2867 cm-1 (CH), 1637 cm-1 (C=C), 1063 cm-1 (C=O) clearly suggested the isolated compound was β –sitosterol. Further the structure was established by ¹HNMR spectrum of this compound which resembled with the data published in literature¹¹⁻¹⁴.

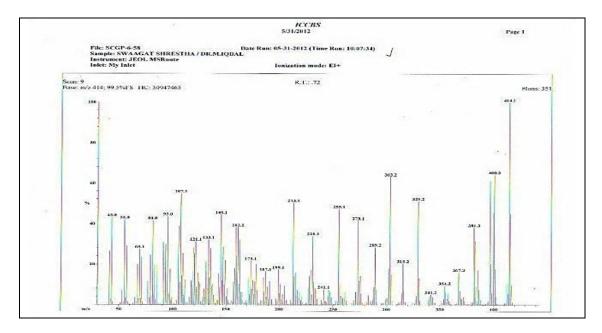


Figure 2: Mass Spectra

Chemical Shift (δ)	Proton count	Probable assignment
1.47	2H	С-1-Н
1.56	2H	С-2-Н
3.61	1H	С-3-Н
2.22	2H	С-4-Н
-	-	C-5-
5.30	1H	С-6-Н
2.03	2H	С-7-Н
1.67	1H	С-8-Н
1.48	1H	С-9-Н
-	-	C-10-
1.53	2H	С-11-Н
1.48	2H	С-12-Н
-	-	C-13-
1.50	1H	С-14-Н
1.59	2H	С-15-Н

Table -1: 560 MHz¹HNMR spectral data of β -sitosteral in Acetone.

1.84	2H	С-16-Н
1.50	1H	С-17-Н
0.78	3Н	С-18-Н
1.01	3Н	С-19-Н
1.66	1H	С-20-Н
0.95	3Н	С-21-Н
0.87	2H	С-22-Н
1.05	2H	С-23-Н
1.48	1H	С-24-Н
1.50	1H	С-25-Н
0.83	3Н	С-26-Н
0.85	3Н	С-27-Н
1.04	2H	С-28-Н
0.88	3Н	С-29-Н
2.00	1H (OH)	С-30-Н

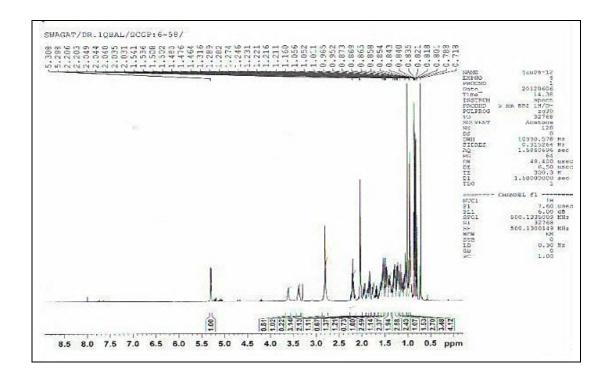


Figure3: ¹HNMR Spectra

Conclusion

The structure of β –sitosterol was established by physical, chemical and spectroscopic methods as well as comparison of its spectral data with those in the literature and direct comparison with authentic sample. This is the first report of occurrence of β –sitosterol in *Corydalis govaniana*.

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