



Asian Journal of Pharmaceutical Analysis and Medicinal Chemistry

Journal home page: www.ajpamc.com



ISOLATION AND CHARACTERIZATION OF β - SITOSTEROL FROM THE LEAVES OF *DIPTERACANTHUS PROSTRATUS NEES*

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ABSTRACT

Dipteracanthus prostratus nees (Acanthaceae) commonly known as Pottakanchi in Tamil contains a number of phytoconstituents viz, alkaloids phytosterols, glycosides, proteins, amino acids, flavonoids and terpenoids. The objective of the present with solvent study was to isolate and characterize phytoconstituents from the chloroform extract of *Dipteracanthus prostratus nees* leaves. Chloroform extract was subjected to column. Chromatography and eluted mixtures of increasing polarity composed of benzene and ethylacetate to isolate phytoconstituents. The structure of the isolated compound was established on the basis of elemental analysis and spectroscopic evidences (IR, UV, ¹HNMR, ¹³CNMR, MS) A β -sitosterol was isolated from the chloroform extract of the leaves of the plant. The yield of compound was 0.056% w/w m.p 136⁰C to 138⁰C. λ max in EtOH: 212nm, Rf value 0.4 in {toluene: ether: cyclohexane} (5:2:1) *Dipteracanthus prostratus nees* contains β -sitosterol which may be responsible for various pharmacological activities of the plant.

KEYWORDS

Dipteracanthus prostratus nees, β -sitosterol, Acanthaceae and Chloroform extract.

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INTRODUCTION

Dipteracanthus prostratus nees (Acanthaceae) is a prostrate perennial herb. It contains a number of phytoconstituents viz alkaloids, phytosterols, glycosides, amino acids, phenolic acid, enzymes, vitamins, sugar, flavonoids, gums and mucilage, terpenoids¹ etc. The plant is used for the treatment of a number of disease². The present study deals with isolation and characterization of β -sitosterol from the leaves of *Dipteracanthus prostratus nees*.

MATERIALS AND METHOD³⁻⁷

Collection and authentication of the plant material

D.prostratus plants were collected from Shervaroyan Hills, Salem, Tamilnadu. The plant was authenticated by Botanist Dr. A. Balasubramanian (ABS Botanical Conservation, Research and Training center) Siddha Research consultant (Ayush). The leaves were separated from the plant dried under shade.

Extraction and isolation of compound

The shade dried and coarsely powdered leaves were successively extracted with petroleum ether, chloroform and ethanol using Soxhlet apparatus. Chloroform extract was chromatographed on a silica gel column and eluted with solvent mixtures of increasing polarity, composed of petroleum ether, benzene and chloroform. All the fractions were monitored on TLC. Fractions collected petroleum ether: benzene (30:70) was pulled together these fractions showed a single spot of same R_f value in TLC. It was evaporated in water bath (50-60°C) to offer a solid residue. The residue was dissolved in a mixture of Chloroform: Ethanol (40:60) with little warming on a water bath. It was left undisturbed in refrigerator when needle shaped crystals of β -sitosterol was obtained. The structure of the isolated compound was established on the bases of spectroscopic evidences (IR, UV, ¹H-NMR, ¹³C-NMR, Maaas).

RESULTS AND DISCUSSION

Characterization of the compound

Phytochemical analysis (Salkowski's test and Libermann-Burchad test) of the compound confirmed its steroidal nature. It is assumed to be a compound containing steroidal nucleus. The β -

sitosterol is white crystalline needles like substances with melting point 136-138°C. (Figure No.1) λ_{max} in ethanol: 212nm. (Figure No.2) IR spectroscopic analysis, the absorption bands are 3373.5cm⁻¹ that is characteristic of O-H stretching. Absorption at 2924.3 and 2853.9 is due to aliphatic (C-H) stretching other absorption frequencies include 1615.2cm⁻¹ as a result (C=C) stretching however this band is weak, at 1451.2cm⁻¹ is a bending frequency of cyclic (CH₂). The absorption frequency at 1038cm⁻¹ signifies cycloalkene. The absorption frequencies resemble the absorption frequencies observed for β -sitosterol.

The proton NMR showed (FigureNo.3) the proton singlet at δ 3.32(C-H) revealed the existence of signals for olefinic proton δ 5.18 (singlet), 4.56 (multiplet), 2.83 (multiplet). Angular methyl proton at 0.69(s) 0.80(s), 1.02(s) corresponding to C₁₈ and C₁₉ proton respectively (FigureNo.4). The ¹³C-NMR has shown recognizable signals 150.77 and 140.7ppm, which are assigned C₅ and C₆ double bonds respectively. The value of angular methyl group (C₁₉) spectra show twenty nine carbon signal including six methyl, nine methylene, eleven methane and three quaternary carbons. The alkene carbons appeared at δ 150.77, 140.70, 121.48 and 118.89.

(FigureNo.5) Mass spectra of this compound suggested that its molecular mass is 414(M.F-C₂₉H₅₀O) having characteristic fragments observed at M/Z 414, 396, 381, 329, 303, 289, 273, 255, 231, 213, 199, 173, 159, 145, 119, 95, 81, 69, 55. The above IR, ¹H-NMR, ¹³C-NMR and MS spectral data and their comparison with those described in literature showed the structure of β -sitosterol confirmed.

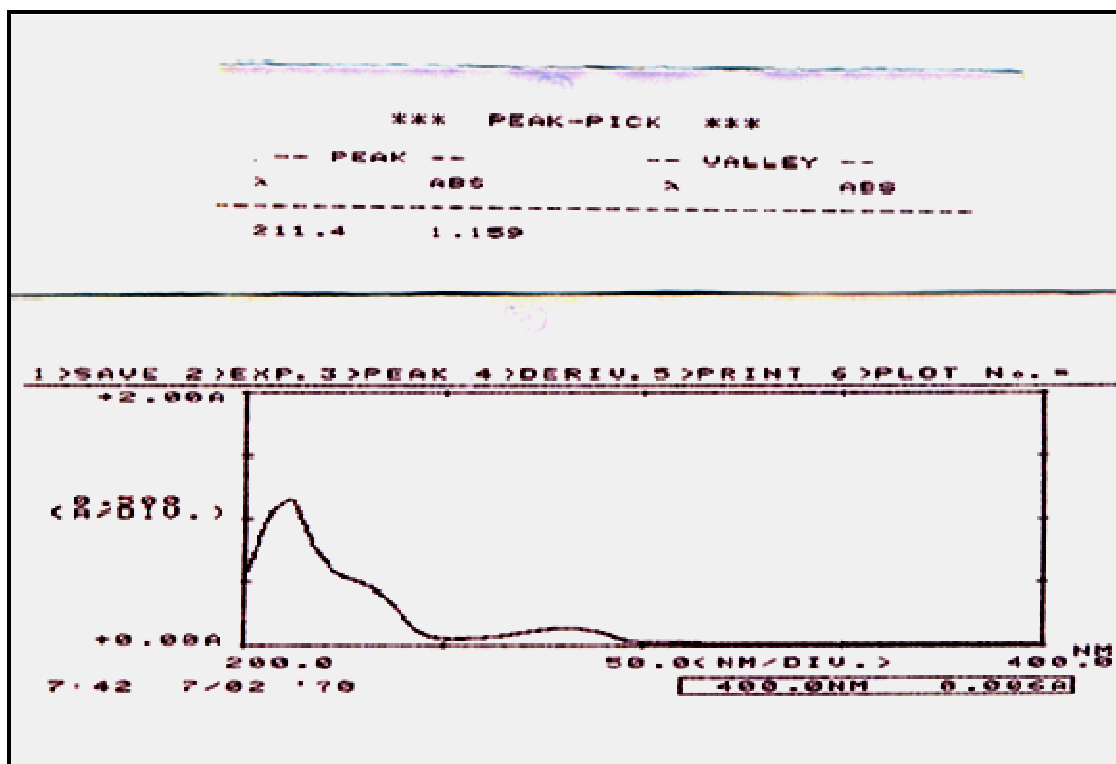


Figure No.1: UV spectral maxima of β -Sitosterol

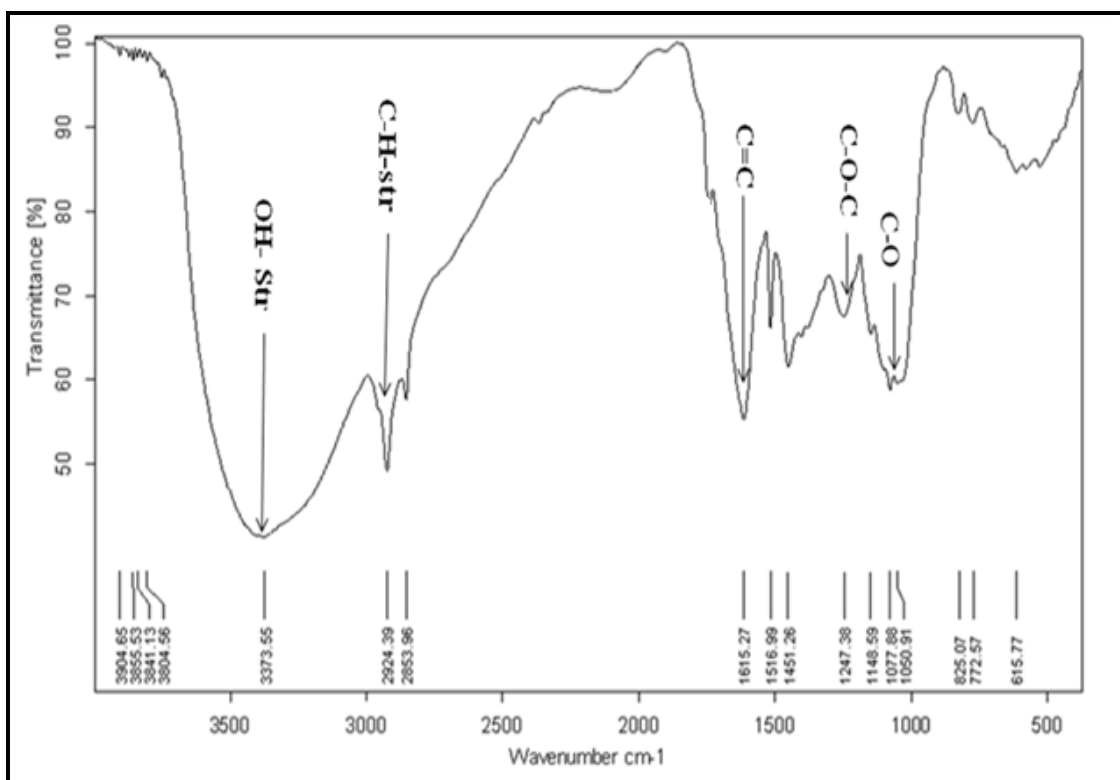


Figure No.2: FT-IR Spectrum of β -Sitosterol

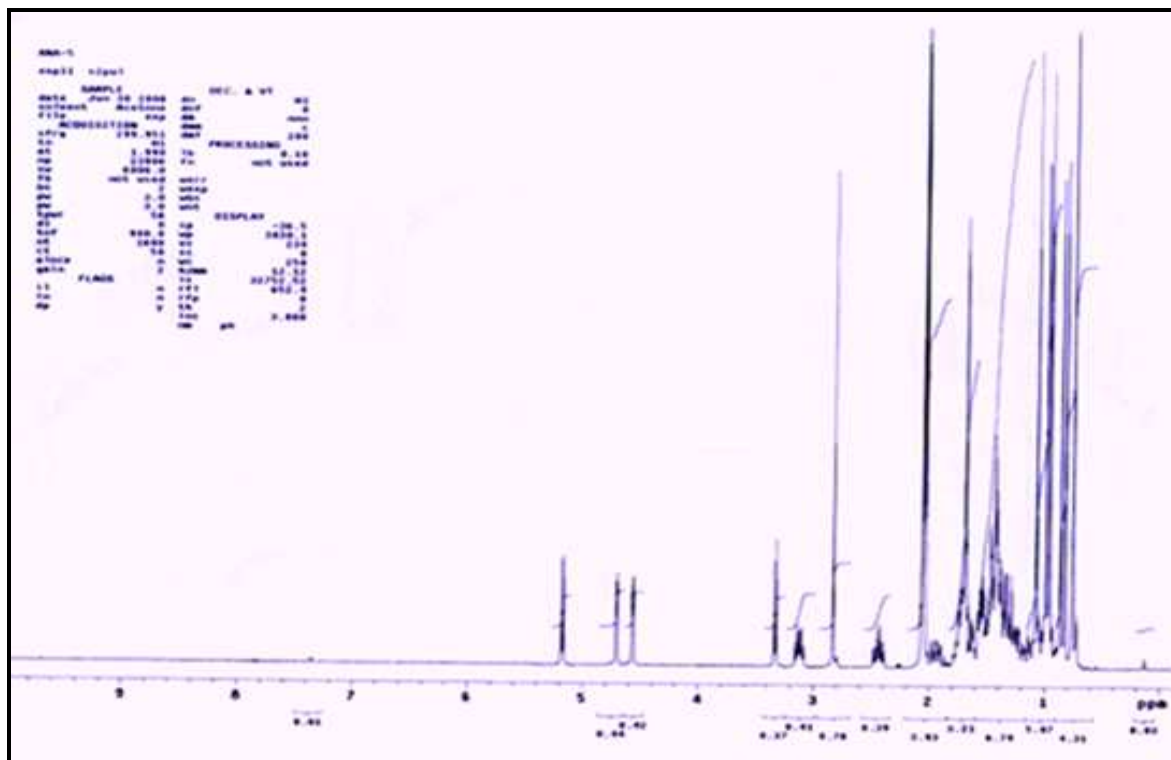


Figure No.3: ^1H NMR Spectrum of β -Sitosterol

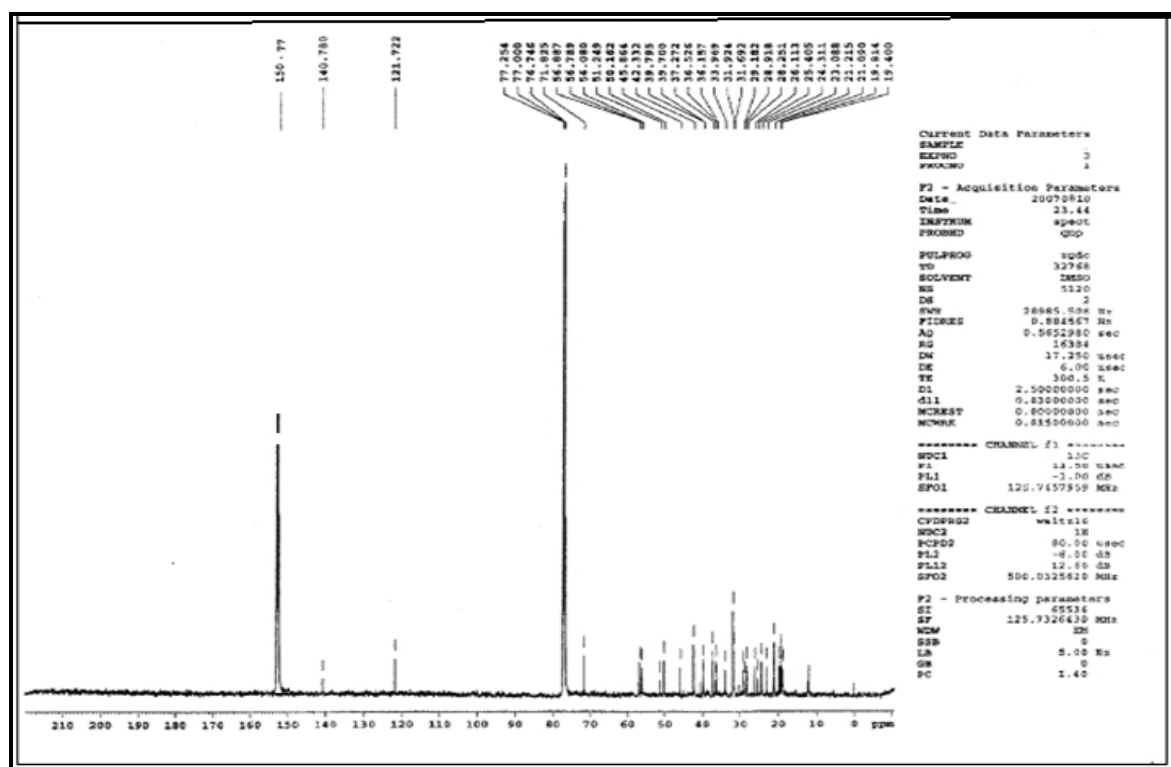


Figure No.4: ^{13}C NMR Spectrum of β -Sitosterol

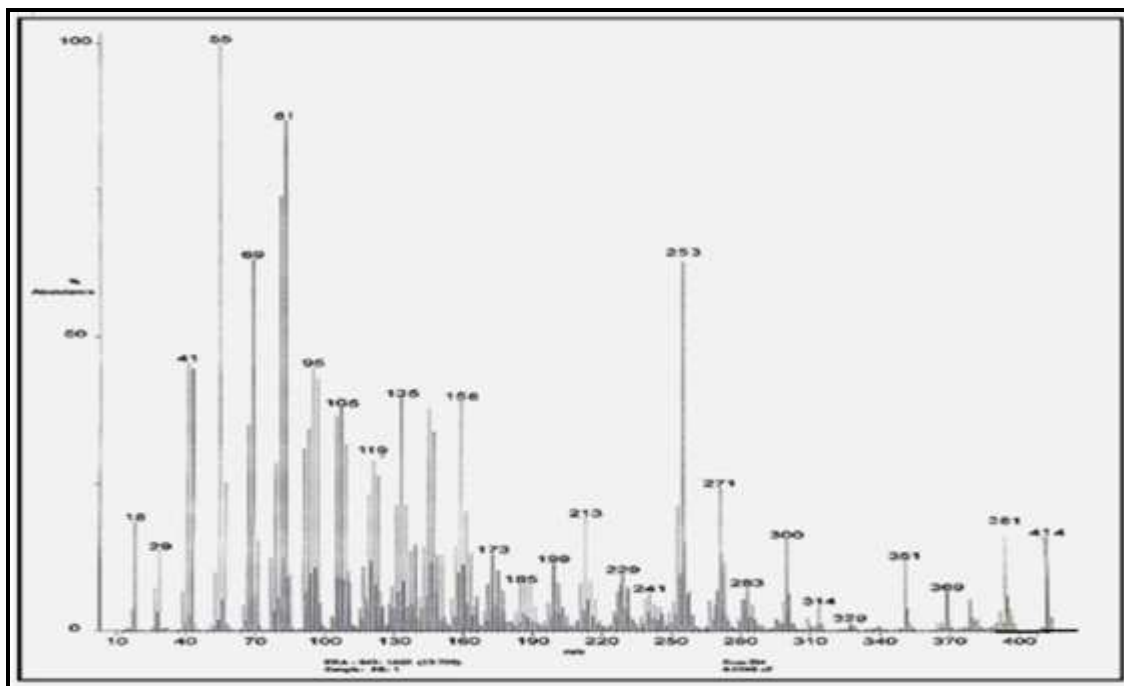


Figure No.5: Mass Spectrum of β -Sitosterol

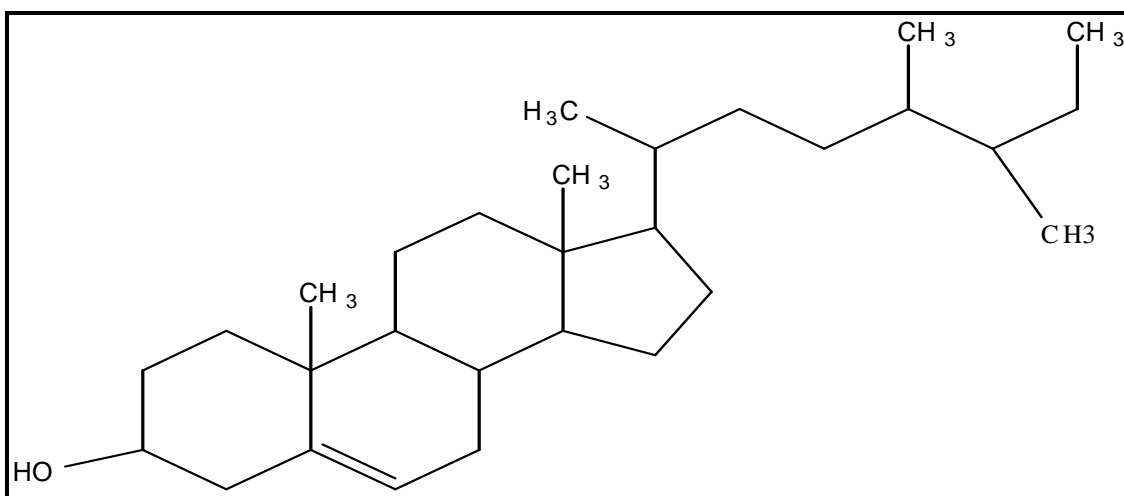


Figure No.6: Structure β - Sitosterol

The isolated compound may be β - Sitosterol (Figure No.6).

CONCLUSION

β - Sitosterol was isolated and characterized from chloroform extract of *D. prostratus* leaves and this is a phytosterol. β -Sitosterol reduces carcinogen-induced cancer of the colon. It shows anti-inflammatory, antipyretic, anti-arthritic, anti-ulcer, insulin releasing and oestrogenic effects and

inhibition of spermatogenesis. β - Sitosterol is mainly known and used for its cholesterol lowering property. But studies have shown that the photochemical may have other health benefits easing symptoms of benign prostatic enlargement, reducing risk of cancer and prevention of oxidative damage through its anti oxidant activity.

ACKNOWLEDGEMENT

A special acknowledgement is owed to Dr. B. Jaykar professor and principal of Vinayaka Mission's College of Pharmacy, Salem, India for his tremendous help, encouragement, timely advice and facilities.

CONFLICT OF INTEREST

We declare that we have no conflict of interest.

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Please cite this article in press as: J. Banu Rekha and B. Jaykar. Isolation and Characterization of β -Sitosterol from the Leaves of *Dipteracanthus Prostratus Nees*, *Asian Journal of Pharmaceutical Analysis and Medicinal Chemistry*, 3(2), 2015, 76 - 81.