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Isolation and Characterization of Friedelin from Azima TETRACANTHA Lam Roots

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ABSTRACT

Azima tetracantha Lam., commonly known as Mulsangu or Chulli in India, is a medicinal plant belonging to the Salvadoraceae family. This spiny shrub is native to South Asia, Africa, and Sri Lanka and is widely used in traditional medicine, including Ayurveda. The plant is valued for its numerous medicinal properties, such as anti-diabetic, antimicrobial, antioxidant, diuretic, and antirheumatic activities. The focus of this research is to isolate the phytochemical friedelin from the powdered dried root of the plant, which is believed to contribute to its anti-diabetic, anti-inflammatory, and antioxidant properties. The roots were air-dried and extracted using a polarity gradient of n-hexane, chloroform, and ethanol. Purification of the methanol extract through column chromatography resulted in the isolation of friedelin. Its structure was confirmed through spectral analysis. The study aims to support the traditional use of Azima tetracantha in treating various ailments by validating the bioactive components responsible for its therapeutic effects.

Keywords: ***Azima tetracantha Lam***, friedelin, Phytochemical isolation, Anti-diabetic properties, Column chromatography

INTRODUCTION

India has an ancient heritage of traditional herbal medicine. Materia medica of India provides lots of information about the folklore practices and traditional aspects of therapeutically important natural products. Indian traditional medicine is based on various systems including Ayurveda,Siddha and Unani.The evaluation of these herbal drugs is mostly based on phytochemical, pharmnacological and allied approaches including various instrumental techniques like chromatography, microscopy and others[1].

Azima tetracantha is a low, spinous, rigid bush, woody at the base, with numerous green, herbaceous branches that are bluntly quadrangular and pubescent when young, has leaves measuring 2.5-4.5 cm in length[2]. Azima tetracantha is a versatile medicinal plant traditionally used in Indian medicine . Its root, root bark, and leaves are consumed with food as a remedy for rheumatism, and it serves as a powerful diuretic for treating conditions like rheumatism, dropsy, dyspepsia, and chronic diarrhea. It is also used as a stimulant tonic after childbirth. Known for its acute anti-inflammatory properties, Azima tetracantha is employed to treat cough, phthisis, asthma, smallpox, and diarrhea. A decoction of the stem bark is considered astringent, expectorant, and antiperiodic[3].

MATERIALS AND METHODS

Experimental procedure:

Distilled n- hexane, chloroform (CHCl3), and ethanol (EtOH) were used as solvent to extract the plant root. n- hexane and ethyl acetate were used as solvent systems in the chromatographic method .A cold maceration technique was used for the extraction of the sample. Gravity column chromatography (CC) was carried out using Merck silica gel 60 (70-230 mesh). Thin layer chromatography (TLC) was performed on 0.20 mm precoated silica gel aluminium sheets (Merck Kieselgel 60 F254). Spots were visualised with UV light (254 nm and 365 nm) and exposed to the iodine crystal. The 1H NMR (400 MHz)) spectra were recorded on a Bruker Avance 400 spectrometer. Chemical shifts were recorded in ppm relative to tetramethylsilane (TMS) in deuterated chloroform (CDCl3). The infrared spectra were measured using ATR-FTIR. The melting point was measured on a Leica Gallen III Kofler micromelting point apparatus and was uncorrected.

Plant Material

The roots of *Azima tetracantha* were collected from local area in Tirunelveli district. The plant material was identified and authenticated by Mr.D.Chelladurai, Research officer -Botany,C.C.R.A.S ,Govt of India,(Retired), Thyagaraja Nagar,Tirunelveli-627011.A voucher specimen was submitted at C.L. Baid Metha College of Pharmacy, Chennai.

Extraction and Isolation of Compounds

The powdered material was charged in an aspirator bottle and successively extracted with hexane,chloroform,ethanol by cold maceration method for 72 hrs.After decantation and filtering, nearly 80% of the solvent was removed by distillation over boiling water bath remaining under reduced pressure [4].TLC of the extracts where done using solvent system hexane:ethyl acetate(6:4).

The ethanolic extract gave clear distinguishable spots and thus the extract further purified over a silica gel column (150 g silica gel; column size 90×20 cm) .The column was eluted with hexane and ethyl acetate starting from the ratio100:0 to 0:100. 30 fractions (1-30) were collected and each fraction was subjected for preliminary phytochemical screening[5]. The fractions from10 to 30 gave positive reactions for triterpenoids and these fractions were subjected to TLC analysis by using solvent system developed by trial and error method – ethyl acetate:methanol (9:1)[6].

Similar spots with same R_f value was observed for the fraction from 10 to30 and based upon the results ,the fraction from 10 to 30 were pooled and this pooled fraction was dried in an oven at and this gives white powder. It was recrystallised from methanol.The solid melted between 245-246°C with an Rf value of 0.43 ethyl acetate:methanol (9:1).and it was subjected to spectral studies and the compound was named as AT.

Spectral Data of isolated compound AT

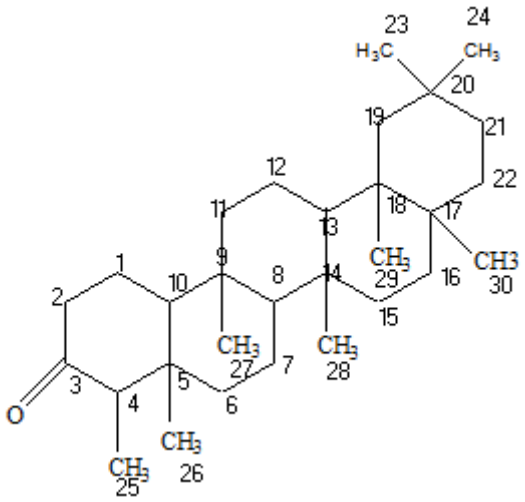
IR Spectrum shows spectral bands at 2930-2875cm^{-1} (C-H stretching)and 1715 cm^{-1}(C=O KETONE). The ^{1}H NMR reveals signals for seven singlet of methyl signals at δ 1.17(H-28),1.04(H-27),1.01(H-26),1.00(H-30),0.95(H-29),0.86(H-25),0.72(H-24),a doublet of methyl proton at δ 0.89(H-23),a methine proton of CH at δ2.25 (H-4)respectively. There is no vinylic signal observed.

13C NMR (100 MHz, CDCl3): δ (ppm) 22.2 (C-1), 41.5 (C-2), 213.2 (C-3), 58.1 (C-4), 42.1 (C-5), 41.3 (C-6), 18.2 (C-7), 53.1 (C-8), 37.5 (C-9), 59.5 (C-10), 35.6 (C-11), 30.6 (C-12), 39.6 (C-13), 38.3 (C-14), 32.4 (C-15), 35.6 (C-16), 30.1 (C-17), 42.8 (C-18), 35.3 (C-19), 27.9 (C-20), 32.6 (C-21), 39.1 (C-22), 6.8 (C-23), 14.6 (C-24), 18.0 (C-25), 20.2 (C-26), 18.5 (C-27), 31.9 (C-28), 35.0 (C-29), 31.8s (C-30); EIMS *m/z* (%): 399 (17), 327 (42), 281 (83), 147 (58), 73 (100). EIMS *m/z* (%):

96.05(100%),426.38[M^+](29%),302.26(8%),274.22(16%),273.22(30%)246.18(25%),218.17(16%),205.16(40%),191.15(26%),179.15(12%),164.12(15 %),125.10(10%),123.08(66%),121.07(26%),109.07(12%),81.04(30%),69.04(16%),55.00(21%).

RESULTS AND DISCUSSION

The powered root of Azima tetracantha.Lamm was cold macerated with with hexane,chloroform,ethanol . The ethanol extract was selected for the column chromatography to isolate an active component , AT and was identified as friedelin from spectral analysis.



The IR spectra of the isolated compound gives spectral bands at 2930-2875 cm^{-1} (C-H strectching) and 1715 cm^{-1} (C=O ketone).The NMR spectrum shows the presence of the angular and side chain methyl groups in the molecule. The ^{1}H NMR reveals signals for seven singlet of methyls, a doublet of methyl and a methane proton respectively.There is no vinylic signal observed. The mass spectra of the component isolated was also done, It shows a parent molecular ion peak [M^+] at m/z 426.3 which corresponds to molecular formula $C_{30}H_{50}O$.From the spectral data, we found that the compound may be similar to the triterpenoid friedelin.

CONCLUSION

From the results obtained, it can be concluded that the root of Azima tetracantha.Lam contains friedelin,a triterpenoid chemical compound ,which is responsible for many of the medicinal properties of the plant Azima tetracantha.

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