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Phytochemical, FTIR and NMR analysis of crude extract of *Acacia planifrons* seeds

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Abstract

Acacia planifrons is commonly known as kudai vel. Various extracts of ethanol, hexane, chloroform, aqueous, and ethyl acetate was prepared from the seeds based on the polarity of the solvents using maceration method. Phytochemical screening of these extracts revealed the presence of alkaloids, saponins, proteins and amino acids, terpenoids, flavonoids, tannin, and steroids in the sample. Fourier Transform Infrared (FTIR) spectroscopy revealed the presence of various functional groups such as –OH, -COOH, -CH_2, and C=O from the IR absorption bands in the high wave region at 3456 cm^{-1} and 2939 cm^{-1} and the active compounds were identified by comparing the retrieved compounds with the standard chart. The number of protons present and the electronic state of the protons in the various compounds was analysed using H^1 NMR (Nuclear magnetic resonance). Further studies are to be done for the isolation of the active compounds that can be used to determine its therapeutic implications for the development of new drugs.

***Keywords*:** Acacia planifrons, Phytochemical, Alkaloids, Fourier Transform Infrared spectroscopy, functional groups, Nuclear magnetic resonance.

INTRODUCTION

Medicinal plants play a prominent role in inhibiting various pathogenic substances since the ancient times. The species of medicinal plants are of great significance in medicinal services. A major part of the total populace relies upon conventional plant-based medications. In the day to day life a numerous Medicinal herbs and spices are utilized as home grown cures and the various types of bioactive compounds that can act against predators like insects and fungi can be isolated from them. The umbrella thorn, (*Acacia planifrons*), is a species of Acacia of the family Fabaceae. Its common Tamil name is Kudai Vel and is a local to India and Sri Lanka. It is a prickly bush around 7m high [1]. Acacia is consumed as a drug by mouth for elevated cholesterol; bad tempered entrails disorder (IBS), and weight reduction. It is also used as a prebiotic to advance "great" microbes in the digestive system and to expel poisons from the body. Acacia is utilized in assembly utilized as a pharmaceutical fixing for throat or stomach aggravation and as a film-shaping operator in peel-off skin masks. As Acacia is a good dietary fiber, it makes the individuals to feel full so, that they may quit eating sooner than they generally would. This leads to weight loss and decrease in cholesterol levels. Acacia species are generally known as 'Babool' in India and have been medicinally used from a long time for the treatment of skin, sexual, stomach and tooth issues [2].

The phytochemical contents in therapeutic plants are the mixtures intensifying the demonstration specifically or in a indirect way avert or treat illness. The maceration method is a traditional technique used for extraction mainly used by the ayurvedas to treat several diseases based on various criteria. The samples are immersed in various solvents and incubated for 24 hrs. Phytochemicals are separated as essential and optional constituents relying on their capacities in plant digestion. FTIR and H^1 NMR are used for the identification of functional groups of the compounds present in the sample and to detect the number of protons present in the compound.

MATERIALS AND METHODS

Collection of plant material and preparation of various extracts:

Seeds of *Acacia planifrons* were collected from the Tamilnadu Forest Department, Chennai, Tamilnadu, India and were shade dried for 6 hours. The plant seed were ground to form fine powder and was filtered with a large pore size filter paper. Based on the polarity of various solvents (Hexane, Ethanol, Chloroform, Ethyl acetate) the sample was immersed in the ratio 1g of sample: 10 ml of solvent and was incubated for 8 hrs.

Phytochemical screening:

All the extracts were subjected to phytochemical screening using standard procedure to identify the phytoconstituents as described by Tiwari et., al [3]. Various test like Alkaloid, carbohydrate, cardiac glycosides, glycosides, saponins, protein, phenol, terpenoids, flavonoids, tannins, steroids were tested.

FTIR (Fourier Transform Infrared Spectroscopy) analysis of the sample:

FT-IR spectrometer (Varian Instruments, shimadzu) outfitted with a ceramic source, KBr bar splitter, and deuterated triglycinesulfate (DTGS) indicator with aggregate reflection (ATR) connection. The ATR inspecting gadget used a DuraSample IR single-pass jewel covered inner reflection adornment (Smiths Detection, Danbury, CT,USA) and a predictable contact weight was connected by method for a hardened steel bar and an electronic load show. The information retrieved was taken to ensure that the window (2 mm in measurement) of the ATR testing device was secured totally by fiber tests. Around five estimations for individual fiber tests was re-examined at various areas were gathered over the scope of 4000– 600 cm^{-1} at 4 cm^{-1} and 16 cm^{-1} co-included outputs. All spectra were given in absorbance units and no ATR standard adjustment was connected. Bringing in the spectra to the GRAMS IQ application in Grams/AI (Version 9.1, Thermo Fisher Scientific, Waltham, MA, USA), the mean

range was taken for each example and afterward was smoothed with a Savitzky– Golay work (polynomial = 2 and focuses = 11). The spectra were standardized by dividing the intensity of the individual band in the 1800–600 cm^{-1} district with the normal force in this1800– 600 cm^{-1} area, and resulting PCA portrayal was performed in the1800– 600 cm^{-1} IR locale, with mean focusing (MC) and Savitzky– Golay first-subordinate (2 degrees and 13 points) otherworldly pretreatment, and in addition with cross-approval technique. With the utilization of the Grams/AI program, incorporated powers of 4 groups at 2900, 1372, 895, and 664 cm^{-1} were assessed in the individual scopes of 3000 to 2800, 1410 to 1290, 910 to 875, and 684 to 650 cm^{-1} from standardized spectra. Independently, the unearthly set was stacked into Microsoft Excel 2007 to execute straightforward calculation examination [4][5].

Proton Nuclear Magnetic Resonance (H^1-NMR):

The zone under the plots gives information about the amount of protons present in the molecule, the circumstance of the signs (the mixture move) reveals information as for the engineered and electronic state of the protons, and the part configuration gives information about the amount of neighbouring (vicinal or geminal) protons. The abscissa shows the compound move (δ) estimations of the particular kind of protons and the ordinate exhibits the powers of the signs. For example, proton NMR data of a monoterpene, citral ($C_{10}H_{16}O$), detached from lemon grass oils. The indications of protons associated with inundated carbon particles, for instance, methyl, methylene, and methine clusters appear between δ 0.8 and 2.4 ppm. The signs some place in the scope of 4.8 and 5.9 ppm identify with the olefinicmethine. Moreover, the banner from 9.0 to 10.0 ppm is ordinary for aldehyde bundles [6][7][8].

RESULTS AND DISCUSSION:

The phytochemical screening shows the presence of alkaloids in ethanol, ethyl acetate, and hexane, saponins in ethanol, hexane, chloroform and aqueous extract, proteins and amino acids in ethanol, terpenoids, flavonoids, tannin, and steroids in the various extracts of sample is listed in Table 1. The FTIR spectroscopy was carried out to ascertain functional groups and the spectrum was recorded in diffused reflectance mode. The FT-IR spectrum of IR absorption bands in the high wave region at 3456 cm^{-1} and 2939 cm^{-1}; attributed to -OH, -CH and -CH_2 asymmetric and symmetric stretching vibrations respectively is shown in Figure 1. The observed band at 3273 cm^{-1} can be assigned to O-H, C=O stretching of –COOH functional group and the other bands in the spectral range are assigned to bending vibrations of CH, C-O-C and CH_3 groups as well as to skeletal bending bonds is listed in Table 2 and pictorial represented in Figure 1. The retrieved compounds were compared with standard chart for the identification of active compounds. The H^1 NMR experiments was carried out and the spectrum showed fine tertiary singlets at δ 0.63.0.65, 0.68, 0.74, 0.90, 0.97 and one more secondary hydroxyl group showed broad triplet at δ 2.02-3.87 and two olefinic protons at δ 5.34 to 7.26 representing the exocyclic double bond are listed in the Table 3.

Table 1: Phytochemical analysis of *Acacia Planifrons* seeds using various extract

Tests	Ethanol	Ethyl acetate	Chloroform	Hexane	Aqueous
Alkaloid	+	+	-	+	-
Carbohydrates	-	-	-	-	-
Glycosides	-	-	-	-	-
Saponins	+	-	+	+	+
Proteins & Amino acids	+	-	-	-	-
Phenol	-	-	-	-	-
Terpenoids	+	-	+	+	+
Flavonoids	-	-	-	-	+
Tanins	-	+	-	+	+
Steroids	+	-	-	-	-
Cardiac glycosides	-	-	-	-	-

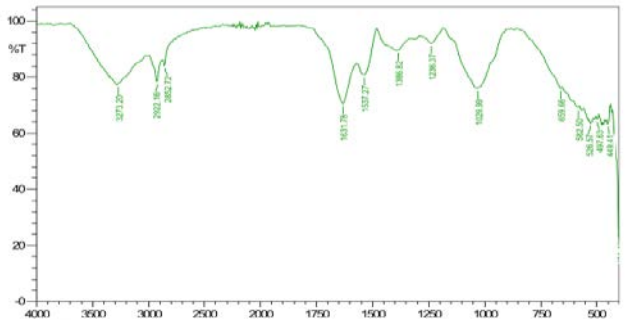

Figure 1: spectra of Fourier Transform Infrared Spectroscopy

Table 2: Compounds retrieved from FTIR spectrum

Spectrum	Compounds
3273.02	Alcohols (O-H stretch)
2922.16	Alkanes and alkyles (C-H stretch)
2852.72	Alkanes and alkyles (C-H strech)
1631.78	C=C-C(O)-OH or Ar-C(O)-OH. C=O stretch
1573.27	Amides (N-H)
1386.62	Alkanes and Alkyles (CH3 C-H bond)
1236.37	Alkyl halides (C-F stretch or ether)
1029.99	Ethers (Ar-O-R =C-O-C)
659.66	-
582.50	-
497.63	-
449.41	-

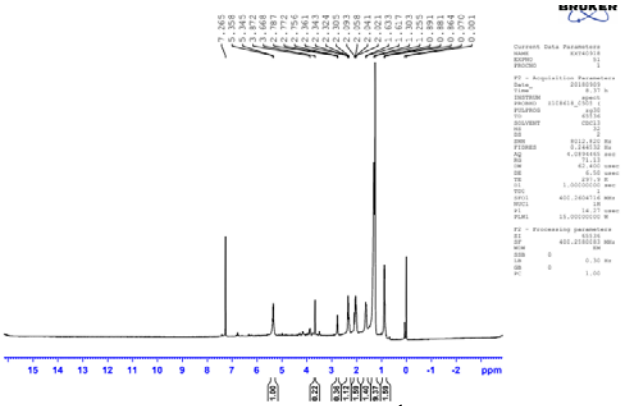

Figure 2: Spectra of H^1 NMR

Table 3 Compounds retrieved from H^1 NMR

PPM	Compounds
7.26	Aromatic phenol or OH
5.35	Vinylic or amide
5.34	Vinylic or amide
3.87	α to oxygen
3.66	Alcohol ,aniline
3.78	α to oxygen ,OH ,aniline
2.77	α to nitrogen
2.75	α to nitrogen
2.36	Benzylic
2.34	Benzylic
2.32	Benzylic
2.30	Benzylic
2.09	α to carbonyl
2.05	α to carbonyl
2.04	Alkyl
2.02	Alkyl
1.63	Alkyl
1.61	Alkyl
1.30	Alkyl
1.25	Alkyl
0.89	Alkyl
0.88	Alkyl
0.86	Alkyl
0.07	-
0.001	-

CONCLUSION

The dried powder of the seeds of *Acacia planifrons* was extracted with Ethanol, Ethyl acetate, Chloroform and Hexane respectively and was subjected to phytochemical screening that showed the presence of alkaloids, saponins, proteins and amino acids, terpenoids, flavonoids, tannin, and steroids in the extracts of sample. The chemical compounds were characterized on the basis of H^1 NMR and FTIR spectroscopy. The FTIR analysis of the seeds of *Acacia planifrons* reviled the presence of various functional groups such as carboxylic acids (-COOH), ketones (C=O), methylene ($-CH_2-$), alkenes (C=C) and alcohols (OH). The H^1 NMR was used for the retrieval of the number of protons present and the electronic state of the protons in the various compounds. Further research on various activities such as antibacterial, antifungal, anti-inflammatory, antipyretic has to be done for the determination of therapeutic uses of the isolated compounds.

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Conflicts Of Interest

There is no conflict of interest.

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