



Phytochemical screening, GC-MS and FTIR Analysis of Petroleum ether extract of *Pithecellobium dulce* (Roxb.) Benth.

A. M. Janani*, V. M. Annapandian, P. Natarajan, P. Mumthaj

Sankaralingam Bhuvaneshwari College of Pharmacy

* Corresponding author

Ms. A. M. Janani

Sankaralingam Bhuvaneshwari College of Pharmacy

3/77 – C, Anaikuttam Road,

Anaikuttam-626130,

Sivakasi, Tamil Nadu.

Email: jananimurugan1998@gmail.com

Mobile number: 9514223276.

Abstract:

The present study was aimed to estimate the phytochemical screening, GC-MS, FTIR analysis of the non-polar leaf extract of *Pithecellobium dulce*. The bioactive components was identified in petroleum ether extract based on their retention time and peak area. The FTIR analysis was done to identify the various functional groups present in the non-polar extract. Thus, the results obtained from the above analysis, the plant extract of the non-polar solvent contains various components have been identified in GC-MS and functional groups in FTIR analysis.

Keywords: GC-MS, FTIR, *Pithecellobium dulce*, functional group, petroleum ether

1. Introduction

The *Pithecellobium dulce* (Roxb.) Benth. belongs to the family of Leguminosae. It is an evergreen tree which is common and naturalized in the greater part of India and is also found in Southeast Asia. The height of *Pithecellobium dulce* is commonly found to be 10-15mtrs and it ranges from about 15-18mtrs. *Pithecellobium dulce* is one of 100-200 species within this genus and this species, is the one that became widespread outside its origin. The morphological features of the *Pithecellobium dulce* leaves are bipinnate, with 2 pairs of 2 kidney shaped leaflets. Thin spines are in pairs at the base of leaves and range from 2 to 15 mm in length¹. The tree is a host plant for the caterpillars of the red-bordered pixie, three-spot grass yellow and several moths. The plant originated from Brazil, Argentina, Bolivia, Colombia, etc., it is one of 18 species in this genus. It has been distributed naturally in many countries like India, Huawei, tropical Africa, and especially along the coast. The plant has been used traditionally for Antiseptic, Lightens Skin, Prevents Hair Loss, Treats Oily Scalp, Aids Weight Loss, Regulates Blood Circulation, Controls Blood Sugar Levels, Boosts Immune System, Relieves Inflammation, Cures Mouth Ulcers, Prevents Cancer, Cures Acne and Pimples, Removes Dark Spot, Natural Skin Moisturizer, Used to treat Venereal diseases².

2. Material and methods

2.1 Collection: The plant was collected nearby areas of Sivakasi district, Tamilnadu in the month of October.

2.2 Part used: Leaves

2.3 Authentication

The plant species was authentically verified by Dr. N. Senthilkumar, Head and Associate Professor of Botany, Ayya Nadar Janaki Ammal College, Sivakasi, Virudhunagar Dist and was certified as *Pithecellobium dulce* (Roxb.) Benth., of family Leguminosae (Fabaceae).

2.4 Extraction

2.4.1 Preparation of plant material

The collected *Pithecellobium dulce* (Roxb.) Benth. leaves was air dried and the coarsely powdered material was taken in Soxhlet apparatus and extracted with petroleum ether. The temperature was maintained at 40-50°C. After the extraction, the excess solvent was completely removed by distillation method and the obtained dried cured extract was stored in an individual glass bottles in desiccator.

2.5 Preliminary Phytochemical screening

The petroleum ether extract of *Pithecellobium dulce* leaves were subjected to phytochemical screening for the presence of various phytoconstituents^{3,4,5} according to standard methods.

2.6 Gas Chromatography-Mass Spectrometry analysis (GC-MS)

GC-MS is a combination of gas chromatography and mass spectrometer and it is one of the hyphenated analytical techniques containing a detection feature of mass spectrometry to estimate different substances within a test sample and GC which separates the volatile and thermally stable constituents present in the sample and finally GC-MS fragments the analyte to be determined on the basis of its mass.

2.6.1 Principle of GC MS

The GC works on the principle that when the sample is heated the individual substance present in a mixture will be separated. The sample is injected into the GC inlet where it is vaporized and enters into a chromatographic column by the helium which is used as a carrier gas. Then, the sample automatically moves through the column and the compounds present in the mixture are separated by their relative interaction with the coating of the column (stationary phase) and the carrier gas (mobile phase) which proceeds through a heated transfer line and terminates at the entry to ion source where compounds eluting from the column and a beam of electrons that ionize the molecules in the samples

leads to the formation of molecular ion and smaller ions with individual relative abundances that provide a 'fingerprint' for that molecular structure and then the ions are separated and detected by the mass analyser. The compounds were identified based on the comparison of their retention indices (RI), retention time (RT), mass spectra of WILEY, NIST library data of the GC-MS system and literature data⁶.

2.7 Fourier-transform infrared spectroscopy (FTIR)

FTIR spectroscopy is a reliable and sensitive method for detection of bio molecular composition and it is a most powerful tool for identifying the different types of chemical bonds that is functional groups present in compounds.

2.7.1 Principle of FTIR

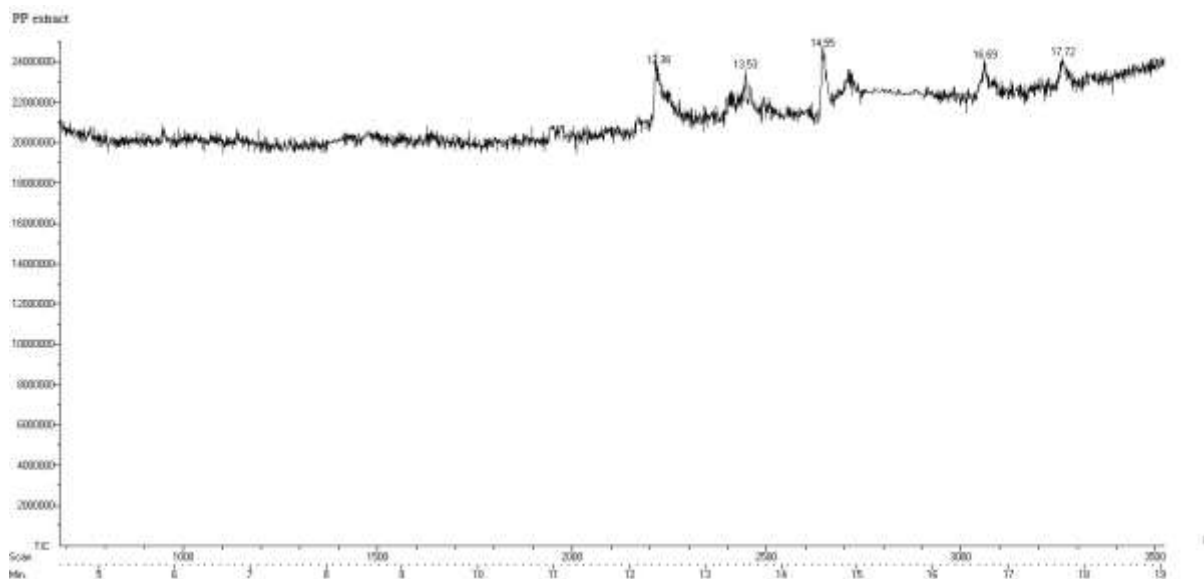
FTIR is based on the principle that when the infrared (IR) radiation passes through a sample, some of the radiation will be absorbed by the sample and some of the radiations that passes through the sample is recorded. Because different molecules contains different structures that produces different spectra which be used to determine and distinguish among molecules.

2.7.2 Procedure of FTIR

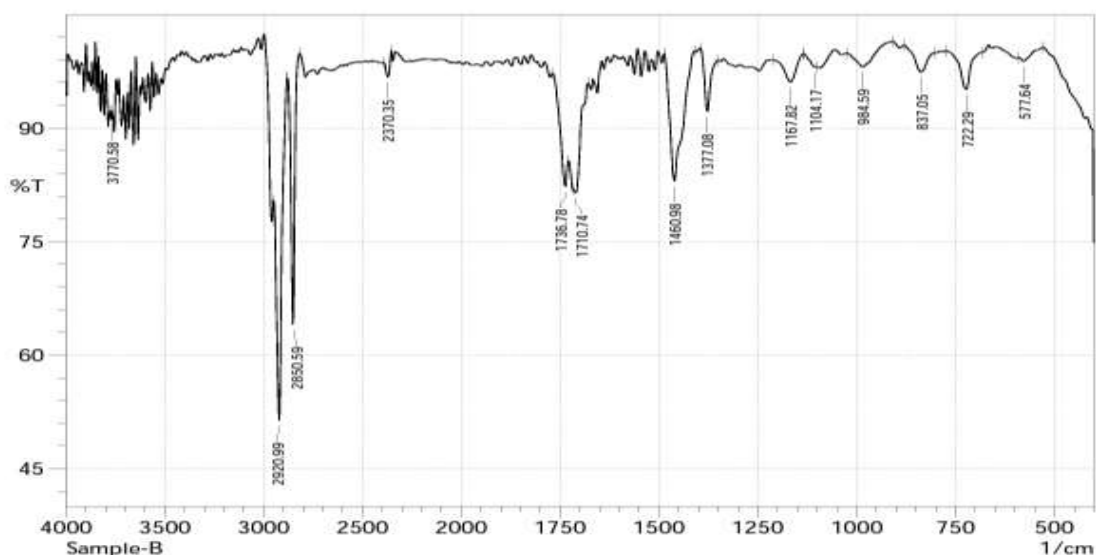
The FTIR analysis uses dried powders of the solvent extract of the plant material. A translucent sample disc was prepared using 10mg of the dried extract powder was encapsulated in 100 mg of KBr pellet which was then loaded in FTIR spectroscope, with a scan range from 400 to 4000 cm^{-1} and a resolution of 4 cm^{-1} is used⁷.

3. Results**Table 1: Preliminary Phytochemical analysis of *Pithecellobium dulce* (Roxb.)Benth leaf petroleum ether extract**

S.No	Plant constituent	Petroleum ether extract
1.	Steroids	+
2.	Flavonoids	-
3.	Alkaloids	+
4.	Carbohydrates	+
5.	Proteins	-
6.	Tannins	+
7.	Glycosides	+
8.	Saponins	-
9.	Triterpenes	+
10.	Fixed oil	+

Figure : 1 GC MS CHROMATOGRAPHY MASS SPECTROSCOPY ANALYSIS OF Petroleum**extract of *Pithecellobium dulce* (Roxb.) Benth****Table 2: Phytocomponents identified in petroleum ether extract of *Pithecellobium dulce* (Roxb.) Benth. by GC MS**

Retention time	Name of the compound	Molecular formula	Molecular weight	Peak area %
12.38	38 -Eta chloro 5 alpha cholestane 5,6, beta-diol 6 acetate	$C_{31}H_{52}O_4$	488.7	36.36
13.53	n-3-p-chlorophenyl-5-p-nitrophenyl thiene-2 yl 1-methylpiperidine 2-imine	$C_{20}H_{14}ClN_3O_2$	363.8	18.18
14.55	9,19-Cyclolanost-7-en-3-ol	$C_{30}H_{50}O$	426.7	27.23
16.69	1,16-Cyclocorynan-16 carboxylic acid, 17 (acetyloxy)-19,20 didehydro-10-methoxy-methyl ester	$C_{24}H_{28}N_2O_5$	424.5	9.05

Figure 2: FTIR of Petroleum ether extract of *Pithecellobium dulce* (Roxb.) Benth.**Table 3: FTIR peak values of Petroleum extract of *Pithecellobium dulce* (Roxb.) Benth.**

S.No	Peak value	Functional group
1.	2920.99	C-H Aromatic
2.	2850.59	C-H Aliphatic
3.	1736.78	C=O of esters and ketones
4.	1710.74	C=C Phenolic esters of aliphatic acids
5.	1460.98	CH ₂ methylene bend
6.	1377.08	C-O-C Aliphatic
7.	1167.82	C-O Stretching
8.	722.29	C-C Bending
9.	984.59	C-H Bending
10.	3770.58	OH
11.	577.64	CH ₃ Angular methyl
12.	837.05	C-C-H stretching

4. Discussion

The petroleum ether extract contains the presence of steroids, alkaloids, carbohydrates, tannins, glycosides, triterpenes and fixed oils.

There were four components identified from the GCMS 38 –Eta chloro 5 alpha cholestane 5,6, beta-diol 6 acetate (36.36%), n-3-p-chlorophenyl-5-p-nitrophenyl thiene-2 yl 1-methylpiperidine 2-imine(18.18%), 9,19-Cyclolanost-7-en-3-ol(27.23%),1,16-Cyclocorynan-16 carboxylic acid, 17 (acetyloxy)-19,20 didehydro-10-methoxy-methyl ester(9.05%).

The FTIR analysis revealed the presence of alkane, ketone, ester, methyl and hydroxyl groups.

5. Conclusion

The various bioactive compounds and functional groups have been identified through GC-MS and FTIR analysis in non-polar solvent extraction. This analytical study maybe useful for the further research works.

6. Reference

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