



Phytochemical Screening and FT-IR Spectral Studies of Aqueous and Chloroform Extract of *Desmodium Gangeticum* leaves

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ABSTRACT

*Chemical compounds produced by plants are known as Phytochemicals. Phytochemicals help to resist bacteria, fungi, viroous infections. They are also known as phytonutrients they may help prevent chronic diseases, including cancer. Phytochemicals present in fruits, vegetables, grains, leaves, flowers, roots and other plant foods. They will give lots of health benefits .Numerous phytonutrients are found in commonly consumed plant foods .In present study The leaves of *Desmodium gangeticum* were collected from the local field of Perambalur, India. It deals with the Qualitative Preliminary Phytochemical screening of *Desmodium gangeticum*(L) .Qualitative preliminary Phytochemical analysis was performed in aqueous and chloroform extract of *Desmodium gangeticum*(L).It revealed the presence of alkaloid, flavanoid, terpenoid,saponin, steroid, tannin and phenolic compounds, whereas steroids and volatile oil were absent.The Chloroform extract of *Desmodium gangeticum* contain more compounds when compared to other solvents. The Chloroform extract of *Desmodium gangeticum* showed of the presence of as saponin, alkaloids, steroids, terpenoids, coumarin, flavonoids, tannin, phenolic compound, and quinone were confirmed in suitable chemical tests. The aqueous extract of *Desmodium gangeticum* contain alkaloid, terpenoid, tannin, saponin and phenolic compound. Moreover, the highest yield was also observed in Chloroform extract and hence this was selected for further studies .The chloroform extract contains more phytochemical constituents when compared to other extracts. The functional group in *Desmodium gangeticum* also confirmed by FT-IR spectroscopic technique. FT-IR measurement was carried out to identify the possible biomolecules in *Desmodium gangeticum*. This spectrum shows lot of absorption bands indicates the presence of active functional groups in the *Desmodium gangeticum*. Some Phytoconstituent were separated in *Desmodium gangeticum* extract by thin layer chromatography technique. TLC of plant extract in chloroform reports three spots for various phytochemicals. The reported spots have enough space separation and having different R_f values showing the presence of number of different phytochemicals.*

Key words:*Desmodium gangeticum, TLC, FT-IR, Extract, alkaloids, plant leaf extracts*

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INTRODUCTION

Natural compounds extracted from plants, particularly higher plants, have been suggested as alternative sources for antibiotics[1].The chemical features of these constituents differ considerably among different species. Because they constitute a potential source of bioactive compounds that have been useful to maintenance of health in humans. Traditionally used medicinal plants produce a variety of compounds of known therapeutic properties .The substances that can either inhibit the growth of pathogens or kill them and have no or least toxicity to host cells are considered for developing new antimicrobial drugs. In recent years, antimicrobial properties of medicinal plants are being increasingly reported from different parts of the world .It is expected that plant extracts showing target sites other than those used by antibiotics will be active against drug-resistant microbial pathogens [2].

Plants secondary metabolites such as alkaloids, flavonoids ,anthocyanins, quinine, lignins, steroids and terpenoids have lots of commercial application as fragrance, drug, dye, flavour, insecticide etc., Such fine phyto chemicals are extracted ,isolated and purified from plant materials by using different solvents[3]. Most of the secondary metabolite structural speciality is generated by differentially modifying common backbone structures, with the derived compounds having potentially excellent biological activities[4] Differential changes of common backbone structures can differ the biological activity of a number of plant hormones and secondary metabolites like auxins, glucosinolates, gibberellins and phenylpropanoid derivatives[5].

The chemical characters of Natural compounds extracted from plants differ considerably among different plant species. This approach is attracting because they form a potential source of bioactive compounds that have been claimed by the general public as comparatively safe and often act at multiple and novel target sites thereby reducing the potential for resistance[6].

Medicinal plants of India Traditional medicines are used by about 60 % of the world's population. These are not only used for primary health care not just in rural areas in developing countries, but also in developed countries as well where modern medicines are predominantly used[7]. While the traditional medicines are derived from medicinal plants, minerals, and organic matter, the herbal drugs are prepared from medicinal plants only.

Use of plants as a source of medicine has been inherited and is an important component of the health care system in India. In the Indian systems of medicine, most practitioners formulate and dispense their own formulations; hence this requires proper documentation and authentication through research. In western world also, the use of herbal medicines is steadily growing with approximately 40 per cent of population reporting use of herb to treat medical illnesses within the past year[8]. Public, academic and government interest in traditional medicines is growing exponentially due to the increased incidence of the adverse drug reactions and economic burden of the modern system of medicine.

The biological impact of small molecular changes is significant enough that the pharmaceutical industry is creating combinatorial chemistry technologies to generate the same structural diversity *in vitro*. Plants synthesize a bewildering variety of phytochemicals but most are derivatives of a few biochemical motifs. Alkaloids contain a ring with nitrogen. Many alkaloids have dramatic effects on the central nervous system. Caffeine is an alkaloid that provides a mild lift but the alkaloids in plant cause severe intoxication and even death. Phenolics contain phenol rings[9]. Terpenoids are built up from terpene building blocks. Each terpene consists of two paired isoprenes. The names monoterpenes, sesquiterpenes, diterpenes and triterpenes are based on the number of isoprene units. The fragrance of rose and lavender is due to monoterpenes. The carotenoids produce the reds, yellows and oranges of pumpkin, corn and tomatoes. Glycosides consist of a glucose moiety attached to an aglycone. The aglycone is a molecule that is bioactive in its free form but inert until the glycoside bond is broken by water or enzymes[10].

The different plant materials, for example, *Morinda tinctoria* leaf extract, *Ananas comosus*, *Cymbopogon flexuosus* extract and *Camellia sinensis* are used to get ready different nano particles like Ag, Au nanoparticles[11]

MATERIALS & METHODS

Collection of plant material

The leaves of *Desmodium gangeticum* were collected from the local field of Perambalur, India. Leaves were shade dried, coarsely powdered with an electrical blender, dried leaves were ground into powder.



Fig:1 *Desmodium gangeticum* plant

Preparation of different fractions of *Desmodium gangeticum*

Chloroform, and aqueous extract were obtained by Solvent extraction method

Chloroform extraction

Coarse powder of the medicinal plant *Desmodium gangeticum* leaves was taken and weighed separately up to 100 grams and mixed with 300 ml of Chloroform. Beaker was closed with aluminum foil and left for 72 hours at room temperature. The extract was filtered through three layered muslin cloth and condensed into the powder by evaporation in water. The condensed powder was stored at 40 C and utilized throughout the studies[12].

Aqueous extraction

Coarse powder of the medicinal plant *Desmodium gangeticum* leaves was taken and weighed up to 100g and dissolved in 300ml of sterile distilled water. This substance was boiled for 30 minutes. Then the extract was filtered through three layered muslin cloth and condensed in to solid form at 40o C using hot air oven. The extract was weighed to find out the extraction value, and stored in a sterile container at 4° C for further use.

Qualitative Phytochemical Analysis

Phytochemical Analysis

Alkaloids

Two ml of extracts were measured in test tubes to which picric acid solution was added. An orange coloration indicated the presence of alkaloids.

Antraquinone

To 1 ml of the extract, 2 ml of 5% KOH was added separately. Then the solution was filtered. Change in colour was observed. Pink colour showed the presence of anthraquinones.

Coumarin

To 1 ml of all the extract, 1 ml of concentrated sulphuric acid was added and was allowed to stand for some time to develop colour. Development of red colour showed the presence of coumarin.

Flavonoids

To 5 ml of different extract, 1 ml of 10% NaOH solution was added. From the sides of the beaker 2 drops of concentrated HCl was added. Turning of Yellow colour solution to colourless is an indication for the presence of flavonoids.

Glycosides

25ml of diluted sulphuric acid was added to 5ml of extract in a test tube and boiled for 15 minutes, cooled and neutralized with 10% NaOH, then 5ml of Fehling solution was added. Glycosides are indicated by a brick red precipitate.

Phenol

To 1 ml of extract few drops of alcohol and ferric chloride solution was added and allowed for few minutes. Development of yellow colour showed the presence of phenol.

Saponins

Saponins were detected using the froth test. 1g of the sample was weighed into a conical flask in which 10ml of sterile distilled water was added and boiled for 5 minutes. The mixture was filtered and 2.5 ml of the filtrate was added to 10ml of sterile distilled water in a test tube. The test tube was stoppered and shaken vigorously for about 30 seconds. It was then allowed to stand for half an hour. Honeycomb froth indicated the presence of saponins[13].

Steroids

The extract powder was dissolved in two ml of chloroform in a dry test tube. 10 drops of acetic anhydride and two drops of concentrated sulphuric acid were added. The solution become red, then blue and finely bluish colour indicated the presence of steroids.

Tannins

To a portion of the extract diluted with water, 3-4 drops of 10% ferric chloride solution was added. A blue colour was observed for Gallic tannins and green colour indicated the presence of catecholic tannins.

Terpenoids

Four milligrams of extract was treated with 0.5 ml of acetic anhydride and 0.5 ml of chloroform. Then concentrated solution of sulphuric acid was added slowly and red violet colour was observed for terpenoid.

TLC

Thin layer chromatography (TLC) is a chromatographic technique used to separate mixtures. TLC was performed on aluminium foil, Which was coated with a thin layer of adsorbent After the sample has been applied on the plate, a solvent or solvent mixture was drawn up the plate via capillary action.⁴ Because different Components have different rate of adsorption separation is achieved.

FT-IR

In the FT-IR instrument, the sample is placed between the output of the interferometer and the detector. An interferogram of a reference (sample cell and solvent) is needed to obtain the spectrum of the sample. a computer performs a Fast Fourier Transform, which results in a frequency domain trace (*i.e* intensity vs. wavenumber). The detector used in an FT-IR instrument respond quickly because intensity changes are rapid [14,15]. Pyroelectric detectors or liquid nitrogen cooled photon detectors be used.

RESULT AND DISCUSSION

The preliminary phytochemical analysis was carried out for different extracts of *Desmodium gangeticum*. [16] .It revealed the presence of alkaloid, flavanoid, terpenoid, saponin, steroid, tannin and

phenolic compounds, whereas steroids and volatile oil were absent. The Chloroform extract of *Desmodium gangeticum* contains more compounds when compared to other solvents. The Chloroform extract of *Desmodium gangeticum* showed the presence of saponin, alkaloids, steroids, terpenoids, coumarin, flavonoids, tannin, phenolic compound, and quinone were confirmed in suitable chemical tests. The aqueous extract of *Desmodium gangeticum* contains alkaloid, terpenoid, tannin, saponin and phenolic compound [17]. (Table 1). Moreover, the highest yield was also observed in Chloroform extract and hence this was selected for further studies.

Table-1: Preliminary Phytochemical Screening Of *Desmodium Gangeticum*

S.NO	TEST	Water	Chloroform
1	Alkaloids	+	+
2	Terpenoids	-	+
3	Steroids	-	+
4	Coumarins	-	+
5	Tannins	+	+
6	Flavonoids	-	+
7	Phenols	+	+
8	Volatile oils	-	-
9	Quinone	-	+
10	Saponin	+	+

TLC analysis

TLC analysis also suggested the presence of different kinds of phytochemicals in leaves extract. Thin layer chromatography was performed on plant extracts using different solvent systems Methanol:Water: Acetone (18:9:1).

TLC of plant extract in chloroform reports three spots for various phytochemicals. The reported spots are separated with enough space and having various R_f values showing the presence of at least three phytochemicals in chloroform extracts. In our study, the most suitable TLC system for analysis was shown to be Methanol:Water: Acetone (18:9:1) with the largest discriminating power. Three bands found in this method and its R_f values were 0.4, 0.45 and 0.48. These values indicate the presence of phenolic compound [18].

Table 2 FT-IR Analysis of chloroform extract of *Desmodium gangeticum* leaves

Sno	Frequency	Types of Bonds	Group Present
1	3435.46	O-H stretch, H-bonded	alcohols, phenols
2	2832.16	C-H stretch	Alkanes
3	2719.36	C-H-O, C-H stretch	Aldehydes
4	2361.12	C-H-O, C-H stretch	Aldehydes
5	2092.40	C≡ bond N stretch	Carboxyl
6	1631.09	N-H bend	Primary amine
7	1133.33	C-H wag (-CH ₂ X)	alkyl halides
8	1362.54	C-H rock	Alkanes
9	997.84	C-H bond	Alkenes
10	924.84	o-H bend	Carboxylic acid

FT-IR ANALYSIS

FT-IR measurement was carried out to identify the possible biomolecules in *Desmodium gangeticum*. This spectrum shows lot of absorption bands indicates the presence of active functional groups in the *Desmodium gangeticum*. The intensity peaks are slightly increased for the period of 3435,2832,2719,2361 cm^{-1} as well as some intensity peaks decreased like 1362, 997, and 924 cm^{-1} . Fig: 1 shows the band at 3435 correspond to O-H Stretching vibrations of alcohol. The peak at 2719 represents to C-H in plane bend to alkenes. The peak at 997 corresponds to C-H, C-Br stretching vibrations to alkyl halides. The weak band at 1045 indicates C-O, C-N stretching vibrations and it corresponds to the presence of alcohols, carboxylic acids, ethers, esters and aliphatic amines in the seed extract (Table.2). FT-IR spectra showing the presence of IR peaks assigned to polyphenols and also the existence of IR bands characteristic of amide I and amide II groups specific for proteins/enzymes suggest that flavonoids and proteins present in aqueous petal extracts of ornamental plants could be responsible controlling pathogen[19].

CONCLUSION

The present study deal with the Qualitative preliminary Phytochemical screening of *Desmodium gangeticum*(L) .Qualitative preliminary Phytochemical analysis was performed in aqueous and chloroform extract of *Desmodium gangeticum*(L). The preliminary phytochemical analysis was carried out for different extracts of *Desmodium gangeticum* .It revealed the presence of alkaloid, flavanoid, terpenoid,saponin, steroid, tannin and phenolic compounds, whereas steroids and volatile oil were absent.The Chloroform extract of *Desmodium gangeticum* contain more compounds when compared to other solvents. The Chloroform extract of *Desmodium gangeticum* showed of the presence of as saponin, alkaloids, steroids, terpenoids, coumarin,flavonoids,tannin, phenolic compound, and quinone were confirmed in suitable chemical tests. The aqueous extract of *Desmodium gangeticum* contain alkaloid, terpenoid, tannin,saponin and phenolic compound.Moreover, the highest yield was also observed in Chloroform extract and hence this was selected for further studies. The chloroform extract contains more phytochemical constituents when compared to other extracts. The functional group in *Desmodium gangeticum* also confirmed by FTIR spectroscopic technique. Some Phytoconstituent were separated in *Desmodium gangeticum* extract by thin layer chromatography technique.

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CONFLICT OF INTEREST

The authors declare that they have no conflict of interest in this work

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