Pharmacognostical Characterization, Phytochemical Screening and Finger Print Profile of the Plant Desmodium gangeticum DC

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ABSTRACT
Herbal medicine, also called botanical medicine or phytomedicine, refers to the use of any plant seeds, berries, roots, leaves, barks or flowers for the medicinal purposes. From the ancient days the conventional medicines and herbalism is becoming more mainstream for the treatment of various chronic diseases. Herbalism is a traditional medicinal or folk medicine practices based on the uses of plants and plant extracts. Herbalism is also known as botanical medicine, medical herbalism, herbal medicine, herbology, and phytotherapy. Sometimes the scope of herbal medicine is extended to include fungi and bee products, as well as minerals, shells and certain animal parts. The Pharmacognostical studies of the plant Desmodium gangeticum DC, Family-Fabaceae were scientifically validated in this present study. The various physicochemical properties were evaluated like ash values, extractive values etc. The preliminary phytochemical analysis revealed that the dried roots of Desmodium gangieatum DC were extracted with the solvents of chloroform, ethanol and hydro- alcohol, showed that the presence of alkaloids, carbohydrates, glycosides, phytosterol and flavonoids. The chromatographic analysis was studied with the help of Thin layer chromatography.

Keywords Desmodium gangeticum DC, Fabaceae, TLC, Finger print, Extractive values

INTRODUCTION
Traditional and folklore medicines play an important role in health services around the globe. About three quarters of the world population relies on plants and plant products for health care. The plant Desmodium gangeticum DC, Family-Fabaceae has been used in folklore medicine in the treatment of various ailments. Many of the Ayurvedic formulations contain this medicinal plant and considered as a Master of Medicinal Plant in Ayurveda due to its wide uses in formulations. Being universal, role of plants has always stood a golden mark to exemplify the outstanding phenomenon of symbiosis. The existence of life in the universe is not endurable without the plants. The plants being in tremendous amount are associated with certain unique and a few common active principles known as secondary plant metabolites. These metabolites are referred to as organic compounds that are not directly involved in the normal growth, development or reproduction of organism. Unlike primary metabolites, absence of secondary metabolites does not cause immediate death, but is involved with the long-term impairment of the organism’s survivability or aesthetics and sometimes represents no significant change at all. According to the World Health Organization (WHO), the herbal medicines have been defined as the finished, labeled medicinal products that contain active ingredients, aerial or underground parts of the plant or other plant material or combinations. For the assessment of safety, efficacy and quality of herbal medicines, specific set of guidelines has been set by WHO. As per the estimation of WHO, around 80% of the world’s population presently use herbal medicine for primary health care (WHO technical report series 1996)1. Plant-derived substances have recently become of great interest owing to their versatile applications. Medicinal plants are the richest bio-resource of drugs of traditional systems of medicine, modern medicines, nutraceuticals, food supplements, folk medicines, pharmaceutical intermediates and chemical entities for synthetic drugs2. Desmodium gangeticum DC, a traditional medicinal plant, identifies a well-established role in some of the Ayurvedic preparations that justifies its substantial potential in the modulation of several physiological anomalies. DG hails from the Leguminosae family and is synonymous with Hedyserum gangeticum. The roots of DG constitute as one of the components of the Ayurvedic medicine, Dashmoola, also called Dasamularishtam, a dietary supplement, which has marked its use in treating diseases like rheumatism, jaundice, paralysis, puerperal fever, filaria, edema and post-natal care3,4. Popularly known as Prisniparni in Sanskrit, Desmodium gangeticum DC, an Indian medicinal plant has been widely used by many Ayurvedic and Unani physicians for curing fever, catarract, typhoid, piles, bronchitis, dysentery, asthma and various other

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with a number of branches, light brown lenticels, light yellow fracture fibers, the odor of the plant is not characteristic and taste of the plant is Sweetish and mucilaginous and its contain the Alkaloids, Vitamins, oils and minerals like calcium, phosphorus, magnesium and it is used in the treatment of Antipyretic, digestive, diuretic, aphrodisiac, it is also beneficial in the treatment of typhoid, piles and dysentery.

**MATERIALS AND METHOD**

**Pharmacognostic evaluation**

**Collection and authentication of plant**
The plant was collected from National Botanical Research Institute/Aminabad, Lucknow (U. P) and authenticated from National Botanical Research Institute, Lucknow by Dr. Alok Lahri Ref.No: NBRI/CIF/492/2015, Scientist and Head, Pharmacognosy and Ethnopharmacology Division.

**Preparation of Powder**
The fresh roots of Desmodium gangeticum DC were washed and cut in to small pieces. The same were dried in controlled hot air oven at the temperature of 30-40°C. The dried materials were coarse powdered by means of mechanical grinder and resulting powdered material were used for further studies.

**Physicochemical determination**

- **A. Ash values**
  - **Determination of Ash Values**
  - **Determination of Total Ash**
  - **Determination of Acid insoluble ash**
  - **Determination of Water soluble ash**

**PHARMACOLOGICAL PROPERTIES**

- **Antipyretic, digestive, diuretic**
- **Arsenic poisoning, scurvy, jaundice, fever**
- **Antiseptic**
- **Antimicrobial**
- **Aphrodisiac, aphthous ulcer, cough, toothache**
- **Antidiarrhoeal, anti-ulcer, antispasmodic, anti-diabetic, diuretic, anti-tubercular**

**PLANT PROFILE**

**Botanical source**

Shalparni consists of dried roots of Desmodium gangeticum DC (Fabaceae), Desmodium gangeticum DC is found in the outskirts of Himalayas, up to 5,000 ft and also available throughout India, usually growing in the dry hills areas. Mostly they are used in the medicine as an anti-asthmatic, bronchitis, aphrodisiac, antihelmintic, astringent to the bowels, cures typhoid fevers due to mental disorder, inflammations and piles. Outer Himalayas, up to 5,000 ft. and throughout India to Ceylon and Burma, malay peninsula—malay islands, china, Philippines and tropical Africa. The plant is widely distributed in tropics and sub-tropics. It is cultivated also in the plains.

**Table 1: The extractive values after successive extraction of roots of Desmodium gangeticum DC.**

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Name of solvent</th>
<th>Total yield (gm)</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Petroleum ether</td>
<td>1.60</td>
<td>0.533</td>
</tr>
<tr>
<td>2</td>
<td>Chloroform</td>
<td>9.36</td>
<td>3.12</td>
</tr>
<tr>
<td>3</td>
<td>Ethyl acetate</td>
<td>1.20</td>
<td>0.4</td>
</tr>
<tr>
<td>4</td>
<td>Ethanol</td>
<td>8.50</td>
<td>2.833</td>
</tr>
<tr>
<td>5</td>
<td>Aqueous</td>
<td>7.2</td>
<td>2.4</td>
</tr>
<tr>
<td>6</td>
<td>Hydro alcoholic</td>
<td>11.48</td>
<td>3.8</td>
</tr>
</tbody>
</table>

**Description**

Desmodium gangeticum DC have conserving of systemic tap root, poorly developed but lateral roots are 15-30 cm long, and 0.1-0.8 cm in thickness, uniformly cylindrical...
The percentage of alcohol-soluble extractive with reference to the air dried drug was then calculated.

**Determination of water-soluble extractive**

5 gm of air dried coarsely powdered drug was macerated with 100 ml of chloroform water in a closed flask for 24 hrs, shaking frequently during 6hrs and allowed to stand for 18 hrs. The extract was then filtered and evaporated 25 ml of filtrate to dryness in a tared flat bottomed shallow dish and dried at 105˚C to constant weight and weighed. The percentage of water-soluble extractive with reference to the air dried drug was then calculated. Results are present in the Table No.5 & 6.

**Phytochemical Analysis**

_**Extraction with different solvent**_

Shade dried roots of *Desmodium gangeticum* DC were extracted with different solvents like Petroleum Ether, Chloroform, Ethyl Acetate, Ethanol and Aqueous by using hot extraction method.

**Chemical Test**

- **Test for alkaloids**
  - Extracts or fractions were treated with Mayer’s reagent (Potassium-mercuric Iodide), appearance of cream coloured ppt. confirmed alkaloids.
  - Extracts or fractions were treated with Wagner’s reagent (Iodine in potassium iodide solution), when appearance of reddish brown ppt. confirmed alkaloids.
  - Extracts or fractions were treated with Dragendorff’s reagent (solution of Potassium Bismuth Iodide), when appearance of reddish brown ppt. confirmed alkaloids.

- **Test for Carbohydrates**
  - Molisch’s test: Extracts or fractions were treated with α-naphthol followed by addition of Conc. Sulphuric acid, when appearance of purple colour confirmed the presence of carbohydrates.
  - Benedict’s test

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Aerial parts of *Desmodium gangeticum* DC

Root of *Desmodium gangeticum* DC

Legume of *Desmodium gangeticum* DC

Figure 1a: Different images of Desmodium gangeticum DC
Figure 1: Showing the extractive yield of the dried roots extract of Desmodium gangeticum DC

Figure 2: Showing the Ash values of the dried root extracts of Desmodium gangeticum DC

Figure 3: Showing the extractive values of the dried root extracts of Desmodium gangeticum DC

Figure 4: Showing the Rf Values of the dried root extracts of Desmodium gangeticum DC
Table 2: Qualitative phytochemical results

<table>
<thead>
<tr>
<th>Qualitative screening</th>
<th>Phytocemical substance</th>
<th>Pet. Ether</th>
<th>Chloroform Acetate</th>
<th>Ethyl Acetate</th>
<th>Ethanol</th>
<th>Hydro-alcoholic</th>
<th>Aqueous</th>
</tr>
</thead>
<tbody>
<tr>
<td>ALKALOIDS</td>
<td>Mayer’s reagent</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Wagner’s reagent</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>Hager’s reagent</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Dragendorff reagent</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>CARBOHYDRATES AND GLYCOSIDES</td>
<td>Molish’s test</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Fehling’s test</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Benedict’s test</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
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<tr>
<td></td>
<td>Legal test</td>
<td>-</td>
<td>-</td>
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<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Borntrager test</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>kellarkillani test</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Balijet’s test</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Saponins</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Foam test</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

Extracts or fractions were treated with Benedict Reagent (173gm sod. Citrate + 100gm sod. Carbonate + 17.3gm copper sulphate) followed by boiling for 2 min then allowed to cool, when appearance of red, yellow or green ppt. depending on amount of sugar confirmed the presence of carbohydrates.

Tests for Flavonoids and Coumarins

Mineral acid test
Extracts or fractions were treated with Conc. Sulphuric acid, when appearance of yellow orange colour confirmed the presence of Flavonoids.

Sodium Hydroxide test
Extracts or fractions were treated with 10% sod. Hydroxide solution, when appearance of yellow orange colour confirmed the presence of Flavonoids.

Shinoda’s test (Mg-HCl reduction test)
Extracts or fractions in alcohol were added few fragment of magnesium ribbon followed by addition of conc. Hydrochloric acid drop by drop, appearance of pink to crimson red color after few minutes confirmed the presence of Flavonoids.

Test for steroids

Libermann-Burchard Sterol reaction
To the extracts or fractions glacial acetic acid was added followed by addition of conc. Sulphuric acid, when a colour change from rose, through red, violet and blue to green confirmed the presence of steroids.

Salkowski reaction
To the extracts or fractions chloroform was added followed by addition of conc. Sulphuric acid, when appearance of red colour in chloroform layer confirmed the presence of terpenoids.

Test for tannins

Ferric chloride test
To the extracts or fractions freshly prepared Ferric chloride solution was added, when appearance of blue-black or brownish colour confirmed the presence of tannins.

Results are tabulated in the Table No 2 & 3.

TLC of Alkaloids

Presence of alkaloids in various extracts and fractions was investigated by using the following mobile phases.

- Benzene: Methanol (95: 5)
- Methanol: Chloroform 40:60
- Ethyl alcohol
- Petroleum Ether
- Water: Alcohol
- Methanol: Chloroform 40:60
- Aqueous

The extractive values of alkaloids were determined from the extracts by quantitative chromatography (TLC).

RESULT AND DISCUSSION

The plant Desmodium gangeticum DC belongs to family Fabaceae. The roots of Desmodium gangeticum DC was universally used as a medicine. The roots were included in Ayurvedic pharmacopoeia of India, Vol-III. So we felt in worthwhile to validate it scientifically the folk claims for its therapeutic activity. The plant had been investigated in systemic way covering its pharmacognostical, phytochemical and TLC profile aspect.

Extractive value

The extractive values of Desmodium gangeticum DC showed that more fractions of material were available in hydroalcoholic (3.8%), chloroform (3.12%), ethanol (2.83%) and aqueous (2.4%). This showed that more fractions are available in these fractions compare to that petroleum ether (0.53%) and ethyl acetate (0.4%). These values may be due to presence of alkaloids present in the drug. values are tabulated below in (Table No. 1)

Pharmacognostical and phytochemical studies
Table 4: TLC of Chloroform, ethanol and hydroalcoholic extracts of Desmodium gangeticum DC.

<table>
<thead>
<tr>
<th>S. No.</th>
<th>TLC of extract</th>
<th>RF Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Chloroform extract/fraction</td>
<td>0.52</td>
</tr>
<tr>
<td>2.</td>
<td>Ethanol extract/fraction</td>
<td>0.76</td>
</tr>
<tr>
<td>3.</td>
<td>Hydro-alcoholic extract/fraction</td>
<td>0.61</td>
</tr>
</tbody>
</table>

Table 5: Ash values of the dried roots of Desmodium gangeticum DC

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Type of ash</th>
<th>% (W/W)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Total ash</td>
<td>11</td>
</tr>
<tr>
<td>2</td>
<td>Acid insoluble ash</td>
<td>2.54</td>
</tr>
<tr>
<td>3</td>
<td>Water soluble ash</td>
<td>1.065</td>
</tr>
</tbody>
</table>

Table 6: The alcohol soluble extractive values and the water soluble extractive values with different polarity of the solvent were carried out and result are as follows

<table>
<thead>
<tr>
<th>S. No</th>
<th>Type of extract</th>
<th>% (w/w)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Alcohol soluble</td>
<td>4.696</td>
</tr>
<tr>
<td>2</td>
<td>Water soluble</td>
<td>13.136</td>
</tr>
</tbody>
</table>

DISCUSSION

Desmodium gangeticum DC is one of the important ingredients of Dasmulakwatha of Ayurveda. The species Desmodium gangeticum DC is a plant that has been confused with other species due to their relative similarities, therefore many times some other materials are mixed or adulterated during the preparation of medicines. As standardization of a crude drug is an integral part of establishing its correct identity for its inclusion in herbal pharmacopoeia. For which pharmacognostic parameters and standards must be established. The total cash value is particularly important in the evaluation of purity of drugs, i.e. the presence or absence of foreign inorganic matter such as metallic salts and/or silica. As there is no pharmacognostical work on record of this much valued traditional drug, the present work was taken up with a view to lay down standards which could be useful to detect the authenticity of this medicinally useful plant.

CONCLUSION

In conclusion, the pharmacognostical features examined in the present study that includes physico-chemical and TLC may serve as a tool for identification/validation of the raw material and standardization of its formulations in fixing quality control parameters as well as answer to the latest

The taxonomical identification of the plant, its morphological characteristics further provided additional information about this plant drug. Analytical parameter like ash values and extractive values were carried out. The total ash, acid insoluble ash, water soluble ash values were carried out and the result are present in (Table No 5 & 6). Qualitative phytochemical analysis Both successive extracts and hydro-alcohol (Maceration) were subjected to chemical test for the detection of the phytoconstituents. The result revealed alkaloids, carbohydrates, glycosides, phytosterol and flavonoids are reported. (Table No 2 & 3). Thin layer chromatography

The Rf values of chloroform, ethanol and hydro-alcoholic extracts were found to be 0.52, 0.61 and 0.91 respectively. Values are present below:
GMP norms and FDA guidelines on standardization of herbal drugs. The present study revealed different physicochemical parameters which can be used for authentication of the crude samples. Physico-chemical as well as various aspects of the sample were studied and the sample of Desmodium gangeticum DC exhibits a set of physicochemical characters, which will help to identify the drug in dried condition. It has been concluded from this study that estimation of pharmacognostical evaluation is highly essential for raw drugs or plant parts used for the preparation of compound formulation drugs. The periodic assessment is essential for quality assurance and safer use of herbal drugs. As per phytochemical investigation, the main constituents present are alkaloids, carbohydrates, glycosides, phytosterol and flavonoids. The therapeutic activity of this plant may be due to the presence alkaloids. Further studies on the isolation of the compound will be carried out in future.

ACKNOWLEDGEMENT
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CONFLICT OF INTEREST
The authors declare that there is no conflict of interest

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