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Research Article

PHARMACOGNOSTICAL AND PHYTOCHEMICAL EVALUATIONS OF BARK OF *Pithecellobium dulce* – A MADRAS THORN PLANT

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A B S T R A C T

Pithecellobiumdulce is used Ayurvedic and Unani systems and other herbal medicine systems. The bark of *Pithecellobium dulce* has a good medicinal value. The present work is an attempt to study the pharmacognostical features, phytochemical and physico-chemical of a Madras thorn plant *P.dulce* were established by standard methods as prescribed in the Ayurveda Pharmacopoeia of India. The transverse section (TS) of bark revealed the presence of cork, phelloderm, phellogem, cortex, secondary phloem and medullary rays. Phytochemical studies showed the presence of carbohydrates, proteins, amino acid, steroids, flavonoids, alkaloids and tannins. Physico-chemical evaluation established, Ash values - Total, acid insoluble, water soluble and sulphated ash values were 6.88%, 0.88%, 3.63% and 10.21%, respectively. Extractive values - Alcohol and water soluble extractive values were 10.78% and9.36% respectively.The Methanol and Aqueous bark extracts (µg/ml) yielded 56.385 ± 0.100 and 35.170 ± 0.200, µg/ml gallic acid-equivalent phenolic content and 1.486 ± 0.051 and 6.287 ± 0.322, µg/ml quercetin-equivalent flavonoid content respectively. The result of this study is a detailed account of the distinct pharmacognostical features, phytochemical and physico-chemical of a Madras thorn plant *P.dulce*.

INTRODUCTION

*Pithecellobium dulce*plant iscomprehensively used by the mankind in the treatment of various ailments and complication. Phytochemicals are naturally occurring secondary metabolites derived from plants which may be biologically active. Locally, the plant*Pithecellobium dulce* Benthis known by various names such as Vilayti chinch, Mithi imli, Madras thorn, etc.It is belongs to the family *Leguminosae* or *Fabaceae(subfamily Mimosoideae)* family are assessed for the distribution of phytochemical constituents of medicinal importance like alkaloids, flavonoids, tannins, saponins, terpenoids, phenols and steroids like sitosterol, campesterol, stigmasterol and spinasterol in their aerial part like bark.(Kirtikar KR *et al*.,1975) (Chopra RN *et al*., 1992)(Rao GN *et al*., 2010)(Nigam SK *et al*.,1968).

A Madras thron is evergreen, spiny tree that reaches 22 meters heights.(Orwa C, Mutua A *et al* ., 2009). It has short trunk andtwining branches having milky white latex. Flower white or pale greenish white with fragrant odour. The bark of *P. dulce* issmooth and pale whitish-grey in color, they become rougher and eventually start peeling when gets matured. The bark is lenticellate, often with horizontal ribs encircling the trunk and branches. The bark and leaves show antidiabetic and anti-inflammatory activity.(Katekhaye S, Nagmoti D *et al*., 2012) (Katekhaye S, Kale M *et al*., 2012)

The bark of the plant which contains catechol type tannins is reported to be used as febrifuge, dermatitis and eye inflammation(Sukumaran M, Vetrichelvan T *et al*., 2009) (Pithayanukul P, Ruenraroengsak P *et al*., 2005)but Literature survey did not provide sufficient information about pharmacognostical and phytochemical studies of bark of plant. The present study was therefore exhibit out to deliver essential Pharmacognostical detail, physicochemical analysis, qualitative and quantitative phytochemical analysis about the bark of *Pithecellobium dulce*.

MATERIAL AND METHODS

Collection of Plants: Taxonomically identified experimental bark of *P. dulce* was collected from Dist: Jalgaon, T: Chopda, Maharashtra and authenticated at Maharashtra Association for the Cultivation of Science, Agharkar Research Institute, Pune – 411004 (Certificate number AUTH 19-129). The plant material was thoroughly cleaned, powdered, and sieved for further analysis.

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Macroscopic studies

Organoleptic characters

In Organoleptic evaluation, appropriate parameters like taste, odor, size, shape and color of the bark were studied. (Anonymous, 2005) Morphological characters

A morphological investigation of the plant bark was conducted. (Bentley R. Trimen H *et al* ., 2006)

Microscopic studies

Transverse Section (TS) of P. ducle bark

TS of bark was taken and stained with phloroglucinol to study about the general anatomical characteristic features such as structure of cork, cortex, phloem parenchyma, sieves tubes and lignified phloem fibers were separated by medullary rays and also to determine the characters of cell contents such as starch grains, stone cell and prismatic calcium oxalate crystals.(Khandelwal KR *et al*., 2008)

Physicochemical studies

In the physicochemical evaluation, ash values, viz. Total ash, acid insoluble ash, water soluble ash and sulphated ash and extractive values, viz. alcohol soluble extractive value, water soluble extractive value and loss on drying were determined as per standard procedures. The ash values represent the inorganic salts present in the drug. Extracts obtained by exhausting crude drugs are indicative of approximate measures of certain chemical compounds they contain, their diversity in chemical compounds, diversity in chemical natures and properties of the contents of drug. The percentage w/w values were calculated with reference to the air dried drug. (Khandelwal KR *et al*., 2008)

Phytochemical studies

Phytochemical analyses were performed by preliminary phytochemical screening to identify various secondary metabolites present in the plant bark using the standard procedures described by [12] and Quantitative Determination of Phytochemical Constituents of phenol and flavonoids.

Quantitative estimation of total Phenolic content

Total phenolic content of test extracts (Methanol and Water) was determined using Folin–Ciocalteu reagent.In this method, the blue colour formed due to the polyphenols present in the extract was measured at 660 nm using UV spectrophotometer.The extract (0.1mL) was mixed with the Folin-Ciocalteu phenol reagent (0.2 mL), water (2 mL) and sodium carbonate (15 % w/v, 1 mL), and absorbance was measured at 660 nm using spectrophotometer (Shimadzu 2405) after 2 h incubation period at 50 ºC for 10 min. All the experiment was performed in triplicate. The total phenolic content is expressed as µg gallic acid equivalents (GAE).(Gelareh M, Zahra E *et al*., 2009)

Quantitative estimation of total flavonoid content

Total flavonoid content oftest extracts (Methanol and Water) was determined using reported method .Sample solution (0.5 mL), ethanol (1.5 mL), Al(NO3)3(0.1 mL, 10%), CH3COONa (0.1 mL, 1 M) and water (2.8 mL) were thoroughly mixed and kept at ambient temperature for 40 min. The absorbance of reaction mixture was measured at 415 nm using spectrophotometer (Shimadzu 2405). All the experiment was performed in triplicate. Total flavonoid content was calculated according to a standard curve established with quercetin. The total flavonoid content was expressed as µg quercetin equivalents (QE). (Liu CT, Ching YW *et al*., 2005)

RESULTS AND DISCUSSION

Botanical description

Domain: Eukaryote
Kingdom: Plantae
Phylum: Spermatophyta
Subphylum: Angiospermae
Class: Dicotyledonae
Order: Fabales
Family: Fabaceae,Leguminosae
Genus: Pithecellobium
Species: *Pithecellobium dulce*

Vernacular Names

English: Sweet Tamarind
Telugu: Seema Chinta
Hindi: Jangle jalebi
Bengali: Dekhani babul
Tamil: Kodukkaapuli
Kannada: Kottampuli
Malayalam: Korukkapul

Macroscopic studies

Organoleptic characters

In Organoleptic evaluation, appropriate parameter such as colour, taste, odour, size, shape, and colour of the bark wasstudied. Bark is grey and smooth in young trees, turning to slightly rough and furrowed in old trees. Bark exudes reddish-brown gum when injured.They are brownish black in colour, cylindricalin shape with slight and feeble odour and pungent withslightly astringent taste.

Morphological character

The barks of *P. dulce* are light whitish-greyin color, they become slightly rougher, furrowed,longitudinally fissured , flaking off in irregular scales and eventually start peeling when gets matured. The inner bark of *P. dulce* is smoothish, thick, light brown and bitter or astringent.The bark is lenticellate, often with horizontal ribs encircling the short trunk and branches.

Figure 1 *Pithcellobium dulce* bark

Microscopic studies

Transverse section of bark

Cork made up of 3-4 layers of thin walled rectangular cells, some with brownish matter.

Phellogen containing two layers of colourless cellulosic cellsand Cortex made up of 10-15 layers of polygonal cellulosic parenchymatous cells. Few of these cells found to contain calcium oxalate crystals and a few starch grains. Starch grains are present in cortical parenchyma. Scattered group of 3-7 stone cells were present in cortex region. Groups of lignified stone cells arranged in tangential rows below the phelloderm. The parenchymatous cells contain prismatic calcium oxalate crystals and a few starch grains. Secondary phloemconsists of phloem parenchyma, sieves tubes and lignified phloem fibers were separated by medullary rays. Phloem parenchyma and medullary ray cells contain starch grains and prisms of calcium oxalate crystals.

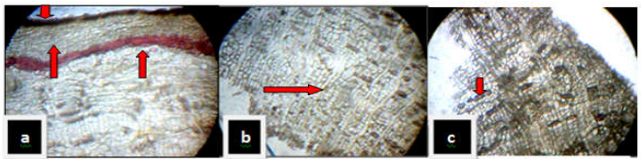

Figure 2 Transverse section of *Pithcellobium dulce*bark. (a) Cork cell, parenchymatous cells and sclerenchymatous cell (lignified stone cells), (b)Medullary ray cell, (c)lignified phloem fibers

Powder microscopy

The powder preparation of the bark was studied under the microscope and the following inclusions were observed.
Cork made up of 3-4 layers of thin walled rectangular cells.Phloem parenchyma and medullary ray cells contain starch grains and prisms of calcium oxalate crystals.Lignified phloem fiberswere separated by medullary rays.Stone cells were present in cortex region

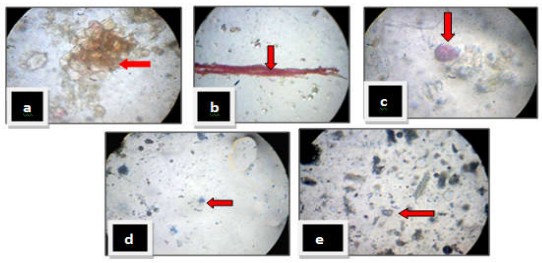

Figure 3 Powdered microscopy of *Pithcellobium dulce* bark. (a) Cork cell,(b) lignified phloem fibers, (c) stone cell, (d) starch grain, (e) Prismatic calcium oxalate crystal

Physicochemical studies

The physicochemical evaluation like ash values, viz. total ash, acid insoluble ash, water soluble ash and sulphated ash and extractive values, viz. alcohol soluble, water soluble and ether soluble extractive values and loss on drying were calculated and recorded (Table 1). Ash values - Total, acid insoluble, water soluble and sulphated ash values were 6.88%, 0.88%, 3.63% and 10.21%, respectively. Extractive values - Alcohol and water soluble extractive values were 10.78% and 9.36% respectively. The moisture content (Loss on drying) was 3.43%.

Phytochemical studies

The results of phytochemical screening of the Methanolic and water extracts of *P. dulce* bark revealed the presence of carbohydrate, glycoside, saponins, flavonoid, phytosterols, tannins and phenolic compounds shown in [Table 2]. These secondary metabolites are known to possess various pharmacological effects and may be responsible for variousactions of *P. Dulce.* Quantitative analysis [Table 3] showed that generally quantities of phenol and flavonoids were found in the Methanol and Aqueous bark extracts examined with the quantities obtained in extracts (µg/ml) yielded 56.385 ± 0.100 and 35.170 ± 0.200, µg/ml gallic acid-equivalent phenolic content and 1.486 ± 0.051 and 6.287 ± 0.322, µg/ml quercetin-equivalent flavonoid content respectively.

Table 1 Physicochemical studies of *Pithcellobium dulce* bark

Parameter	Values in % W/W
Total ash	6.88
Acid insoluble ash	0.88
Water soluble ash	3.63
Sulphated ash	10.21
Alcohol soluble extractive value	10.78
Water soluble extractive value	9.36
The moisture content	3.43

Table 2 Preliminary phytochemical screening of *Pithcellobium dulce* bark extracts

Test of extracts	PDBME	PDBWE
Carbohydrates	----	+
Proteins	+	+
Amino acid	----	----
Alkaloids	----	+
Flavonoids	+	+
Glycosides	----	----
Phenols	+	+
Steroids	+	+
Tannins	+	+

+: Presence,**---:** Absence, PDBME:*Pithcellobium ducle* bark methanol extract, PDBWE: *Pithcellobium dulce* bark water extract

Table 3 Quantitative phytochemical screening of *Pithcellobium dulce* bark extracts

Parameter	Values in % W/W	
	PDBME	PDBWE
Total Phenol content	56.3851	35.1703
Total Flavonoid content	1.4862	6.2874

PDBME: *Pithcellobium ducle* bark methanol extract, PDBWE: *Pithcellobium dulce* bark water extract

CONCLUSION

The present study concludes for the first time, the pharmacognostical and physicochemical parameters of the bark of *P. ducle.* This will provide useful information for identification of the plant materials. It also provides data for identification of biologically active phytoconstituents. These observations could be helpful in setting up of the diagnostic characters for the identification and preparation of a monograph and herbal section of Indian herbal Pharmacopeia.

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