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Research Article

PRELIMINARY PHARMACOGNOSTIC AND PHYSICO-CHEMICAL EVALUATION OF ROOT AND LEAVES OF *PATHA* (*CYCLEA PELTATA* (LAM.) HOOK.F. & THOMS)

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ABSTRACT

The quality control of herbal crude drugs and their bio constituents is of paramount importance in justifying their acceptability. The crude drugs can be identified systematically on the basis of their morphological, histological, chemical, physical and biological studies. Cyclea peltata (Lam) Hook. f. Thoms belonging to the family Menispermaceae is a common plant seen in South India. It is mentioned in Ayurvedic classics by the name Rajapatha and is one of the important drugs used in Avurveda therapeutics and is used widely in different formulations in medical practice. The aim of the study is to evaluate the preliminary pharmacognostical and phytochemical characters of Cyclea peltata (Lam) Hook. f. Thoms. Pharmacognostical evaluation of the plant Cyclea peltata (Lam) Hook. f. Thoms. was done by studying the macroscopic and microscopic features of leaf and root of the plant. The physical parameters studied included foreign matter, moisture content, volatile oil, total ash, acid insoluble ash, water soluble extractive, alcohol soluble extractive, fibre content and sugar content. The preliminary phytochemical analysis included qualitative chemical analysis, Thin Layer Chromatography, High Performance Thin Layer Chromatography and Atomic Absorption Spectroscopy. Various pharmacognostic and preliminary phytochemical characters observed in this may help in standardization, identification and carrying out further research in Cyclea peltata (Lam) Hook. f. Thoms.

KEYWORDS: Cyclea peltata (Lam) Hook. f. Thoms., Patha, Pharmacognosy, Phytochemistry.

INTRODUCTION

Cyclea peltata (Lam) Hook. f. Thoms is commonly known as velvet leaf. It is a slender twining shrub frequently climbing on tall trees. This species is found throughout South India. The useful part of this plant is root. Acharya Caraka mentioned this plant in Sandhaniyamahakashaya (healing promoters).^[1] The drug is found to be an ingredient in many of the Ayurvedic preparations. It is indicated for the treatment of skin diseases, vomiting, diarrohea. It is also claimed to possess wound healing and antimicrobial property. The researches have proven the anti-ulcer,^[2] anti bacterial^[3] and anti oxidant^[4] activities of the plant. The objective of the present study is to evaluate the pharmacognostic and phytochemical parameters of the plant which will help in the authentication of the plant.

MATERIALS AND METHODS

Collection of plant material

The fresh samples of root and leaves of *Patha* (*Cyclea peltata* (Lam) Hook. f. Thoms) were collected from their natural habitat near Sasthamcotta, Kollam district, Kerala. Samples were authenticated by the pharmacognosist and the voucher specimens were

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kept in Lab of Dept. of Dravyagunavijana, Government Ayurveda College, Trivandrum. They were shade dried and packed in zip lock polythene bag and labeled.

Pharmacognostic study

Macroscopic evaluation

Fresh root and leaf of the plant were taken for morphological and histological studies. The organoleptic characters of fresh plant of (*Cyclea peltata* (Lam) Hook. f. Thoms), viz. colour, feature, shape, taste, odour were analyzed.

Microscopic evaluation

Materials: Sharp blades, Safranin stain, glass slides, cover slips, glycerin, Petri dishes, brushes and digital microscope. For microscopical studies transverse sections of stem, root and leaf were prepared and stained as per standard procedure.^[5] The powders of drugs were studied microscopically and the characters were observed.

Physico-chemical evaluation

Physico-chemical analysis including the analyzing parameters such as the foreign matter,

moisture content, ash values, fiber content, volatile oil content, sugar content, different extractive values, qualitative analysis, heavy metal analysis by Atomic Absorption Spectroscopy and chromatographic techniques like TLC, HPTLC were done. The procedures were done as per mentioned in Ayurvedic Pharmacopoeia of India.

Reagents used

Xylene, dilute Hydrochloric acid, Petroleum ether, Cyclohexane, Acetone, Acetic anhydride, concentrated Hydrochloric acid, concentrated Sulphuric acid, Magnesium ribbon, neutral Ferric chloride, Benzene, Chloroform, Ethyl acetate, Potassium permanganate, Acetic acid, Fehling's solution, Sodium bicarbonate, Dragendroff's reagent, Ferric alum, Ethanol, Lead acetate, Sodium oxalate, Ethanol, and distilled water.

Apparatus

Dean and Stark's apparatus, Clevenger apparatus, Soxhlet apparatus, silica crucible, Bunsen burner, round bottomed flask, measuring jars, beakers, conical flask, funnel, glass rods, watch glass, electronic balance etc.

RESULTS AND DISCUSSION

Results of macroscopic evaluation

Macroscopic evaluation is the quickest and simplest method and is an important standardization parameter. Macroscopically, *Cyclea peltata* (Lam) Hook. f. Thoms. is a slender, hispid twining shrub (Figure 1). The organoleptic characters of root and leaf of fresh plant of *Cyclea peltata* (Lam) Hook. f. Thoms were summarized in Table No: 1 and Table No: 2 respectively.

Results of microscopic evaluation

Transverse section of leaf of *Cyclea peltata* (Lam) Hook. f. Thoms (figure 2)

Leaf has planoconvex midrib which is semicircular on the abaxial part and flat on the adaxial part. Epidermal cells of the midrib consist of circular, thick walled, wide cells with prominent cuticle. Inner to epidermis 2-3 layers of small collenchymatous cells. Palisade layer of lamina extends upto the adaxial median zone leaving a gap in the middle. Ground tissue of midrib is parenchymatous, midrib has a single large collateral vascular strand placed in the central part with thick walled xylem elements and horizontal band of phloem elements. Vascular bundle is covered by a sclerenchymatous sheath. The adaxial epidermis of lamina consist of wide and thick rectangular cells, abaxial epidermis is narrow and oblong and uniseriate trichomes are present in the abaxial surface. Mesophyll is differentiated into 2 layers of long cylindrical palisade cells and 6-7 layers of small lobed spongy parenchyma cells.

2. Transverse section of root of *Cyclea peltata* (Lam) Hook. f. Thoms (fig 3)

TS of the root is circular in outline. It shows a broad central core of stele filling the greater part of the area, a narrow ring of cortex, a thin brownish strip of skin. Outer skin consists of rows of rectangular cells, of which outer 6-8 rows compressed with dark brown walls. Phellogen or cork cambium is formed of one or two rows of cells. Phelloderm consists of 3-5 rows of tangentially elongated narrow rectangular or oblong cells. A narrow, annular, uninterrupted strip of 2 or 3 rows of elliptic oblong stone cells with thick pitted walls found inner to the cortex.

Stele: its tissues are medullary parenchyma composed of broad rectangular thin walled radially extended cells thickly packed with starch grains. Semicircular masses of phloems which almost abut on the ring of stone cells. Primary phloems are light yellowish crescentric arches of compressed tissue just outside the semicircular masses. Each radial strip of wood tissue is composed of a no of scattered xylem vessels of varying sizes.

The powder microscopy of leaf of *Patha (Cyclea peltata* (Lam.) Hook.f. & Thoms) (fig 4)

Reticulate venation can be found. Parts of trichomes, Epidermal peel and Vessel elements are also present.

The powder microscopy of root of *Patha (Cyclea peltata* (Lam.) Hook.f. & Thoms) (fig 5)

Simple or compound starch grains and Prismatic crystals of calcium oxalate were seen. Oil globules tannin containing materials and lignified thickened fibres were present. Group of small sized stone cells and fragments of pitted vessels were found.

Results of physico-chemical evaluation

Preliminary physical and physico-chemical analysis were done and the results are summarized in Table No: 3. The ash values of a drug give an idea of the inorganic composition and other impurities present along with the drug and the value was more in leaf (9.30%) than root (6.89%). Estimation of extractive values determines the amount of the active constituents in a given plant material when extracted with a particular solvent. The high water solubility of the contents than alcohol solubility of both root and leaf suggests aqueous extract for future studies.

Results of preliminary phytochemical analysis

The ethanolic extractive obtained was subjected to qualitative analysis for identification of various plant constituents like steroids, phenols, alkaloids, flavonoids, tannins and results were summarized in Table No:4. The presence of different plant constituents determines the pharmacological action and therapeutic potential of that plant. The preliminary phytochemical screening of root showed the presence of different phytoconstituent groups such as steroid, flavonoid, phenol, alkaloid and tannin. The leaves showed the presence of steroid, alkaloid and tannin.

Results of TLC and HPTLC

The spots obtained in TLC gives a rough idea about the study plant constituents. The best separation was achieved using Toluene: Acetone (9:1) for *Patha* leaf and Chloroform: Ethyl acetate: Formic acid (5:4:1) for *Patha* root. The plates were first viewed through UV-fluorescence viewing cabinet (365 nm) and the Rf values of the spots were noted (Table No:5). HPTLC was done and the HPTLC profile was shown in figure 6 and 7.

Results of Atomic Absorption Spectroscopy

Atomic absorption spectroscopy is used in the determination of heavy metal elements and some non-metal elements in atomic state and the results were shown in Table No:6. Four heavy metalscopper, cadmium, iron and lead contents were found within permissible limits. Hence the drug is not contaminated by heavy metals and can be used safely for internal administration.

Characters	Root
Shape	Cylindrical; cut surfaces are circular with radial structures traversing the porous tissue
Size	Diameter ranging from 1-2cm.
Surface characters	Outer skin is fairly smooth and very thin. In fresh roots it is so soft and could be easily scraped. Root has transverse constrictions and curves at frequent intervals. Prominent lenticels are found.
Colour	Surface is slate-grey or light brown tint
Fracture	Short of Ayurveda
Odour	No characteristic smell
Taste	Mild bitter

Table 1:	Macrosco	pic eval	luation	of root
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Table No 2: Macroscopic evaluation of leaves			
CHARACTERS	Leaves		
Insertion	Cauline		
Phyllotaxy	Alternate HAPR UPA		
Туре	Simple		
Shape	Ovate, Cordate at base, acute at apex		
Size	2.5cm to 10cm long, 2.5 to 3.75cm broad		
Margins	Entire		
Venation	Reticulate venation, 5 pairs of veins		
Texture	Coriaceous		
Colour	Upper surface is shiny and dark green, lower surface is paler		
Odour	Not distinct		
Taste	Slightly bitter		

Table 3: Preliminary physicochemical analysis

S.No	Name of experiment	leaf	root
1	Foreign matter	Nil	Nil
2	Moisture content	9.9	11.9
3	Volatile oil	Traces	Traces
4	Total ash (%)	9.30	6.89
5	Acid insoluble ash (%)	1.55	2.56
6	water soluble extractive (%)	5.77	16.76
7	Alcohol soluble extractive (%)	5.56	3.28

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8	Fibre content (%)		8.42	15.25
9	Sugar content	Reducing sugar (%)	0.70	0.81
		Total sugar (%)	1.21	1.34

Table 4: The results of Qualitative chemical analysis of alcoholic extract

Sl no	Chemical constituent	Leaves	Root
1	Steroid	++	++
2	Flavonoid	-	+
3	Phenol	-	+
4	Alkaloid	+	+++
5	Tannin	+	+
6	Saponin	-	-

Table 5: Rf values of spots obtained in Chromatography

Solvent system with ratio	Sample	No of spots	Rf value
	leaf	3	0.92
Toluene: Acetone (9:1)			0.75
			0.52
	root	4	0.15
Chloroform: Ethyl acetate: Formic acid -5:4:1			0.65
			0.77
			0.95

Table 6: Atomic absorption spectroscopy

Samples	Heavy metal concentration (in ppm)			
	Cu	Cd	Fe	Pb
Root	0.1812	0.0300	10.4995	0.0871
Leaves	0.2162	0.0340	11.4200	0.1084

Figures





Figure 2: transverse section of leaf of Patha

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Figure 3: transverse section of root of Patha



Figure 4: the powder microscopy of leaf of Patha



Figure 5: the powder microscopy of root of *Patha*



HPTLC plate with solvent system Chloroform : Ethyl acetate : Formic acid -5:4:1

Fig 6: HPTLC of Patha leaf

CONCLUSION

In the present study, pharmacognostic and phytochemical screening were carried out as per pharmacopoeia and WHO guidelines. The macroscopical and microscopical evaluations were done to ascertain the standard reference value for the standardization of plant material. This study revealed that the phytochemical constituents may be responsible for various pharmacological activities of this medicinal plant. These results may help in standardization, identification and in carrying out further research in Cyclea peltata (Lam) Hook. f. Thoms.

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HPTLC plate with solvent system Toluene: Acetone (9:1)

Fig 7: HPTLC of *Patha* root

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