International Journal of Phytomedicine 2 (2010) 277-283

http://www.arjournals.org/ijop.html

Research Article

ISSN: 0975-0185

Pharmacognostical and phytochemical studies on *Pothos scandens* L.

Lalitharani[1], V.R. Mohan[1]*, A. Maruthupandian[1]

***Corresponding author:**

V.R. Mohan

1 Ethnopharmacology unit,
Research Department of Botany
V. O. Chidambaram college,
Tuticorin 628008, Tamil Nadu,
India.
E.mail:
vrmohan_2005(at)yahoo.com

Abstract

Pothos scandens L. known to the *Kanikkars* as '*Paraioutan'* is an important medicinal plant. The *Kanikkar* tribe, inhabitants of Agasthiarmalai Biosphere Reserve, Western Ghats, Tamil Nadu, India. use leaf and stem of this plant to reduce body heat and also helps in conception. The present investigation deals with the pharmacognostic studies of the leaf and stem of the said plant. Pharmacognostic studies include microscopic, physicochemical constant (ash & extractive values), Fluorescence analysis and preliminary phytochemical evaluations.

Keywords: *Pothos scandens* L, *Kanikkars*, Pharmacognosy

Introduction

India has an ancient heritage of traditional medicine. The Material Medica of India provides a great deal of information on the folklore practices and traditional aspects of therapeutically important natural products. Indian traditional medicine is based on various systems including Ayurveda, Siddha, Unani and Homeopathy. The evaluation of these drugs is primarily based on phytochemical, Pharmacological and allied approaches. With the emerging worldwide interest in adopting and studying traditional systems and exploiting their potential based on different health care systems, the evaluation of the rich heritage of traditional medicine is essential [1]. There is a need for documentation of research work carried out on traditional medicine. With this backdrop, it becomes extremely important to make an effort towards standardization of the plant material to be used as medicine. The process of standardization can be achieved by step wise pharmacognistic studies [2]. These studies help in identification and authentication of the plant material. Correct identification and quality assurance of the starting materials is an essential prerequisite to ensure reproductive quality of herbal medicine which will contribute to its safety and efficacy. Simple Pharmacognostic techniques used in standardization of plant material include its morphological, anatomical and biochemical characteristics [3].

The *Pothos scandens* L. belongs to the family Araceae. It is commonly known as Paraioutan in *Kanikkar* tribals of Agasthiarmalai Biosphere Reserve, Western Ghats, Tamil Nadu. *Kanikkar* tribe, use leaf of this plant for reduce body heat and helps in conception [4]. However, perusal of literature reveals that Pharmacognostic information as *P. scandens* is totally lacking, hence in the present investigation was undertaken. The object of the present study is to evaluate various Pharmacognostic standards like microscopy of leaf and stem, ash values, extractive values, fluorescence analysis and preliminary phytochemical analysis of *Pothos scandens* leaf.

doi:10.5138/ijpm.2010.0975.0185.02039

Materials and Methods

Fresh leaf and stem materials were collected from the well grown trees found in the natural forest of Agasthiarmalai Biosphere Reserve, Western Ghats, Tamil Nadu. Identification and confirmation were done by Botanical Survey of India, Southern circle, Coimbatore, Tamil Nadu, India. Where voucher specimens were deposited in the Ethnopharmacology unit, Research Department of Botany, V.O.Chidambaram College, Tuticorin . For anatomical investigations standard microtomy techniques [5] were followed. T.S. of 10 to 12 µm thickness were prepared. These microtome sections were stained with 0.25% aqueous Toluidine blue (Meta chromatic stain) adjusted to pH 4.7 [6]. Photomicrographs were taken with Nikon trinocular photomicrographic unit. The most accepted descriptive terms were being used to describe the leaf and stem anatomy [7, 8].

Physicochemical and Fluorescence analyses

These analyses were carried out as per the standard procedures [9]. In the present study, the powdered leaf was treated with various chemical reagents like aqueous 1N Sodium hydroxide, alcoholic 1N sodium hydroxide, 1N hydrochloric acid, 50% sulphuric acid and concentrated nitric acid, picric acid, acetic acid, ferric chloride, conc.HNO_3 + NH_3 and their extracts were subjected to fluorescence analysis in day light and UV light (254 nm and 366 nm). Various ash types and extractive values were determined by following the standard methods [10].

Preliminary phytochemical analyses

Shaded dried and powdered leaf samples were successively extracted with Petroleum ether, benzene, chloroform and ethanol. The extracts were filtered and concentrated using vacuum distillation. The different extracts were subjected to qualitative tests for the identification of various phytochemical constituents as per the standard procedure [9,11].

Results

Microscopic features

Anatomy of Leaf (Plate-I, Fig.1): The lamina is isobilateral thin with smooth surfaces and noded at the regions of the vascular bundles of the lateral veins. The leaf-margin is conical and slightly bent down. The marginal part of the lamina is 40µm thick; the submarginal part is 70µm thick. The middle portion of the lamina becomes slightly thicker measuring 90µm thick.

The vascular system of the lamina has a horizontal row of vascular bundles which are circular and are smaller and larger alternating with each other in the lamina. The lamina becomes swollen into node in the region of the larger bundles. The smaller vascular bundles are 30µm in diameter. They have a central cluster of fine six small thick walled xylem elements surrounded by wider thick walled bundle sheath cells. The larger bundles are 70 -80µm in diameter. They are also circular with small central core of phloem, large cluster of xylem. The vascular strand is surrounded by sclerenchyma bundle sheath .

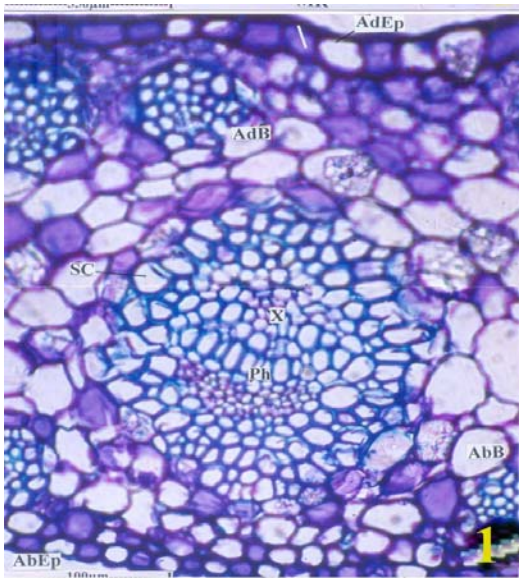

Fig.1- T.S. of leaf with large vascular bundles.

Towards the midrib, the leaf becomes planoconvex with flat adaxial side and convex abaxial side. The midrib is about 350µm thick

and 550µm wide. The midrib portion has an adaxial row of circular smaller bundles and abaxial row of similar type of circular smaller bundles.

The adaxial and abaxial bundles have small nest of phloem elements surrounded by a wide zone of thick walled angular xylem elements and wide bundle sheath of sclerechyma cells.

In the median portion of the midrib lies a wide circular midrib bundle. This bundle has a central core of arc-shaped phloem elements and adaxial was of angular xylem elements. The bundles have two or three layers of sclerenchymatous bundle sheath.

The leaf has an adaxial layers of rectangular thick walled cells. The cells are 20µm thick. The abaxial epidermis is comparatively narrow; the cells are circular with thick outer tangential walls. In between the epidermal layers occur four or five layers of circular or elliptical, more or less compact mesophyll tissue.

Anatomy of Stem (Plate-I, Fig.2) :

The stem is more or less circular in sectional view with flat lateral sides. It is 1.5mm in vertical plane and nearly 2mm in horizontal plane. The stem is clearly differentiated into outer cortex and cortical vascular bundles and central circular stele and steler vascular bundles.

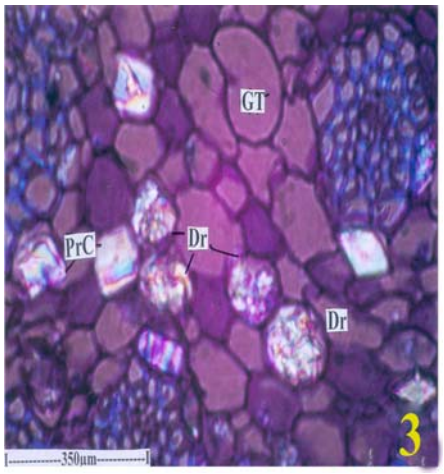

Fig-2- T.S. of Stem

There is a distinct, continuous epidermal layer of thick walled, lignified circular cells. The cortex is 350 µm wide. It has circular or angular parenchymatous ground tissue. The cortical vascular bundles are in two whorls; the outer wholes of bundles are smaller measuring 70 – 100 µm in diameter. The bundle has a central, single wide, circular xylem elements and less conspicuous small cluster of phloem elements. It has wide, thick sclerenchymatous bundle sheath. The inner cortical bundles are larger, measuring 150µm in diameter. There is a wide circular xylem elements in the centre, which is 70µm in diameter. A cluster of phloem elements occurs on the outer end of the xylem. The entire vascular strand is surrounded by a wide, highly thick walled sclerotic sheath.

The stellar portion has fairly distinct endodermoid layer of less thick walled cells. Inner to the endodermoid layer, the ground tissue is sclerenchymatous which forms a thick outer cylinder. The stellar vascular bundles are scattered and densely occupy the stele. The stellar bundles have a wide, circular, thin walled metaxylem element and two to three metaphloem elements and a small cluster of protophloem elements. The metaphloem elements are 150µm wide.

Cell inclusions:

Calcium oxalate crystals are abundant in the mesophyll tissue of the midrib, cortical parenchyma and stellar ground tissue. The crystals are predominantly prismatic type; occasionally druses may also occur in the midrib tissue (Plate- I, Fig.3). In the stem, the crystals are located all along the outer periphery of endodermoid layer (Plate-I, Fig.4) and in the pith parenchyma.

Powder analysis of the drug:

The results of the ash and extractive values of *Pothos scandens* leaf drug powder are depicted in Table - 1. The total ash content of the powdered leaf is 11.34% and extractive value in water is more than in other solvents. The results of fluorescent analysis of leaf powder of *Pothos scandens* are shown in Table - 2. The leaf powder shows the characteristic flurosecent green colour treated with 1N aqueous NaOH, and picric acid under short UV light. The result of preliminary phytochemical screening of leaf extracts of

Pothos scandens are presented in Table -3. The ethanol extracts of the leaf shows the presence of alkaloid, catachin, coumarin, tannin, saponin, flavonoid, phenol, sugar, glycoside and xanthoprotein.

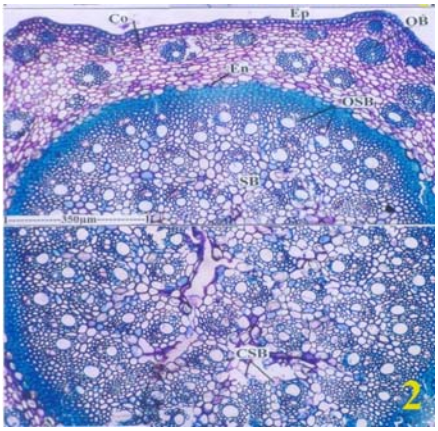

Fig.3- Prismatic crystals druses in the midrib cells

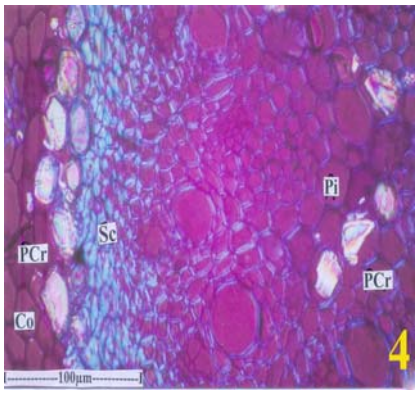

Fig.4- Prismatic crystals in the cortex near the endodermoid layers and pith cells

Table. 1. Ash and extractive values of the powdered leaf of *Pothos scandens*

Ash Values		
S.No	**Type of Ash**	**% of Ash**
1	Total ash value of powder	11.34 ± 0.03
2	Water soluble ash	5.10 ± 0.01
3	Acid insoluble ash	0.25 ± 0.03
4	Sulphated ash	13.98 ± 0.12
Extractive Values		
S.No	**Nature of extract**	**Extractive value (%)**
1	Petroleum ether	3.30 ± 0.06
2	Benzene	2.51 ± 0.01
3	Chloroform	3.31 ± 0.03
4	Acetone	8.20 ± 0.11
5	Methanol	9.89 ± 0.07
6	Ethanol	7.72 ± 0.03
7	Water	11.34 ± 0.21

Table.2: Fluorescence analysis of the powdered Leaf of *Pothos scandens*

Experiments	**Visible / Day light**	**UV Light**	
		254nm	**365nm**
Drug powder as such	Green	brown	Brown
Powder + 1N NaOH (aqueous)	Yellow	Fluorescent green	Green
Powder + 1N NaOH (alcohol)	Green	Green	Yellow
Powder + 1N HCL	Light brown	Light green	Brown
Powder + 50% H_2So_4	Pale yellow	Green	Greenish yellow
Drug powder + Nitric acid	Brown	Light green	Brown
Drug Powder + Picric acid	Fluorescent yellow	Fluorescent green	Green
Drug Powder + Acetic acid	Light green	Green	Orange
Drug Powder + Ferric chloride	Yellowish brown	Green	Brown
Drug Powder + HNO_3 + NH_3	Pale yellow	Pale yellow	Light green

Table. 3. Preliminary phytochemical screening of leaf extracts of *Pothos scandens*

Test	**Petroleum ether**	**Benzene**	**Chloroform**	**Ethanol**
Alkaloid	-	-	+	+
Anthraquinone	-	-	-	-
Catachin	-	+	-	+
Coumarin	+	-	+	+
Flavonoid	-	+	-	+
Phenol	+	+	+	+
Quinone	-	-	-	-
Saponin	-	-	+	+
Steroid	-	+	-	-
Tannin	+	-	+	+
Terpenoid	-	+	+	-
Sugar	-	-	+	+
Glycoside	-	-	-	+
Xanthoprotein	-	+	+	+

Fixed oil	+	-	+	-

Discussion

The present study attempts a modest comprehensive investigation of the leaf and stem of *Pothos scandens*. Since the whole plant of *Pothos scandens* as the folklore claims has therapeutic qualities the present investigation has laid down a set of anatomical features of the leaf and stem which can be employed for its botanical diagnosis. The salient features of identification of the fragmentary sample are;

- The leaf with thick planoconvex midrib and uniformly thin, noded lamina.
- The midrib with one large median vascular bundle and 6 small vascular bundles along the abaxial and adaxial, subepidermal regions.
- The lamina with single horizontal, median row of larger and smaller bundles.
- All vascular bundles are collateral with abaxial arc of phloem and adaxial mass of xylem elements; vascular bundles ensheathed by a thick layer of thick walled liginified sclerenchyma cells.
- Calcium oxalate crystals of prismatic as well as druses types abundant in the ground parenchyma cells.
- The stem is circular with flat lateral sides, comprising of wide cortex and cortical vascular bundles and thick, sclerenchymatous endodermoid layer eucircular with stele with many circular, discrete vascular strands forming atactostele.
- Outer vascular bundles of the cortex are smaller and inner cortical bundles are larger.
- Cortical bundles have single, wide, circular metaxylem element and a small cluster of phloem elements; the bundles have thick sclerenchymaous bundle sheath.
- Steler bundles have one wide circular metaxylem and two or three protoxylem elements and small cluster of phloem on the outer portion of the protoxylem. The bundles have thick sclerenchyma sheath.
- Calcium oxalate prismatic crystals are abundant along the endodermoid layer as well as ground parenchyma cells.

The physical constant evaluation of the drug is an important parameter in detecting adulteration or improper handling of drugs [12]. Equally important in the evaluation of crude drugs, is the ash value and acid insoluble ash value determination. The total ash is particularly important in the evaluation of purity of drugs, i.e., the presence or absence of foreign organic matter such as metallic salts and/or silica [13].The ash value of *Pothos scandens* leaf is 11.34%. This ash value is indicative of the impurities present in the drug. Since the ash value is constant for a given drug, this value is also one of the diagnostic parameters of the drug. In the present study, *Pothos scandens* leaf has more water soluble ash than acid insoluble ash. The ash value is generally the index of the purity as well as identity of the drug.

Many phytocompounds fluoresce when suitably illuminated. The fluorescence colour is specific for each compound. A non fluorescent compound may fluoresce if mixed with impurities that are fluorescent. The fluorescent method is adequately sensitive and enables the precise and accurate determination of the analyze over a satisfactory concentration range without several time consuming dilution steps prior to analysis of pharmaceutical samples [14].

In the present study, the powdered leaf of *Pothos scandens* emitted green under day light, brown under short UV and brown in long UV light. Presence or absence of certain important compounds in an extract is determined by colour reactions of the compounds with specific chemicals which act as dyes. This procedure is a simple preliminary pre-requisite before going for detailed photochemical investigation. Various tests have been conducted qualitatively to find out the presence or absence of bioactive compounds. Different chemical compounds such as alkaloid, catachin, coumarin, tannin, saponin, flavonoid, phenol, sugar, glycoside and

xanthoprotein are detected in *Pothos scandens* leaf extracts, which could made the plant useful for treating different ailments as having a potential of providing useful drugs of human use.

Saponins, a group of natural products occur in the leaf extract of *Pothos scandens*. In plants, the presence of steroidal saponins like, cardiac glycosides appear to be confined to many families and these saponins have great pharmaceutical importance because of their relationship to compounds such as the sex hormones, cortisones, diuretic steroids, vitamin D etc.,[15]. Saponin reduce the uptake certain nutrients including glucose and cholesterol at the got through intra-luminal physicochemical interactions. Hence, it has been reported to have hypocholesterolemic effect and thus may aid lessening metabolic burden that would have been placed in the liver [16].

Several authors reported that flavonoids, sterols/terpenoids, phenolic acids are known to be bioactive antidiabetic principles [17,18]. Flavonoids are known to regenerate the damaged beta cells in the alloxan induced diabetic rats [19]. Flavonoids act as insulin secretagogus [20]. Most of the plants have been found to contain substances like glycosides, alkaloids, terpenoids, flavonoids etc, which are frequently implicated as having antidiabetic effects [21].

Since the plant, *Pothos scandens* is useful in traditional medicine for the treatment of various ailments, it is important to standardize it use for as a drug. The Pharmacognostic study of the *pothos scandens* has been carried out for the first time. The pharmacognostic constant for the various parts of above said plant, the diagnostic microscopic features and the numerical standards reported in this work could be useful for the compilation of a suitable monograph for it proper identification.

References

1. Prakash PM. *Ficus racemosa* Linn – An overview. Natural Product Radiance. 2009; 8:84-90.
2. Ozarkar KR. Studies on anti-inflammatory effects of two herbs *Cissus quadrangularis* Linn. and *Valerina wallichi* DC using mouse model. Ph.D. Thesis, University of Mumbai, Mumbai.2005.
3. Anonymous. Macroscopic and microscopic examination: Quality Control Methods for Medicinal Plant Materials, WHO, Geneva.1998.
4. Lalitharani S, Mohan VR, Regini GS. Ethnomedicinal plants used by Kanikkars in Karaiyar of Agasthiamalai biosphere, Tamil Nadu. J. Econ. Taxon. Bot. 2010; 34: 472-477.
5. Johansen DA. Plant Microtechnique. McGraw - Hill Book Company, Inc.New York and London. 1940. pp. 1 – 523.
6. O' Brien TP Feder N dMc Cull ME. Polychromatic staining of plant cell walls by toluidine blue – O. Protoplasma. 1964; 59: 364 – 373.
7. Junikka L. Survey of English Macroscopic bark terminology. IAWA Bull. 1994; 15: 3-45.
8. Trockenbrodt, M. Survey and discussion of the terminology used in bark anatomy. *IAWA Bull*. 1990; 11: 141 - 166.
9. Lala, P.K. Lab Manuals of Pharmacognosy, CSI Publishers and Distributors, Calcutta, 5th (Ed) 1993.
10. Anonymous. Indian Pharmacopoeia, Government of India, Ministry of Health and Family Welfare, The Controller of Publications, Civil Lines, Delhi – 110054. 1996. Vol. I&II.
11. Brindha, P., Sasikala, P. and Purushothaman, K.K. Pharmacognostic studies on Merugan kizhangu. Bull. Med. Eth. Bot. Res. 1981; 3: 84-96.
12. African Pharmacopoeia. General Methods for Analysis Ist ed. 2: (OAU/STRC) Lagos. 1986; p. 123.
13. Musa KY, Katsayal AU, Ahmed A, Mohammed Z, Danmalam UH. Pharmacognostic investigation of the leaves of *Gisekia pharmacioides*. African J. Biotech. 2006; 5: 956-957.

14. Pimenta AM, Montenegro MC, Ara Ujo AN, Mart'inez JC. Application of sequential injections analysis to pharmaceutical analysis. J. Pharm. Biomed. Anul. 2006; 40: 16-34.
15. Evans WC, Saunders WB (Eds.). Trease and Evan's Pharmacognosy Tokyo. 2001, pp. 1- 579.
16. Price KR, Johnson LI, Ferewick H. The chemical and biological significant of saponins in food and feeding staffs CRC Critical Rev. Food. Sci. Nut. 1987; 26: 127-135.
17. Oliver-Bever B. Medicinal plants in tropical West Africa, Cambridge University Press, London. 1986, pp: 245-267.
18. Rhemann AV, Zaman K. Medicinal plants with hypoglycemic activity. J. Ethnopharmacol. 1989; 26: 1-55.
19. Chakaravarthy BK, Gupta S, Gambir SS, Gode KD. Pancreatic beta cell regeneration. A novel antidiabetic mechanism of *Pterocarpus marsupium* Roxb. Indian J. Pharmacol. 1980; 12: 123-127.
20. Geetha BS, Mathew BC, Augusti KT. Hypoglycemic effects of leucodelphinindin derivative isolated from *Ficus bengalensis* Linn. Indian J. Physiol. Pharmacol. 1994; 38**:** 220-222.
21. Loew D, Kaszkin M. Approaching the problem of bioequivalence of herbal medicinal products. Phytpther. Res. 2002; 16**:** 705-711.