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PHARMACOGNOSTICAL INVESTIGATION OF *INDIGOFERA ASPALATHOIDES* VAHL. EXDC.

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ABSTRACT

Indigofera aspalathoides Vahl. Exdc. (Fabaceae) commonly known as "Shivanar vembu" in tamil. It is a erect herb. The whole plant has used to various treatment like leprosy, skin disease, antiseptic, anticancer, toothache, antifungal, anti-inflammatory, hepatoprotective and antitumor. The present investigation focuses on the Pharmacognostical studies include microscopic, physic-chemical constant, fluorescent analysis and preliminary phytochemical evaluations. The total ash content of the whole plant powder is 94% and the extractive value of benzene is more than other solvents. The whole plant powder shows the characteristics fluorescence colour when treated with 50% H_2SO_4, Alcoholic NaOH and 1N NaOH under UV light. The ethanol extracts shows the presence of catechin, flavanoid, phenol, quinine, saponin, steroid, terpenoid, sugar and glycoside. These findings may be suitable for inclusion in in the proposal pharmacopoeia of Indian medicinal plants.

KEYWORDS: *Indigofera aspalathoides*, Pharmacognosy, physicochemical, phytochemical analysis.

INTRODUCTION

People are becoming more aware of medicinal plant resources and many of them utilize these therapeutic interventions and their products in maintaining health and preventing diseases with an ecofriendly touch. Herbal medicines are promising choice over modern synthetic drugs. They show minimum or no side effects and are considered to be safe. Generally herbal formulations involve use of fresh or dried plant parts. They also provide raw materials for pharmaceutical industries and represent a substantial proportion in global drug market. Correct knowledge of such crude drugs is very important aspect in preparation, safety and efficacy of the herbal product. Pharmacognosy is a simple and reliable tool, by which complete information of the crude drug can be obtained.[1-6] *Indigofera aspalathoides* Vahl. Exdc. (Fabaceae) commonly known as "Shivanar vembu" in tamil. It is distributed in the red soil of Western Ghats of Tamil Nadu India. It is an erect tree to 10 meter tall with thick stem and underground root stock. The aerial part of the bark is greyish brown and the leaves are used by the rural folk for curing various ailments like skin diseases, fever, dysentery, cooling, astringent, abdominal pain and hemorrhages. Perusal of the previous literature revealed that this medicinal plant is unexplored, so a detailed systematic pharmacognostic study was carried out.

MATERIALS AND METHODS

PLANT COLLECTION AND AUTHENTICATION

The aerial parts of the tree were collected from the Citraruvi, located in Courtallam hill, Western Ghats, Tamil Nadu. The specimens were preserved identified and authenticated by the Botanical survey of India – Southern circle (Coimbatore) as *Aglaia elaeagnoidea* (JUSS) Benth.(Meliaceae) were preserved in Department of Botany, Sri Parasakthi College for Women Courtallam (Autonomous) Herbarium, Tamil Nadu, India. The stem and leaves were collected, shade dried, powdered in mechanical pulverized and stored in air tight containers for future use.

MACROSCOPIC AND MICROSCOPIC STUDIES

Macroscopic studies were carried out by simple determination, technique like the shape, size, colour, odour, margin and apex. The stem and leaf specimens were fixed in FAA and microtome slides were prepared and stained.[7, 8] Photomicrographs of with different magnifications were taken with Nikon Labphot 2 microscopic unit.

DETERMINATION OF PHYSICOCHEMICAL PARAMETERS

Total ash value, water and acid, soluble and insoluble ash value, and moisture content were determined as per Indian pharmacopoeia.[9, 10]

FLUORESCENCE ANALYSIS

The fine powders of the samples were examined under visible light and UV light (254nm and 365mm). These powders were also treated with acid, alkali and alcohol and changes in colour were recorded under visible and

uv-light.[11]

DETERMINATION OF EXTRACTIVE VALUE

The powdered bark and leaves were successively extracted with Hexane, Dichloromethane, Ethyl acetate, Ethanol, and water in a soxhlet apparatus. The extracts were evaporated using a rotary evaporator and water extract with a freeze dryer. The residues were weighed.

PRELIMINARY PHYTOCHEMICAL ANALYSIS

The preliminary phytochemical analysis of the Hexane, Dichloromethane, Ethyl acetate, Ethanol, and water extracts were carried out using standard methods. The presence and absence of the secondary phytoconstituents were noted.[12, 13]

RESULT AND DISCUSSION

To ensure the quality of herbal products, proper control of starting material is utmost essential. Various techniques are used for the standardization of medicinal plants of therapeutic potential. But identification and evaluation of plant drugs by pharmacognostical studies is still more reliable, accurate and inexpensive. Standardization plays an important role in the production of phytopharmaceuticals of standard quality and purity.[14,15]

MACROSCOPIC CHARACTERS

It is a erect herb, some are small trees up to 5-6 m tall. Stem is dark brown when young, grayish white, branched 0.7 cm to 1.5 cm width. Roots are brown coloured, woody, lateral roots present 0.5 to 3.0 cm width. The leaf is trifoliate, pale green, obanceolate, digitate, sessile and crowded on the young branches, stipules minute and pink coloured flowers (Fig 1&2).

ANATOMY OF LEAFLETS

The leaflets are small thin and have a prominent midrib. The leaf lets are either flat (Fig.1.1) of folded adaxilly up to the middle of the lamina and marginal part of the lamina is bent to a flat posture. Some of the leaflets are totally folded adaxilly into a V-shaped configuration (Fig.1.2). The midrib is 70µm thick and 80 µm wide. The adaxial part of the midrib consists of two or three, wide, thin walled cells are small, squarish and thin walled. There is a single small vascular bundle situated in the median part of the midrib. It is collateral with adaxial cluster of xylem elements (fig.1.3). The vascular bundle is surrounded by a single, layer of parenchymatous bundle sheath. At the lower end of the vascular bundle is situated a wide circular canal. This midrib canal is 30 µm in diameter.

LAMINA

(Fig.2.1) the lamina is 70 µm thick. It consists of an adaxial layer of small thick walled squarish epidermal cells. The abaxial epidermis cells are larger, circular and thin walled. The mesophyll tissue includes adaxial leaves of three layers of vertically elongated palisade cells. On the abaxial side is two layers of short palisade cells. In between the adaxial and axial palisade layers there is a single median layer of squarish, hyaline parenchyma cells (Fig.2.2).

The marginal part of the leaf is as thick as middle part. The epidermal cells of the marginal part are small, circular and thick walled. There is a wide circular canal situated at the margin of lamina (Fig.2.2). The internal structure of the leaf margin is similar to that of the middle part of the lamina.

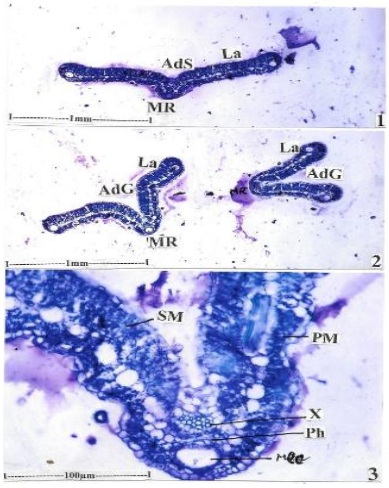

Fig. 1.1; 1.2; 1.3 - T.S. of leaf lamina.

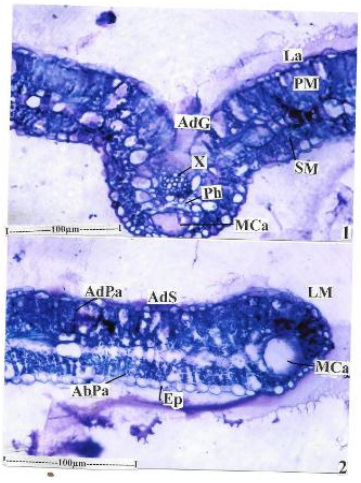

Fig 2.1 ; 2.2 ; - T.S. of midrib of the leaf let.

AdG – Adaxial Groove; Ads- Adaxial side; La- Lamina; MR- Midrib;Mca – Midrib canal; PM- Palisade mesophyll; Ph- Pholem; SM – Spongy mesophyll; X- Xylem, Abpa- Abaxial palisade ; Adpa – Adaxial palisade;Ads- Adaxial side ; Adg- Adaxial Groove; Ep- Epidermis; LM- Leaf margin; Mca- Midrib canal; Mac – Marginal canal; PM- Palisade mesophyll; Ph- Pholem; X- Xylem).

STEM

(Fig. 3.1.3.2 & 4.1, 4.2) shows the stem is circular in sectional view measuring 2mm in diameter. The stem consists of cortical zone with sclerenchyma masses, narrow secondary phloem which surrounds a thick, dense hollow secondary xylem cylinder and central wide parenchymatous pith (Fig.3.1).

The epidermal layer of the stem is intact and in consists of elliptical cells with thick outer tangential walls. The cortex is narrow and it includes four or five layers of angular, thin walled compact parenchymatous cells. Inner of the cortex is a thick, cylinder of isolated masses of sclerenchyma cells (Fig.3; 4.1).

The secondary phloem is wide and includes radial compact lines of sieve elements and parenchyma cells (Fig.4.1).Secondary xylem is a hollow cylinder and it includes vessels and xylem fibres. The vessels occur in several widely separated, uniseriate long radial multiples (Fig. 3.1). The vessels are angular fairly wide and thick walled. Both wide and narrow vessels occur in the radial lines. The wide vessels are 20 µm in diameter; the narrow vessels are 10 µm in diameter (Fig.4.2). In xylem fibres are thick walled and lignified. The cells are angular and occur in compact radial lines. The xylem rays are thin one cell in thickness, straight. The ray cells are thick walled and lignified (Fig. 3.2).

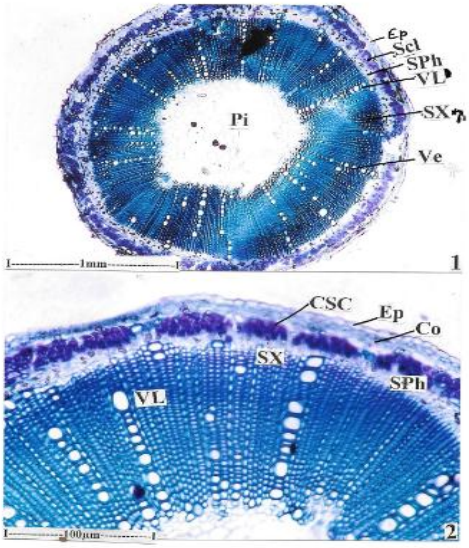

Fig 3.1& 3.2- T.S. of stem – Entire view

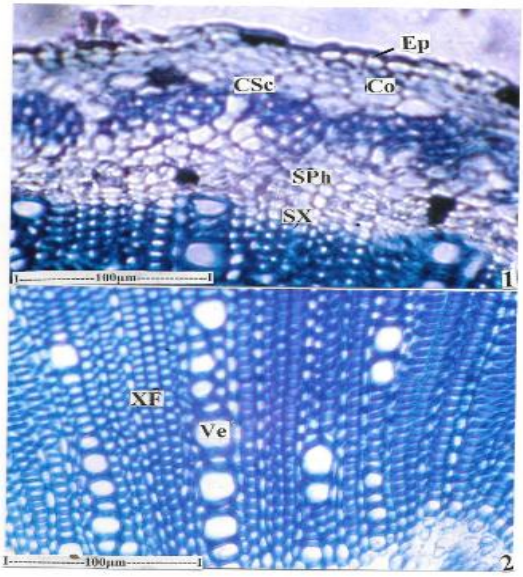

Fig 4.1 & 4.2 T.S. of stem, cortex and secondary phloem (Co – Cortex; CSC – Cortical sclerenchyma ; Ep- Epidermis ; Scl- Sclerenchyma; Sph- Secondary phloem; SX- Secondary xylem; Pi- Pith; VL- Vessel line ; Ve- Vessel, XF- Xylem fibre).

Table 1: Determination of Ash Values, Moisture content and Extractive values.

Parameters	**Ash Value (%)**
Loss of weight on drying	50.65%
Total ash	**94%**
Water soluble ash	2.3%
Sulphated ash	88%
Solvents	**Extractive values (gm)**
Petroleum ether	27%
Chloroform	38%
Acetone	30%
Ethanol	33%
Benzene	55%
Water	47%

Table 3: Fluorescence Analysis of whole plant powder of *Indigofera aspalathoides.*

S.NO	SAMPLE	LEAF		
		Day light	UV (365nm)	UV (254nm)
1	PLANT POWDER	Green	Pale green	Dark green
2	1N Hcl	Green	Pale green	Dark green
3	Nitric acid	Brick red	Dark red	orange red
4	Picric acid	Yellowish green	Pale green	Dark green
5	50% sulphuric acid	Light green	Fluorescence green	Dark green
6	Aqueous NaOH	Dark green	Light green	Green
7	Alcoholic NaOH	green	Pale green	Fluorescence green
8	1 N NaOH	Green	Pale green	Fluorescence green
9	Acetic acid	Green	Pale green	Dark green
10	Ferric chloride	Red colour	Brick red	Brownish red
11	Picric Acid	Yellowish green	pale Green	Dark Green

PHYSICOCHEMICAL PARAMETERS AND PRELIMINARY ANALYSIS

The residue after incineration of plant materials are the ash. The ash value represents the inorganic salts naturally occurring in the crude drug. Total ash was more (94%), followed by sulphated ash(88%) than other ashes. The moisture content of the drug was 50.65% (Table 1). Among the various extracts the results showed greater extractive values in benzene (55%) and water(47%)(Table 2). Ethanol, petroleum ether indicated the concentration of secondary metabolites in the whole plant powder sample. The whole powder showed a specific and diagnostic colorations under ordinary day light and UV light (Table 3). This character is distinct for each species and can be used as a diagnostic feature in the identification of crude drug. Preliminary phytochemicals analysis of various extracts of ***Indigofera aspalathoides*** revealed that the presence of alkaloid, saponins, phenol, glycosides, steroids, flavonoids, amino acids, anthroquinones, catechin, coumarins, quinones and carbohydrates.The concentration of secondary metabolites was strong in ethanol extract of stem than the leaf (Table 4).[19, 20] These secondary metabolites are known to support bioactivity in this plant because these chemicals can interact with the metabolic activities of the microorganism which cause diseases.

CONCLUSION

The present investigation of *Indigofera aspalathoides* Vahl. Exdc. can be concluded that this pharmacognostic study yielded a set of parameters which could serve as an important source of information to ascertain the identity and determination of quality and purity of plant material for future studies. This simple but reliable standardization will be useful to a lay person in using the drug as home remedy and also in the pharmaceutical industry for testing the raw material.

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