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PHARMACOGNOSTICAL STUDIES ON THE ENDEMIC MEDICINAL PLANT – AGLAIA ELAEAGNOIDEA (JUSS.) BENTH. (MELIACEAE)

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ABSTRACT

Aglaia species are known for many biological activities like antipyretic, analgesic, anticancer, hepatoprotective, antidiabetic, anti-inflammatory, immunomodulatory and insecticidal activity. Aglaia elaeagnoidea (JUSS) Benth. (Meliaceae) is an endemic medicinal tree of peninsular India. The stem and leaf of this tree are used by the rural folk similar to other Aglaia species in curing skin diseases, fever,

dysentery, cooling, astringent, abdominal pain, emetic and excessive menstruation. The present investigation is intended to evaluate the pharmacognostical features of stem and leaf of this species. The pharmacognostical parameters were carried out by complete botanical evaluation which includes macroscopic, microscopic, physicochemical and phytochemical analysis. Pharmacognostical standard serve as a reference piece and helps in the further identification and authentication of this taxon.

KEYWORDS: Aglaia elaeagnoidea, Pharmacognosy, physicochemical, phytochemical analysis.

INTRODUCTION

People are becoming more aware of medicinal plant resources and utilize these therapeutic interventions and their products in maintaining health and preventing diseases with an ecofriendly touch. Herbal medicines are the 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] *Aglaia elaeagnoidea* (JUSS) Benth. (Meliaceae) is an endemic medicinal tree of peninsular India commonly called as "Chokkala". 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, WesternGhats, Tamil Nadu. The specimens were preserved identified and authenticated by Director, Institute of Herbal Botany, Plant Anatomy Research Centre, Chennai. *Aglaia elaeagnoidea* (JUSS.) Benth.(Meliaceae) was preserved in Department of Botany, Ayya Nadar Janaki Ammal College (Autonomous) Herbarium, Sivakasi, Tamilnadu, 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 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

Aglaia elaeagnoidea (JUSS) Benth. is a tree to 10 m, with thick stem and root stock. Leaves are compound, leaflets alternate, 3-7 cm long, oblanceolate, with entire margin and bluntly subacuminate tip; petioles and rachis 3-10 cm long. Inflorescences are axillary panicle, shorter than leaves, lepidote scaly 5-10cm long. Flowers roundish polygamodioecious, yellow. Fruit berry, globose 1.2-2cm long. Seed is buff, two loculed, seeds 1 per locule(Fig 1&2).

Microscopic characters of stem

The stem is thick and woody and measures 2.3mm thick. The outer surface consists of thin periderm, and shows the tendency of exfoliating in the form of thin membrane (Fig 3). The cortical zone is fairly wide and parenchymatous. The cortical vascular bundles are collateral with inner few parallel lines of xylem and outer tangential segment of phloem (Fig 4). There are also scattered small masses of fibres found in the cortex. The vascular cylinder is thick and dense with central hollow parenchymatous pith. Secondary phloem occurs in thick cylinder comprising of compact radial lines of sieve elements and their companion cells. In the outer part of the phloem occurs collapsed phloem where the sieve elements are crushed into thick, tangential dark bands. The non-collapsed secondary phloem is 80µm thick. The

secondary xylem is thick hollow cylinder $250\mu m$ thick. It consists of diffusely distributed solitary or less frequently radial multiples of 2 or 3 xylem elements. The vessels are circular, thick walled, mostly solitary or rarely in segments of 2 or 3 elements. The xylem fibres are highly thick walled with narrow, reduced lumen and thick walled lignified fibres. The vessels are wide and circular measuring $40\mu m$ in diameter. The pith is parenchymatous with scattered masses of pith fibres. The pith fibres give mechanical support to the plant tissues.

Transverse section of the bark

The bark consists of deep seated periderm, which is located inner to 2 or 3 cylinder of phloem fibres (Fig.9). The periderm is exposed externally and includes outer phellem and inner phelloderm cells are tabular in shape and are in compact radial files. The cells possess dense accumulation of tannin. The total thickness of periderm is 100µm.

The secondary phloem is very wide consisting of several successive thin cylinders of phloem fibres and tannin containing parenchyma cells (Fig.9). The zone is collapsed phloem and includes crushed tangential lines of sieve elements (Fig.9). The cells of the collapsed phloem have tannin accumulation and thick cylinders of phloem Sclerenchyma cylinders are seen with crushed sieve elements. The inner part of the secondary phloem is narrow zone of non-collapsed phloem. The non-collapsed phloem has narrow phloem rays, intact sieve elements and narrow phloem parenchymatous cells. The sieve elements are angular and occur in compact radial files. The companion cells are located on the lateral walls of the sieve element. The phloem fibres and tannin containing cells are absent or reduced in the non-collapsed phloem (Fig.9).

Leaf

The leaf consists of a thick and prominent Plano convex midrib and smooth and even bilateral symmetrical lamina. The midrib consists of flat adaxial side and wide and thick semicircular abaxial side.

Lamina

The lamina is 230µm thick. It is dorsi ventral and hypostomatic. Some of the palisade cells are highly dilated into wide circular idioblasts and possess a rhomboidal calcium oxalate crystals. Usually only one crystal containing idioblast is 50µm in diameter.

Epidermal tissue

Adaxial epidermal cells are wide, polygonal and compact. The anticlinal walls are fairly thick and straight. The epidermal layer is apostomatic. The abaxial epidermis is stomatiferous. [15, 17] The epidermal cells are comparatively small, polygonal and thin walled. The stomata seem to be cyclocytic type with one or two circles of curved rectangular subsidiary cells. The guard cells are elliptical measuring $20\times20\mu m$ in size. The subsidiary cells are thin walled and encircle the guard cells (Fig 7).

Venation Pattern

The venation pattern as seen from surface is reticulate with fairly distinct vein islets and well developed vein terminations (Fig.5). The vein islets are wide and have thick straight vein boundaries. Within the islet occur either simple or branched vein terminations. The simple vein terminations are curved and slightly dilated at the tip (Fig.5). The branched terminations are dendroid in outline with repeated branch –lets. They are also wavy with dilated ray. Calcium oxalate prismatic crystals are seen in surface view located in the dilated epidermal cells of the leaf (Fig.6). The crystals are $30\times30\mu m$ in size.

Petiole

Both proximal part and distal part of the petiole were studied. The general structure of the distal part of the petiole is similar to proximal part of the petiole. The proximal petiole is circular with flat adaxial side. It is 1.4µm thick. The distal part of the petiole has flat adaxial side with short, thick lateral wings. The petiole consists of a thin layer of semicircular thick walled squarish epidermal cells with thick cuticle. The outer ground tissue includes about 7 layers of collenchymatous tissue and inner zone of circular or angular thin walled parenchymatous ground tissue. The vascular strand is semicircular with adaxial slightly depressed part. It consists of several, parallel rows of narrow thick walled vessel elements and thick walled fibres. The xylem strand consists of incurved adaxial margins and semicircular abaxial part. Outer to the xylem strand occurs a thin layer of small, discrete masses of phloem elements and sclerenchymatous thick sheath enclosing the entire vascular strand (Fig 8.2). In distal part of the petiole the vascular strand consists of arc shaped adaxial part with radial rows of xylem elements and fibres with phloem situated along the outer part. The adaxial median part includes a slightly concave thick segment of xylem strand. Fibres occur both on the outer phloem zone as well as along the inner boundary of the xylem. [16]

Crystal distribution

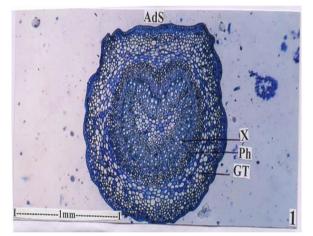
Calcium oxalate crystals of prismatic type and druses type are abundant in leaf and stem. The crystal characters are also one of the important key character in identifying the family Mahogany^[18] (Meliaceae).





Fig: 1. Fig.2.

Twig of Aglaia elaeagnoidea (JUSS) Benth. Bark of Aglaia elaeagnoidea (JUSS) Benth.



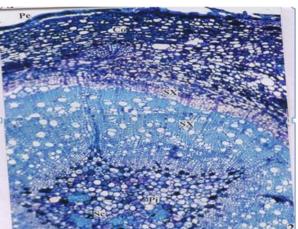
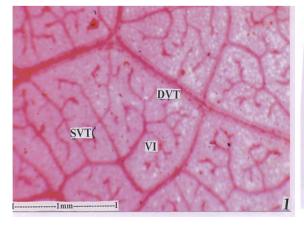


Fig 3. T.S. of Stem

Fig 4. T.S of stem portion enlarged



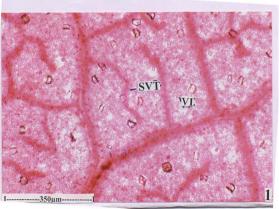


Fig 5 Fig 6

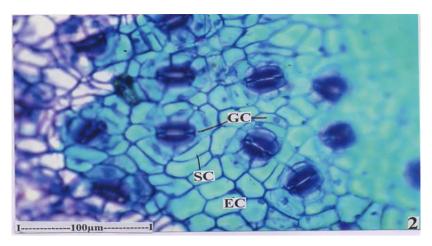


Fig 7 Stomatal morphology

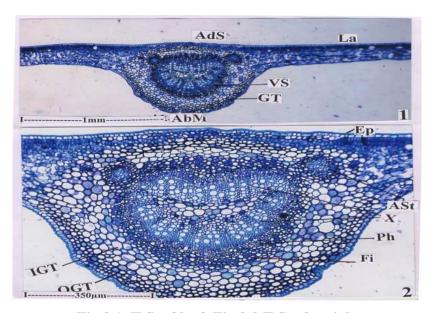


Fig 8.1, T.S. of leaf, Fig 8.2 T.S. of petiole

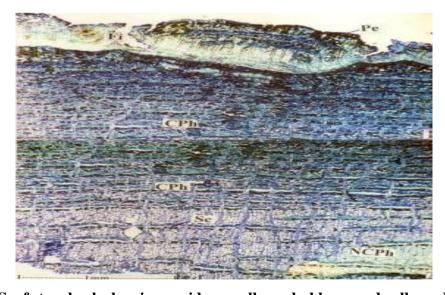


Fig 9. T.S. of stem bark showing periderm collapsed phloem and collapsed phloem.

(Cph- Collapsed phloem, Fi- fibre, Ncph- Non collapsed phloem, Pe- Periderm, Sc-Sclerenchyma, Sph- Secondary phloem, AbE – Abaxial epidermis, AbM- Abaxial Midrib, AdE- Adaxial Epidermis, AdS- Adaxial Side, Cu- Cuticle, Cr- Crystal, Ep- Epidermis, Fi- Fibre, GT- Ground tissue, IGT- Inner ground tissue, OGT- Outer ground tissue, La-Lamina, Ph- Phloem, X- Xylem, Ast- Accessory Strand, DvT- Dendroid vein termination, SvTr- Simple vein termination, VI- Vein Islet, Pe- Periderm, Sc- Sclerenchyma, pi- Pith, GC-guard cells, Sc- Subsidiary cells, St- Stomata).

Table 1: Determination of Ash Values and Moisture content

Downwotons	Ash Value (%)					
Parameters	Leaf sample	Bark sample				
Total Ash value	6.23	8.32				
Acid insoluble ash	16.1	5.2				
Acid soluble	33.24	3.34				
Water soluble	12.24	1.78				
Sulphated Ash	3.13	2.22				
Moisture content (%)	3.1	4.2				

Table 2: Determination of Extractive values

Solvents	Extractive values (gm)					
Solvents	Leaf sample	Bark sample				
Hexane	0.9	0.05				
Dichloromethane	1.7	0.61				
Ethyl acetate	1.1	0.798				
Ethanol	2.5	1.5				
Water	3.2	2.5				

Table 3: Fluorescence Analysis of leaf and Bark powder

	Leaf powd	er		Bark Powder			
Sample	Visible	UV	UV	Visible	UV	UV	
	light	(254nm)	(365nm)	light	(254nm)	(365nm)	
Powder	Green	Pale	graan	Brown	Dark	Brown	
1 Owder	Green	green	green	Diowii	brown		
Powder+1N cl	Green	Pale	Light	Dark	Dark	Brown	
	Green	green	green	brown	green		
Powder+HNO3	Brown	Green	black	Brown	dark green	Green	
Powder Picric	Yellowish	Pale	Light	Yellowish	Dark	Green	
acid	green	green	green	brown	green	Green	
Powder+50%	dark	Green	Pale green	Brown	dark green	Green	
sulphuric acid	green	Green	raie gieen	BIOWII	dark green	Green	
Powder+	Brownish	Green	Light	Dark	Bright	Green	
Aqueous NaOH	green	Gicell	green	brown	green	Green	

Powder+ Alcoholic NaOH	Brown	Dark green	Yellowish green	Yellow	green	Pale green
Powder+Nitric acidwith NH3	Light green	dark green	brown	Brown	light green	Green
Powder+Acetic acid	Pale green	Light green	brown	Brown	Dark green	Pale green
Powder+Ferric chloride	Yellowish green	Dark green	green	Yellowish green	Dark green	Green

Table4: Preliminary phytochemical analysis of various extracts of leaf and bark samples.

Phytochemical	Leaf sample					Bark sample				
Test	Hexane	Dichloro Methane	Ethyl acetate	Ethanol	Water	Hexane	Dichloro methane	Ethyl acetate	Ethanol	Water
Carbohydrates	+	-	-	+	+	+	+	+	+	+
Tannins	-	-	-	-	-	-	-	-	-	-
Saponin	-	+	-	-	-	-	+	+	-	-
Flavonoid	-	-	+	+	-	+	+	++	++	++
Alkaloid	-	++	++	+	-	+	++	++	++	-
Quinones	+	+	-	+	+	-	-	-	-	++
Glycosides	-	-	-	-	-	-	-	-	-	-
Cardiac glycosides	++	++	-	+	+	++	++	++	++	+
Terpenoids	-	-	-	-	-	-	-	-	-	-
Phenols	+	+	+	+	+	-	-	-	+	++
Coumarins	-	-	+	+	-	++	++	++	++	++
Steroids and Phytosteroids	++	+	-	+	+	-	-	-	-	-
Phlobatannins	-	-	-	-	-	-	-	-	-	-
Anthraquinones	-	-	-	-	-	-	-	-	-	-

⁺ Present, ++ strongly present, - absent

Physicochemical Parameters and Preliminary Analysis

The residue after incineration of plant materials is the ash. The ash value represents the inorganic salts naturally occurring in the crude drug. It was more in bark (8.32%) than the leaf (6.23%). The moisture content of the drug was more in stem than the leaf (Table 1). Among the various extracts, the results showed greater extractive values in water extract followed by ethanol and indicated the concentration of secondary metabolites in both samples (Table 2). The leaf and stem 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 *Aglaia elaeagnoidea* revealed 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). These secondary metabolites are known to support bioactivity of this plant because these chemicals can interact with the metabolic activities of the microorganism which cause diseases.

CONCLUSION

The Pharmacognostical and phytochemical evaluation of *Aglaia elaeagnoidea* (A.Juss) Benth leaves and bark provide useful information for identification and authentication of plant. This study yielded a set parameters which could serve as important source of information to ascertain the identity and determination of quality and purity of plant material for future studies.

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