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PHARMACOGNOSTICAL ADN PHYSICOCHEMICAL ANALYSIS OF MALLOTUS PHILIPPINENSIS, (LAM.) MUELL. ARG.

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ABSTRACT

Mallotus philippensis, (Lam.) Muell Arg. plants are used as a purgative, anthelmintic antioxidant, antitumour, antiallargic activity, wound healing and microbial infections. In order to ensure the use of only genuine and uniform material in preparation of herbal formulation, work on standardization was carried out. Macroscopic, microscopic and physico-chemical characters determination have been carried out, which would facilitate quick identification and selection of the drug from various adulterants.

KEYWORDS: Pharmacognosy, Mallotus philippensis, Physicochemical, microscopic, Florescence analysis.

INTRODUCTION

In the last few decades there has been an exponential growth in the field of herbal medicine. It is getting popularized in developing and developed countries owing to its natural origin and lesser side effects. In olden times, vaidyas used to treat patients on individual basis, and prepared drugs according to the requirement of the patients. But the scene has been changed now, Herbal medicines are being manufactured on a large scale in mechanical units, where manufacturers are facing many problems such as availability of good quality raw material, authentication of raw material, availability of standards, proper standardization methodology of drugs and formulations, quality control parameters^[1, 2]</sup> and *etc*. Correct knowledge of such crude drugs is very important aspect in the preparation, safety and efficacy of the herbal products. Pharmacognosy is a simple and reliable tool, by which complete information of the crude drug can be obtained.^[3-5] There is a need for documentation of research work carried out on traditional medicines.^[6] Hence, it becomes extremely important to make an effort towards standardization of the plant material to be used as drugs. The process of standardization can be achieved by stepwise pharmacognostic studies.

Mallotus genus an ethanomedicinal plant of *Mallotus philippensis* (Lam.) Muell Arg locally known as "Kamala" and "Kapilapodi" belonging to family *Euphorbiaceae*. It is a multipurpose, medium sized tree much branched, tolerant and soil improving small tree. It is up to 10-12 meters in height and is widely distributed, throughout tropical India along with the Himalaya from Kashmir East wards up to 5000 ft and subtropical region including the Punjab, Uttar-pradesh, Bengal, Assam,

Burma, Singapore and Mumbai. Artificial propagation is done by sowing fresh seeds in April. The various parts of *Mallotus philippensis* are used in the treatment of skin problem, bronchitis, fungal disease, eye – disease, cancer, diabetes, jaundice, malaria, urinogenital infection, etc.^[7-9] Plants have played an important role in medicine in – vitro Hence, the present study was undertaken to study the pharmacogstical and physicochemical analysis of the leaves of *Mallotus philippensis* (Lam.) Muell. Arg.

MATERIALS AND METHODS

PLANT COLLECTION AND AUTHENTICATION

The aerial parts of the plant were collected from the Citraruvi, located in Courtallam hill, Western Ghats, Tamil Nadu. The plant was identified by Prof.P.Jayaraman, Plant Anatomy Research Centre, West Thambaram, Chennai, Tamil Nadu and the voucher specimen were preserved in Department of Botany, Sri Courtallam Parasakthi College for Women (Autonomous) Herbarium, 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 with toluidine blue.^[10-12] Descriptive terms of the anatomical features are as given in th standard books.^[13] Photomicrographs of with different magnifications were taken with Nikon Labphot 2

microscopic unit.

DETERMINATION OF PHYSICOCHEMICAL PARAMETERS

Physicochemical characters such as ash value, extractive values and loss of weight on drying were determined as per Indian pharmacopoeia.^[14-16]

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.^[17-18]

RESULT AND DISCUSSION

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.

ANATOMY OF THE LEAF

The leaf even and smooth on the adaxial side and highly undulate and densely hairy on the abaxial side. The midrib is very thick projecting much below the lower side of the leaf (Fig. 1.1 - 1.3). The midrib is slightly revised above the adaxial side. It is thick and semicircular on the abaxial side. It is 950 µm thick and 900µm wide. The epidermal layer of the midrib consists of small thick walled and darkly stained cells with echinate tangential outer walls. The ground tissue includes circular compact parenchyma cells. Four or five layers of cells inner to the epidermis are small thick walled collenchyma cells. Both adaxial and abaxial strand are collateral. The xylem elements occur in short more or less compact vertical lines. The xylem elements are angular and thick walled. The lumen of the cell is wide (Fig. 2.). Pholem occurs in the form of an arc beneath the abaxial median vascular bundle is located along the outer border of the xylem segment. The xylem strands of adaxial and abaxial bundles are just aposed (Fig. 2).

LATERAL VEIN

The lateral vein hangs from the abaxial surface of the lamina. It is thick and prominent. It is basically similar to main midrib bundles. It has adaxial and abaxial vascular bundles which collateral and the xylem elements of the two bundles are aposed (Fig. 1.2).

CRYSTAL DISTRIBUTION IN THE MIDRIB

Calcium oxalate crystals of druses are sparsely distributed in the pholem parenchyma and ground parenchyma cells (Fig.3.1). The druses are variable in size. The druses occur in single in the parenchyma cells.

In the pholem parenchyma cells the druses are smaller in size than those located in the ground parenchyma.

STRUCTURE OF THE LAMINA

The adaxial surface of the lamina is smooth and even. The abaxial surface is prominently uneven with thick ridges and furrows. The ridges are due to thick prominently projecting lateral veins. The furrows are formed by presence of large glandular trichomes. The glands have a short thin stock and funnel shaped wide secretary body. The body of the gland is multicellular with darkly stained cells. The glandular trichomes are 50µm in size and 40 µm wide. Apart from the glandular trichomes there are dense cluster of stellate trichomes of non-glandular type are seen (Fig.3.2). The mesophyll tissue includes 4 or 5 layers of vertically elongated compact rectangular cells. There is no distinct differentiation of palisade and spongy mesophyll tissue. The lamina is 100 µm thick. In the region of lateral veins it is 160 µm thick.

STEM

The stem is circular in cross sectional view measuring $3.4 \mu m$ in diameter. The stem consists of intact epidermal layer, thin, cortex, cortical gelatinous fibres and dense hollow vascular cylinder. There is a central pith where the cells are disintegrated (Fig.4.1).

The epidermal cells of the stem are vertically elongated with darkly stained cell inclusions. The cortical one includes about six layers of polygonal thin walled compact parenchyma cells. In the inner boundary of the cortex occurs a thin continuous layer of gelatinous fibres (Fig.4.2 & 5.1). The fibres have inner mucilaginous cells walls. The inner cell wall stain dark purple in colour with toluidine blue stain (Fig.5.1). The secondary phloem is well developed and wide. It includes sieve elements and parenchyma cells in compact radial lines. The phloem rays are slightly wider and straight. The sieve elements are rectangular in shape and the companion cells are located on the lateral art of the sieve elements (Fig.5.1). The secondary xylem includes short radial multiples of vessels or solitary vessels. They are mostly elliptical in shape. Some of the vessels are circular and cell the vessel elements are thin walled. The fibres are thick walled and lignified. The xylem rays are fairly prominent and straight. They include radially oblong thin walled cells.

Calcium oxalate crystals are fairly abundant in the secondary xylem parenchyma cells. They are druses type (Fig.5.3). Prismatic crystals also sparsly seen in the parenchyma cells outside the gelatinous fibre.

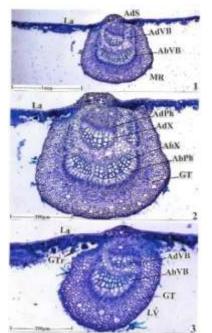


Fig.1.1 – T.S. of leaf through midrib Fig. 1.2. – T.S. of midrib – Enlarged Fig.1.3. – T.S. of leaf through lateral vein

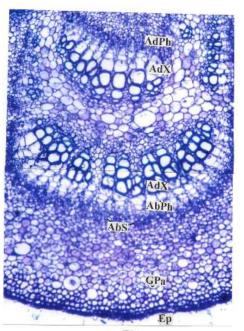


Fig.2.- T.S of midrib of the leaf- Enlarged

(Ads- Adaxial side; AdvB- Adaxial vascular bundle; AbvB- Abaxial vascular bundle; Adph- Adaxial Pholem; Adx- Adaxial xylem; GT- Ground tissue; La- Lamina; LV- Lateral vein; MR- Midrib; Adph – Adaxial phloem; Adx – Adaxial xylem; Abx- Abaxial xylem; Abph- Abaxial phloem; E- Epidermis; Gpa- Ground parenchyma)

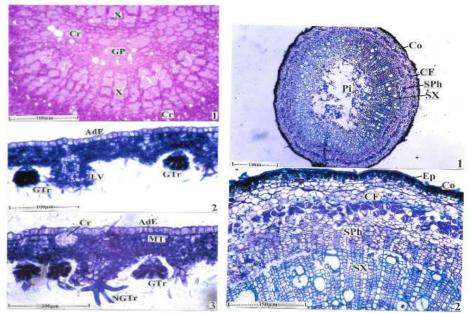


Fig 3.1. – Calcium oxalate crystals distribution in the midrib as seen under polarized light Fig. 3.2. T.S. of lamina showing lateral vein and abaxial concavity with glandular trichomes. Fig.4.1. T.S of stem – Entire view, Fig 4.2- T.S. of stem - A sector enlarged.

(AdE- Adaxial Epidermis; Cr- Crystal; G- Ground parenchyma; GTr- Glandular trichome; Lv- Lateral vein; MT- Mesophyll tissue; NGTr- Non – glandular trichome; X- Xylem; Co – Cortex; CF – Cortical fibre; Ep – Epidermis; Pi – Pith; Sph – Secondary phloem; SX- Secondary xylem)

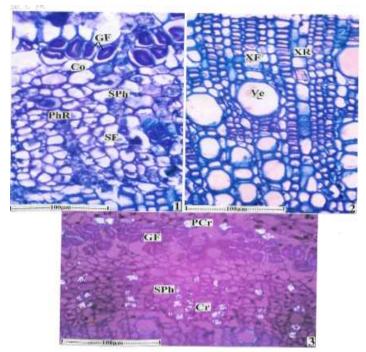


Fig. 5.1 – T.S. of secondary phloem of the stem.

Fig. 5.2 – T.S. of secondary xylem of the stem.

Fig 5.3. – Crystals distribution in the phloem parenchyma of the stem.

(Co- Cortex; GF- Gelatinous fibre; Cr- Crystal; phr – phloem Ray; Pcr – prismatic crystal; Sph – Secondary phloem; SE – Sieve Elements; XF – Xylem fibre; XR – Xylem Ray; Ve – vessel)

Table 1: Physicochemical characters of leaf	powder of <i>Mallotus</i>	philippensis	(Lam.) Muell. Arg.
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S. NO	Description	Percentage (%)
1	Loss of weight on drying	61%
2	Total ash	91%
3	Acid insoluble ash	2.3%
4	Water soluble ash	1.6%
5	Sulphated ash	89%
6	Petroleum ether	43%
7	Chloroform	93%
8	Acetone	66%
9	Ethanol	43%
10	Benzene	42%
11	Water	59%

Table 2: Fluorescence analysis of leaf powder of *Mallotus philippensis*(Lam.) Muell. Arg.

		Leaf		
S.No	Sample	Day	UV	UV
		Light	(365nm)	(254nm)
1	Plain Powder	Green	Pale green	Dark Green
2	1N HCl	Green	Dark Green	Fluorescent Green
3	50% H ₂ SO ₄	Pale Green	Green	Dark Green
4	Aqueous NaoH	Brown	Pale Brown	Dark Brown
5	Alcoholic NaoH	Brick Red	Pale Brick Red	Dark Brick Red
6	NaoH	Orange	Yellowish green	Dark Orange
7	HNo ₃	Orange	Yellow Brown	Dark Brown
8	Ferric chloride	Green	Dark Green	Brown colour
9	Ammonia	Green	Dark Green	Fluorescent Green
10	Acetic acid	Fluorescent Green	Green	Dark Green
11	Picric Acid	Pale Green	Yellowish Green	Dark Green

PHYSICOCHEMICAL PARAMETERS

The percentage of loss of weight on drying, total ash, acid insoluble ash, water soluble ash, and sulphated ash were obtained by employing standard methods of analysis and the moisture content of the *Mallotus philippensis* was 61%. The total ash content of the leaf was found 91%, the acid insoluble ash content of the leaf was found 2.3%, the water soluble ash content of the leaf was found 1.6, and the sulphated ash content of the leaf was found 89%. The extractive values obtained from different solvents were found in the *Mallotus philippensis* among all the extracts chloroform(93%), acetone(66%), and distilled water (59%) showed highest percentage and presented in Table -1.^[19]

FLUORESCENCE ANALYSIS

The fluorescence analysis of the plant powder of *Mallotus philippensis* were determined and presented in Table 2. The powder was examined under ordinary light and UV light (short UV 254nm, Long UV 365nm). The powdered plant drug emitted the characteristic fluorescent green colour when treated with 1N HCl, acetic acid and ammonia, under short UV (254nm), plant drug emitted yellowish green when treated with aqueous NaOH and picric acid, under 365nm and plant drug emitted dark green when treated with ferric chloride, ammonia, and picric acid under 254nm.^[20] This characteristic fluorescence can be used as a diagnostic tool for the correct of the crude drug and to test adulteration if any.

CONCLUSION

In conclusion, it can be stated that the standardization parameters used in the present investigation will provide a way for the standardization of raw materials and prepared formulations of herbal origin as well as answer to the latest GMP norms and FDA guidelines on standardizations of herbal drugs. This could also serve in the identification and preparation of a monograph on the plant.

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