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Research Article

Morpho-Anatomical Features on *Blumea Mollis* (D. Don) Merr. (Asteraceae) Leaves

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Abstract

Background: *Blumea mollis* belonging to Asteraceae family is a significant therapeutic herb which has been in therapy utilized to treat several pathological marque since ages. It can be commonly referred as Suvattru mullangi in telugu. Though it is an essential herb, till date, no pharmacognostical information had been available on its leaves. Numerous adulterations are located in the market.

Objective: The current research was carried out to analyze the Pharmacognostic details for the rapid recognition and authentication of the herb. Materials and methods: The macroscopic and microscopic features with i quantitative microscopy of *Blumea mollis* leaves were performed utilising distinctive chemicals and reagents.

Results: The plant leaves show single layered, wavy walled cells in upper epidermis. Powder study of leaves shows epidermal cells, pigment cells, anomocytic stomata, covering trichomes and lignifie xylem vessles.

Conclusion: The macroscopic and microscopic characteristics of *Blumea mollis* leaves serves as a tool for low cost, rapid identification and authentication of this plant.

Keywords: Blumea mollis; anomocytic stomata; phytochemical analysis; physicochemical parameters.

Introduction

The usage of natural products or natural product-based medication is strengthening worldwide, particularly in the expanding parts of the world, despite the fact that synthetic medicines are readily available and reliable in healing several illnesses, there are people that still choose using traditional folk medicines because of the fewer hazardous outcome. Around 25% of the prescribed medicines on the globe will be of basically plant source [1]. In the developing countries like India, around 80% people depend on traditional plant-based medicines for their prime health care desires [2].

Modern prevalent desire for plant-derived medicines demonstrates its acknowledgement of the validity of numerous traditional promises about the values of natural products in healthcare [3]. For quality control of conventional medications, phytochemical inspections are mostly employed. Therefore, it creates an excellent value to look at chemical constituents and examine pharmacological activity about this herb because of its therapeutic applications, which is very helpful in the field of medicine as new emerging drug [4]. According to the WHO, medicinal plants are the best sources to obtain a variety of new herbal drug.

Blumea mollis (Asteraceae) is a genus of flowering plants widely distributed in Western and Southern plains of India ascending to 2000 ft in the Himalayas [5]. *Blumea* mollis is an agreeably fragrant annual herb with 30- to 60-cm height and generally seen in the flatlands of India, outer Himalaya, Ceylon (veraltet) and Myanmar. It is an annual erect herb, up to 60cm tall; brenchlets ribbed, pubescent. Leaves obovate, 3.5-9.5 X 1-3.5cm, base attenuate,

margin irregular serrate, tip acute, pubescent on both sides. Heads in axillary and terminal corymbs. Achenes angled, sparsely hairy, not ribbed [6]. The leaf of the herb is historically used for skin diseases, and the boiled herb is used for diarrhea [7]. The studies of gas chromatography and GC-mass spectroscopy (GC-MS) affirm that the leaf volatile oil of *B.mollis* comprised 39 substances, and the main chemical substances recognized were linalool, γelemene, copaene, estragole, Allo-ocimene, γ-terpinene and Alloaromadendrene [8]. The genus *Blumea* has reported various pharmacological activities including Anti-bacterial activity, anticancer, hepatoprotective, anti-oxidant, anti-microbial, anti-inflammation, anti-plasmodial, anti-tyrosinase, platelet aggregation, wound healing, anti-obesity and insect resistant activities [9,10].

Though this plant is widely used for its multiple properties, it has not been standardized pharmacognostically and chromatographically. Therefore, the intention of the current research is to assess many pharmacognostic criteria like macroscopy, microscopy, ash values, extractive values, microscopical characters of powdered leaf and development of HPTLC fingerprinting pattern for *B. mollis* leaves.

Materials and Methods

Collection and authentication of plant materials

The leaves of *B. mollis* were collected from V. V. Institute of Pharmaceutical Sciences, Gudlavalleru, Andhra Pradesh, India. The plant specimen was identified by Dr. K. Madhava chetty, Plant taxonomist, Sri Venkateswara University, tirupati.

Organoleptic evaluation

Leaves of *B. mollis* were assessed for their effect on different organs of sense for organoleptic properties. Its color, odor, size, taste and other diagnostic variables was noticed and noted.

Macroscopic analysis

Macroscopic features of the leaf i.e. type of leaf, colour, shape, apex, margin, lamina, base venation and texture had been examined by standard approach.

Microscopic analysis

Study of transverse section

For microscopical investigations, gathered material was washed and fixed in Formalin-Acetic Acid-Ethyl alcohol (FAA) and standard microtome methods had been adopted [11]. Transverse sections of 10 to 15 x 10-6 m thicknesses were carefully taken and stained with phloroglucinol and concentrated HCl. Photographs were taken under a CX-21i Olympus microscope connected to a digital camera. For micromorphological investigation i.e. for leaf constants, fresh material as well as fixed material could be used and standard peel study was followed. Stomatal index, vein islet and vein termination numbers were calculated by using standard methods [12]. For powder study, dried herbal material was finely powdered and then sieved by usingmesh no. 85. The fine powder acquired was stained applying phloroglucinol and Conc. HCl. The stained powder was placed on a slide and then noticed within microscope to find and recognize the botanical characters. The characters noticed had been took pictures of under a CX-21i Olympus microscope attached to a digital camera.

Powdered drug microscopy

The coarsely powder leaf was placed on a glass slide in glycerin and analyzed with microscope. Snap shots of various magnified cellular structures had been undertaken with CX-21i Olympus microscope attached to a digital camera.

Analysis of leaf constants

Several parameters of leaf constants had been assessed implementing standard techniques.

Proximate analysis

The various physiochemical parameters like ash values, extractive values and moisture content had been based upon the standard methods [13,14].

Preparation of extracts

Initially, the plant leaves had been rinsed with normal water to eliminate dirt and other foreign matters and then had been segregated and shade dried. Dried leaves had been then milled to coarse powder after which surpassed above sieve No. 14. The acquired dried out powder leaves of B. mollis (500 g) had been positioned in the tube of Soxhlet apparatus by means of thimble and then kept on heating mental for 6 h for extraction using various solvents from non-polar to polar. The acquired extracts had been strained while hot and dried by evaporation utilising rotary vacuum evaporator and the final dried extracts products had been retained at low temperature in refrigerator for additional investigation.

Preliminary phytochemical screening

Various extracts of *B. mollis* leaves were subjected to phytochemical analysis [14]. A number of identity assessments had been worked to detect existence of primary and secondary metabolites.

Fluorescence analysis

This study was performed in accordance with the standard methods. In the current research, the plant powder was treated with various reagents [13-15].

Results

Organoleptic and microscopic characters

The Organoleptic characteristics of leaf showed in the Table 1.

Organoleptic characters	Observation
Colour	Green
Odour	Aromatic
Taste	No taste
Size	8 to 13 cm
Texture	Rough
Fracture	Fibrous

Table 1: Organoleptic characteristics of leaf of *Blumea mollis.*

Figure 1: Morphological characteristics of Whole Plant of *Blumea mollis*

Macroscopic characters

The leaves are obovate with 2.5-7 cm long, 1-3.5 cm broad. The lamina of leaf is smooth at both the surfaces with entire margin and an acute apex.

Transverse section

The transverse section of *B. mollis* leaf revealed existence of lower and upper epidermis. The epidermis was guarded with a single layer of cuticle. The vascular bundle was between 2-4 layers of cortex. Xylem was lignified, phloem was non-lignified, vascular bundles were arc shaped. The pith was made up of large cells. The anomocytic stomata had been within epidermis. Plenty of uniseraite multicellular covering trichomes was present along with unicellular head with multicellular, uniseriate stalk glandular trichomes. The prominent diagnostic features of leaf had been arc shaped vascular bundle, anomocytic stomata and xylem vessels. These characters may be used for standardization of drugs and moreover utilized for preparing of herbal monographs.

Figure 2: Transverse Section Midrib portion of *Blumea mollis* leaf. Cu: Cuticle; UE: Upper epidermis; Par: Parenchyma; Xy: Xylem, Ph: Phloem; CT: Covering Trchomes; Col: Collenchyma; LE: Lower epidermis.

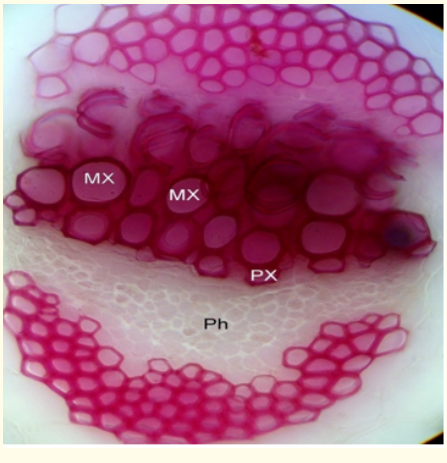

Figure 3: Transverse Section of Midrib portion of *Blumea mollis* showed Vascular Bundles. Ph: Phloem; Xy: Xylem; Mx: Metaxylem; Px: Protoxylem.

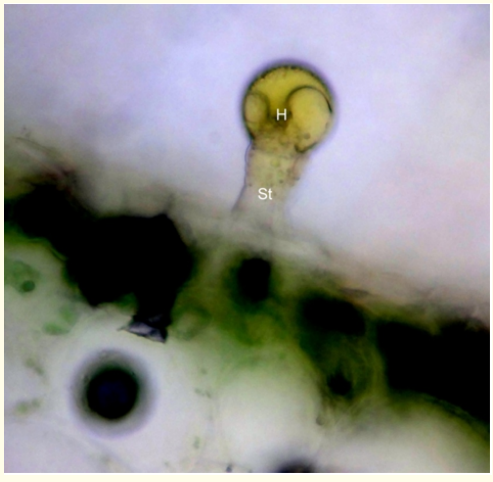

Figure 4: Epidermal cells of *Blumea mollis* leaf showed Glandular trichome. H: Head; St: Stalk.

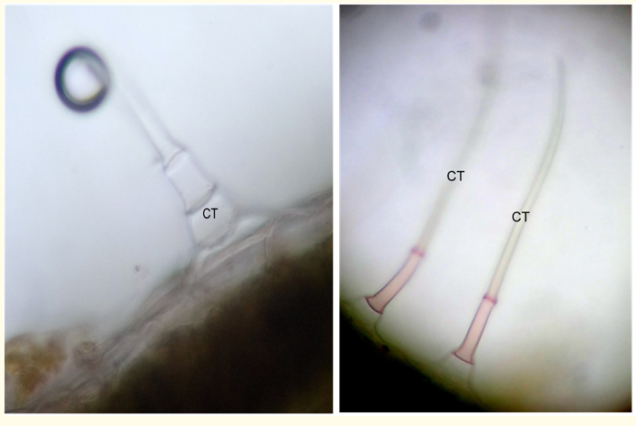

Figure 5: Epidermal cells of *Blumea mollis* leaf showed uniseriate multicellular covering trichomes.

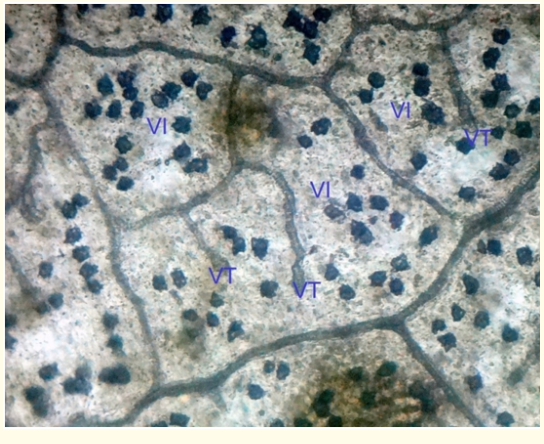

Figure 6: Vein Islet and Vein Termination of *Blumea mollis* Leaf. VT: Vein termination; VI: Vein Islet.

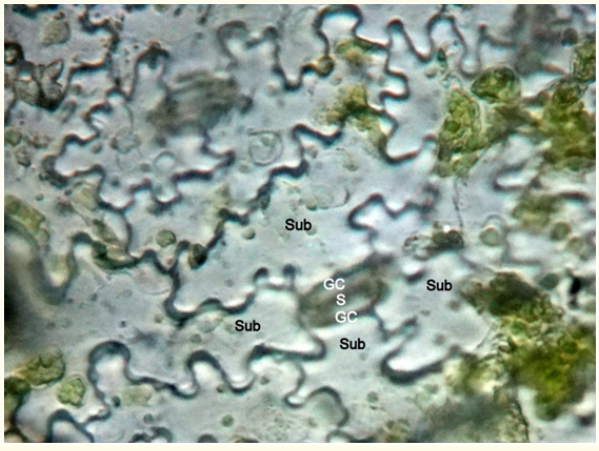

Figure 7: Surface view of epidermal cells of *Blumea mollis* leaf showed Anomocytic stomata. S: Stoma; G: Guard cells; Sub: Subsidiary cells.

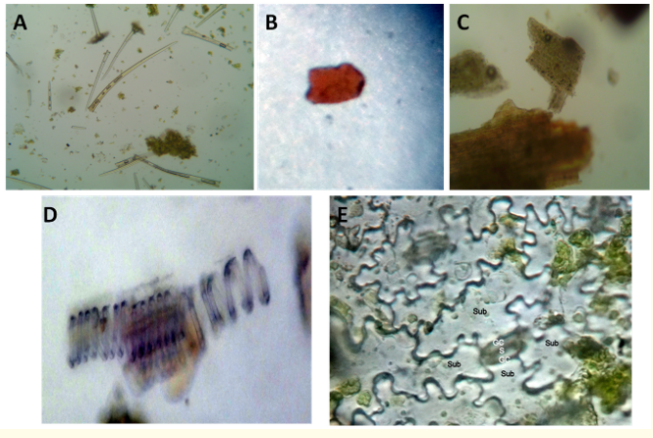

Figure 8: Powder Miscroscopy of *Blumea mollis* leaf powder. (a) Covering Trichomes (b) Pigment cell (c) Epidermal cells (d) Lignified xylm vessels (e) Anomocytic Stomata.

Powdered drug microscopy

Microscopic powder features revealed presence of covering trichomes, pigment cell, polygonal epidermal cells, spiral xylem vessels and anomocytic stomata.

Leaf constants

The leaf constants of leaf of *B. mollis* was tabulated in the table 2.

Physicochemical evaluation

The results of physicochemical parameters are summarized in Table 3.

Parameters	Average Value (per sq. Mm)
Stomatal number (Upper surface)	6.25 ± 1.25
Stomatal number (Lower surface)	11.23 ± 2.98
Stomatal Index (Upper surface)	14.12 ± 3.58
Stomatal Index (Lower surface)	25.33 ± 4.42
Vein Islet number	10.23 ± 1.54
Vein Termination number	26.85 ± 3.85

Table 2: Leaf constants of *B. mollis* Leaf.

Preliminary phytochemical screening

Various phytochemical analysis tests supported that the extracts contain alkaloids, carbohydrates, flavonoids, phenolic compounds, tannins and glycosides, recorded in Table 4.

Parameters	Values %w/w
Moisture content (Loss on drying)	7.89 ± 2.59
Total ash	6.41 ± 1.22
Acid insoluble ash	2.89 ± 0.85
Water soluble ash	3.56 ± 1.75
Petroleum ether soluble extractive value	0.89 ± 0.05
Chloroform soluble extractive value	2.58 ± 0.59
Ethyl acetate soluble extractive value	4.36 ± 1.26
Alcohol soluble extractive value	9.12 ± 2.56
Water soluble extractive value	12.57 ± 1.62

Table 3: Physicochemical parameters of *B. mollis* laef.

Fluorescence analysis

Fluorescence statement of *B. mollis* powdered leaf is tabulated in Table 5.

Phytoconstituents	Method	Pet. ether Extract	Chloroform Extract	Ethyl acetate Extract	Methanolic Extract	n-butanol Extract	Aqueous Extract
Flavonoids	Shinoda Test	-	-	+	+	-	+
	Zn+HCl test	-	-	+	+	-	+
	Lead acetate Test	-	-	+	+	-	+
Volatile oil	Stain test	+	-	-	+	-	+
Alkaloids	Wagner Test	-	+	-	+	-	+
	Hager's Test	-	+	-	+	-	+
Tannins & Phenols	$Fecl_3$ Test	-	-	-	+	+	+
	Potassium dichromate test	-	+	-	+	+	+
Saponins	Foam Test	-	-	-	+	+	+
Phytosterols	Libermann's test	+	+	-	+	-	-
Carbohydrates	Molish test	-	-	-	+	-	-
Acid compounds	Litmus test	-	-	-	-	-	-
Glycoside	Borntragers test	-	-	-	+	-	+
Amino acids	Ninhydrin test	-	-	-	+	-	+
Proteins	Biuret test	-	-	-	-	-	-
Fixed oils & fats	Spot test	+	-	-	-	-	-

Table 4: Preliminary Phytochemical analysis of leaf of *B. mollis* leaf. **"+"** Present and **"–"** Absent.

Solvent used	Visible light	UV light	
		At short (254nm)	At Long (366nm)
Water	Light brown	Light brown	Black
Sodium hydroxide	Light yellow	Yellow	Dark brown
Hydrochloric acid	Dark green	Light green	Black
Nitric acid	Brownishred	Light green	Dark black
Ferric chloride	Dark green	Light green	Black
Picric acid	Yellow	Yellow	Dark brown
Iodine	Light brown	Light green	Dark black

Table 5: Fluorescence statement of *B. mollis* powdered leaf is tabulated in Table 5.

Discussion

B. mollis is used extensively as an ancient traditional medicine for the treatment of wide range of diseases [6,16-18]. But there are no pharmacopeial standards for the correct identification and authentication of leaf of the plant. Thus, in the present study, the leaf part of B. mollis was analyzed for its pharmacognostic features. *B. mollis* is an an agreeably fragrant annual herb having leaves of green color, obovate, base attenuate, margin irregular serrate, tip acute, pubescent on both sides. The TS examination and powder microscopy showed the presence of covering trichomes, anomocytic stomata, pigment cells, epidermal cells and lignified xylem vessles. There are uniseriate hollow trichomes and anomocytic stomata were present on epidermis making these features as important diagnostic characters. Further leaf constant can also be used for identification of leaf and analyzing purity and adulterations.

Physicochemical assessments offer several essential variables like moisture content, ash values and extractive values for different solvents. The ash values established in our research might be useful in stabilizing specifications of purity and quality. The low total ash value show the low amount of inorganic salts. The acid insoluble ash was very low that support the truth that the very small quantity of the inorganic element is insoluble in acid and this is a diagnostic tool [19]. The moisture content of the fresh leaves was 8.72% displaying the leaf dried out very easily after plucking. The extractive values provides a concept regarding the type of the chemical constituents present in the plant and it is helpful for the evaluation of particular constituents soluble in this specific solvent utilized for the extraction and also the perseverance of exhausted materials. The water extractives were greater than those of others [20].

Therefore, these are the ideal choice as solvent for extraction of leaves of *B. mollis.* The fluorescence research demonstrated characteristic fluorescence under visible light and ultraviolet light (short and long wave length), which might be useful in the recognition of adulterants. Preliminary phytochemical testing was carried out to check on the existence of active constituents from the leaves. It reveals the presence of flavonoids, alkaloids, saponins, phytosterols, carbohydrates, glycosides, amino acids, and proteins.

Conclusion

The present pharmacognostical assay of *B. mollis* leaves which includes data on leaf constants, powder microscopy, fluorescence features and physical variables offers an essential analytical tool for recognition, authentication, and detection of adulterants of *B. mollis* leaves, along with development of quality variables from the species.

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