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Alekya K

P.G. Scholar, Department of
Dravyaguna, S.V. Ayurvedic
College, Tirupati, Andhra
Pradesh, India

Bulusu Sitaram

Professor, Department of
Dravyaguna, S.V. Ayurvedic
College, Tirupati, Andhra
Pradesh, India

Pavan Kumar S

Associate Professor, Department
of Dravyaguna, S.V. Ayurvedic
College, Tirupati, Andhra
Pradesh, India

Pharmacognostic evaluation of certain species of *Phyllanthus* used as botanical source of Bhumyamalaki/Tamalaki

Alekya K, Bulusu Sitaram and Pavan Kumar S

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Abstract

Tamalaki/Bhumyamalaki is a herbaceous plant which is very well known for its medicinal value in Ayurveda especially in liver disorders and other common ailments. Various species names have been attributed to Bhumyamalaki by different authors due to varied geographical locations where these plants grow and varied actions exhibited by them. This leads to confusion in selection of the species and accurate identification is essential as their use in practice is becoming more popular. Hence, it is imperative to assess the genetic diversity and phylogenetic relatedness among the *Phyllanthus* species. The aim of present work is to evaluate the morphological, organoleptic characters and anatomical sections of three *Phyllanthus* species taken for study and to establish Macroscopic and Microscopic analysis of drug for future conservative strategies.

Keywords: Tamalaki/bhumyamalaki, pharmacognosy, *Phyllanthus*

Introduction

The genus *Phyllanthus* L. was first described by Linnaeus in 1753 and consists of 833 species^[1] in the world which is one of the largest and most diversified genus in the family Euphorbiaceae and are chiefly distributed in moist humid tropics. In India, it is represented by 40 species^[2]. Before 1985, most of the botanical, pharmacological and phytochemical work of 'Bhumyamalaki' was carried out on *Phyllanthus niruri* L. of Hooker^[3]. Now, it is clarified that *Phyllanthus niruri* is an American species and not at all found in India^[4]. The herbs known as 'Bhumyamalaki' in Indian literature refer to a complex group of *Phyllanthus amarus* Schum. & Thonn., *Phyllanthus fraternus* Webster, *Phyllanthus debilis* Klein ex Wild and *Phyllanthus urinaria* L.^[5] Confusion exists in identification of these species mainly due to referring them all with a common vernacular name, their similarity in gross morphology, close proximity in growth habitat and lack of guidelines to check the authenticity and quality of the medicinal plant sold^[6].

Tamalaki/Bhumyamalaki in India has long been used for the treatment of Jaundice, gonorrhoea, frequent menstruation, dysentery and diabetes^[7, 8]. Various Studies include Antimicrobial activity of *Phyllanthus niruri*^[9] and *Phyllanthus amarus* focuses on its Anti-viral potential especially with Hepatitis B virus^[10, 11]. A single drug remedy (Tamalaki decoction) was mentioned in the treatment of Rajayakshma associated with six symptoms^[12]. Difference of opinion still exists among the various authors of literature in the selection of species. Various species of *Phyllanthus* are being sold in India under the trade name 'Bhuiamli'. During market surveillance of herbal drug, it was observed that almost all the commercial samples, either comprise of *Phyllanthus amarus* Schum & Thonn. or *Phyllanthus maderaspatensis* Linn. or mixture of *Phyllanthus amarus*, *Phyllanthus urinaria* and *Phyllanthus maderaspatensis*^[13]. So, in this context the pharmacognostic evaluation of all the three species has been carried out with the aim to establish their identification which includes both microscopic and macroscopic study including morphological and organoleptic analysis of each species respectively.

Materials and methods

Plant material: Fresh samples of whole plant of *Phyllanthus amarus*, *Phyllanthus maderaspatensis* and *Phyllanthus urinaria* taken for the study.

Collection of Drug: The study area was thoroughly surveyed for the presence of *Phyllanthus* species and Samples were collected from around the campus and nearby areas of Tirumala, Andhra Pradesh, India.

Corresponding Author:**Alekya K**

P.G. Scholar, Department of
Dravyaguna, S.V. Ayurvedic
College, Tirupati, Andhra
Pradesh, India

Authentication of the plant materials was done by the Department of Dravyaguna, S.V. Ayurvedic Medical College, Tirupati by examining the macroscopic and microscopic characteristics of the plant materials.

The species, Cross Sections (Transverse section) were obtained free hand with sectioning blades, then stained in safranin dye and mounted in glycerine. Semi-permanent slides, so prepared, were examined under electronic Digital microscope (1000x). The photography was made with the help of Camera fi2 app linked to mobile camera.

Plan of study

According to the W.H.O (1998), the macroscopic and microscopic description of a medicinal plant is the first step towards establishing its identity and purity.¹⁴

So, this study has been performed in following two divisions:

- Macroscopic study
- Microscopic study

Observations

Macroscopic Study

- *Phyllanthus amarus* Schum & Thonn.
Organoleptic characters: (Dried plant powder)
- Colour : Dark green
- Odour : Pungent
- Taste : Bitter
- Texture : Slightly smooth powder

Root: Hard tap root system which is nearly straight, gradually tapering, with a number of secondary and tertiary roots, external surface light brown, fracture - short.

Stem: Slender, Terete, light brown, 2-6 cm long branching profuse towards upper region bearing 5-10 pairs of leaves.

- *Phyllanthus maderaspatensis* Linn.
Organoleptic characters: (Dried plant powder)
- Colour : Light green
- Odour : Indistinct
- Taste : Slightly bitter
- Texture: Slightly smooth.

Root: The root is taproot with numerous secondary rootlets, light brown coloured with yellow colour fracture. Woody.

Stem: Glabrous, woody at base, 30 to 90 cm long, erect, branches terete below, striate and flattened, particularly at nodes towards apex.

- *Phyllanthus urinaria* Linn.
Organoleptic characters: (Dried plant powder)
- Colour : Dark green
- Odour : Pungent
- Taste : Slightly astringent
- Texture: Slightly rough, free flowing.

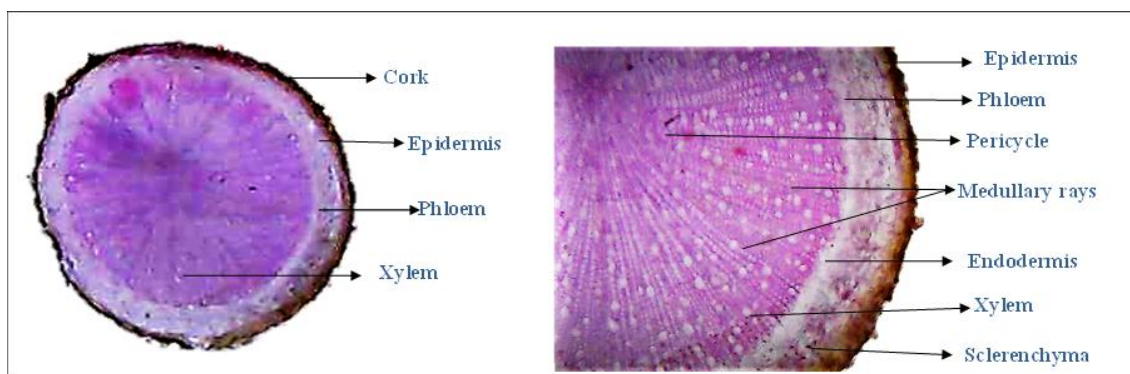
Root: Root is with cluster of small thin rootlets at the end, light reddish brown in colour.

Stem: Stem is Reddish-green, smooth, angled (pentagonal), woody at base, acutely ridged at nodes throughout, glabrous to pubescent.

Table 1: Showing the morphological variations among three species

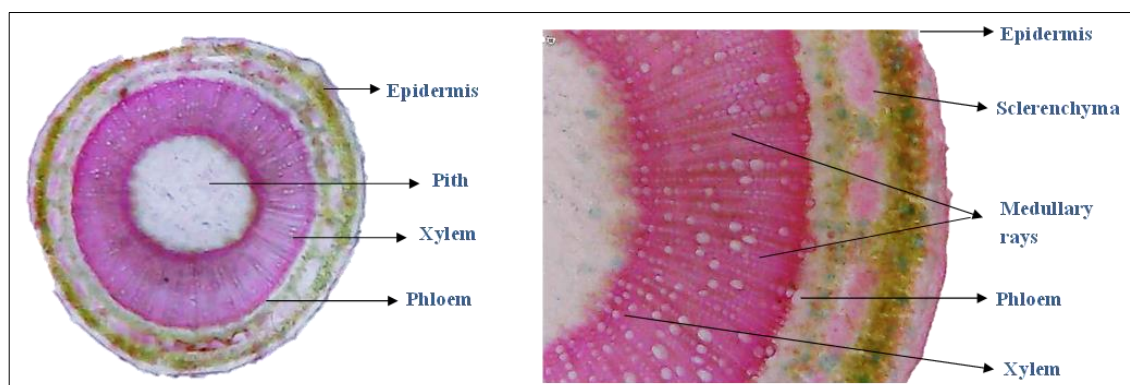
Morphological character	<i>Phyllanthus amarus</i>	<i>Phyllanthus maderaspatensis</i>	<i>Phyllanthus urinaria</i>
Habit	Branching annual herb	Erect/decumbent herb	Erect/procumbent herb
Habitat	Prefers rocky, calcareous sites in humid tropical regions.	Common weed of waste places, garden lawns and cultivated fields, in moist and shady places and prefers black cotton soil.	It grows on river or stream banks, agricultural fields, forest margins, gardens, wastelands.
Height	Upto 80 cm	Upto 1m	Upto 60 cm
Stem shape	Terete	Terete below, striate and flattened above	Terete below, flattened above
Stem colour and form	Greenish, smooth, rounded, glabrous and woody at base.	Greenish, slender, smooth and woody at base.	Reddish green, smooth, pentagonal, glabrous and woody at base.
Leaf shape	Oblong/elliptic oblong	Obovate / spatulate	Oblong/linear oblong
Leaf tip	Obtuse and shortly mucronate.	Rounded mucronate	Obtuse with distinct mucronate
Leaf base	Obtuse / rounded	Cuneate	Unequal
Flower	Unisexual and bisexual cymules	Unisexual and bisexual cymules	Unisexual cymules
Female disc	Flat, deeply five lobed	Six lobed	Six lobed
Capsule	Globose and smooth	Depressed globose and smooth	Depressed globose and prominently echinate.
Seed	Trigonous with 6-7 parallel longitudinal ribs on back	Trigonous, muriculate in fine lines.	Triquetrous with prominent transverse ridges and crossbars.

Microscopic Study



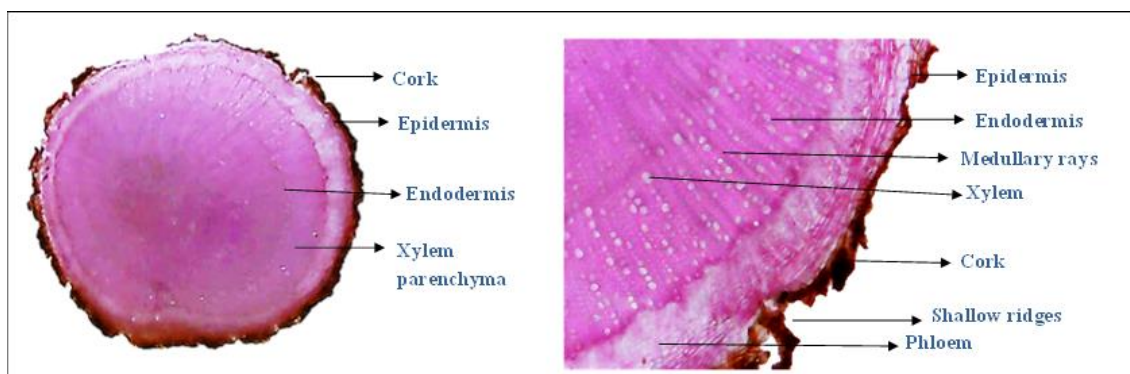
- **Outline** is circular with fissures.
- **Epidermis** is broken with 2 – 4 layers of tangentially elongated and lignified cork cells, which are filled with brown content and oil globules.
- Some of the isolated 2 – 3 celled pericyclic fibres were observed in cortical region.
- **Cortex** ends with single layer of parenchymatous endodermis.
- **Phloem:** The Phloem is about 4 – 6 cells thick.
- **Xylem:** It includes either Solitary or multiples of thin walled circular vessels.

Image 1: Showing the Transverse section of *Phyllanthus amarus* - Root



- **Outline** is smooth and circular covered by thick cuticle.
- **Epidermis** with Epidermal cells which are rectangular and thick walled.
- **Hypodermis:** It is 2 -3 layers thick.
- **Cortex:** Cortical zone was narrow, inner border of the cortex was seen unevenly thick with discontinuous cylinder of cortical fibres.
- **Sclerenchyma:** It is 2 – 3 layers thick
- Larger region of the section is occupied by **pith**; parenchymatous cells are filled with some starch grains and rosette crystals of Calcium oxalate.

Image 2: Showing the Transverse section of *Phyllanthus amarus* - Stem



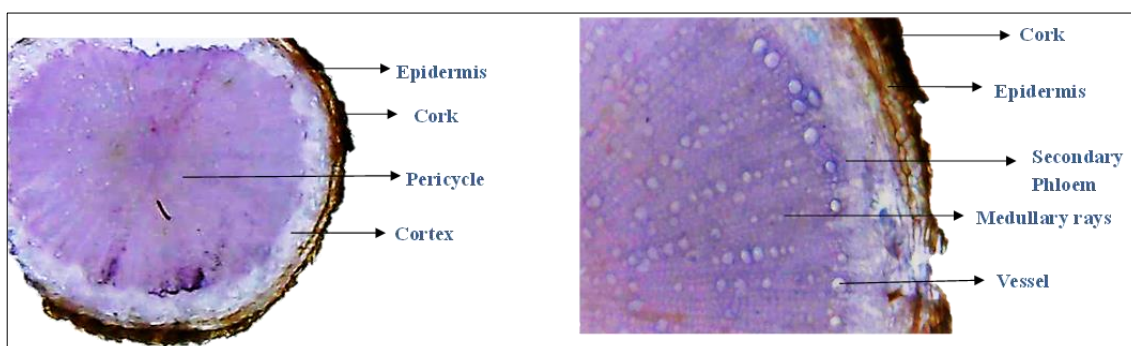
- **Outline** is irregularly circular with shallow ridges.
- **Epidermis** is filled with lignified cork cells with some cells filled with brown content.
- Below epidermis is composed of tangentially elongated 2 – 3 layers of lignified cork cells, some of the cells filled with brown content.
- **Cortex** region is found to be moderately loosely arranged and made up of 4 – 7 layers of simple parenchymatous cells.
- **Medullary** rays are uni-serrate and started from central region and extended up to inner layers of the pericyclic region.

Image 3: Showing the Transverse section of *Phyllanthus maderaspatensis* – Root



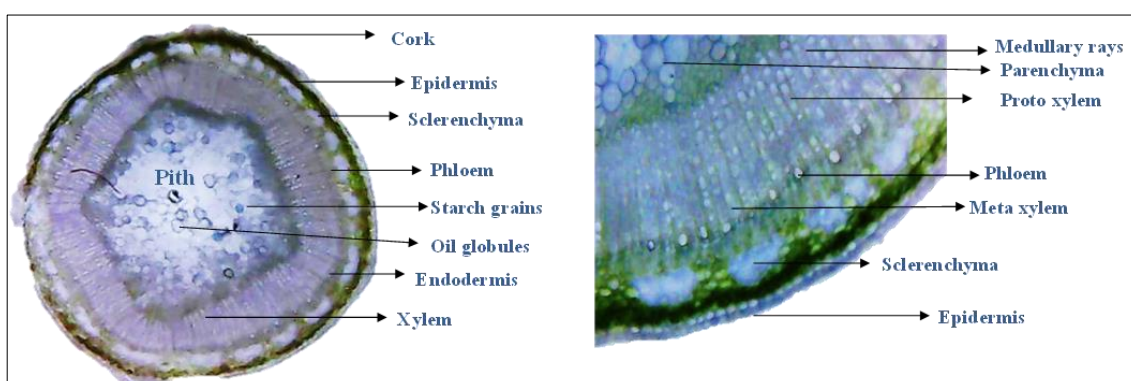
- **Outline** is somewhat Pentagonal in shape.
- **Epidermis** is single layered with compactly arranged cells.
- **Hypodermis** is made up of single layered but contains 2-4 layered sclerenchyma cells.
- **Cortex** is made up of 3 – 5 layers of simple parenchymatous cells.
- **Xylem** is separated by uni-serrate medullary rays. **Phloem** is well developed situated above the xylem with sieve elements and fibres.
- Parenchymatous pith cells are compactly arranged in pith region. **Pith** cells are filled with oil globules, simple and compound starch grains and sphenoid crystals of Calcium oxalate.

Image 4: Showing the Transverse section of *Phyllanthus maderaspatensis* – Stem



- **Cork:** Composed of 2-4 layers of tangentially elongated lignified cork cells, some of the cells filled with brown content and oil globules.
- **Cortex** region is found to be 8 – 10 layered with loosely arranged simple parenchymatous cells which are sacredly isolated with simple starch grains and tannin content.
- **Endodermis:** The innermost layer of the cortex which is endodermis is one layer thick.
- The **Vascular bundles** which are radially arranged contained both xylem and phloem with xylem being more prominent.

Image 5: Showing the Transverse section of *Phyllanthus urinaria* – Root



- Outline is circular with angular projections/ ridges at different places.
- Epidermis is single layered and covered with thick cuticle.
- Collenchyma ranges from 2 – 5 layers. The shape of collenchyma is rectangular.
- Cortex forms narrow zone with polygonal shaped cells of parenchyma.
- Endodermis: It lies immediately outside the pericyclic sclerenchyma which is 2 – 3 layers thick.
- The phloem ranged from 3 – 5 layers while the no. of elongated cells in xylem vessels ranged from 2 – 5 cells.
- Pith, parenchymatous cells are compactly arranged in pith region. Pith cells are filled with oil globules, rarely simple starch grains and some rosette crystals of Calcium oxalate.

Image 6: Showing the Transverse section of *Phyllanthus urinaria* - Stem

Results and discussion

The morphological, macroscopic and microscopic analysis of three species reveals the following facts

- Morphologically variation in the height of the herb, root system was noticed. Stem colour and form varies among species (*Phyllanthus urinaria* being reddish brown in comparison with other two species which are greenish). Stem Fracture is Short (*Phyllanthus amarus*), Yellow (*Phyllanthus maderaspatensis*) and reddish brown (*Phyllanthus urinaria*).
- *P. amarus* can be easily distinguished from its allies by its unique characteristics of terete stem and five tepals.
- Leaf shape is specific in case of *P. maderaspatensis* which is Spatulate with rounded mucronate tip whereas it is oblong or linear oblong in case of *P. amarus* and *P. urinaria* respectively.
- Microscopical studies indicated the presence of highly lignified with brown content in Root sections of *Phyllanthus amarus* and *Phyllanthus urinaria* than *Phyllanthus maderaspatensis* root.
- Stem section of *P. urinaria* and *P. maderaspatensis* possess ridges and furrows which are passed over by a single layer of epidermis. The presence of ridges and furrows were as result of the angular nature of the stem circumference. This feature is absent in *P. amarus* due to rounded stem circumference.
- Narrow cortical zone with unevenly thick cortical fibres was found in *P. amarus* (stem), whereas cortical cells are polygonal shaped in *P. urinaria* (stem) and with simple parenchymatous cells in case of *P. maderaspatensis* (stem).

The word Bhumyamalaki thus reasonable to be considered as a generic name includes different species of *Phyllanthus* which are having their specific action. Among various species available, three species which are most abundant and commonly sold in commercial markets were taken for the study. It is observed that *P. amarus* and *P. maderaspatensis* have five and six prominent tepals respectively, but *P. urinaria* got very minute six tepals. Capsule of *P. urinaria* is echinate, whereas rest two is smooth. Hard tap root, woody tap root, highly fibrous root system are seen in *P. amarus*, *P. maderaspatensis* and *P. urinaria* respectively. The Pharmacognostic study done on three different species of *Phyllanthus* and the data generated is helpful in determining the Quality and Purity of the Drug.

Conclusion

In Ayurveda and folklore practices *Phyllanthus amarus* as Bhumyamalaki, is used in various ailments especially in liver troubles, but other two species viz. *Phyllanthus maderaspatensis* and *Phyllanthus urinaria* are all mixed up and being sold in herbal drug markets of the country by the same vernacular. Considering the medicinal properties of the *Phyllanthus* herbs and their ethno pharmacological value, these species have been assessed for their pharmacognostic study. The macroscopic and microscopic diagnostic characters drawn from the present study by using simple techniques may eventually help to authenticate genuine samples of *Phyllanthus* which are being mixed up and sold in the market as Bhumyamalaki/ Tamalaki.

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