



## Anatomy of *Phyllanthus niruri* L. (Euphorbiaceae)

<sup>1</sup> Rubaiyat Sharmin Sultana\*, <sup>2</sup>Md. Mahabubur Rahaman

<sup>1</sup> Department of Botany, University of Rajshahi, Bangladesh

<sup>2</sup> Department of Crop Botany, EXIM Bank Agricultural University Bangladesh, Chapainawabganj, Bangladesh

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### ABSTRACT

The present study contracts with anatomical characterization of *Phyllanthus niruri* L., belongs to Euphorbiaceae family. Stem, leaf and root were the specimens for the anatomical study to observe treats in transverse section after staining with safranin. The anatomical studies revealed that the transverse section of stem showed the presence of one-layered epidermis covered by a thick cuticle. Hypodermis ranged 2-3 layers was present beneath the epidermis. Cortex was parenchymatous with profusely intercellular space. Cambium was present between xylem and phloem. Xylem was radially arranged and uniserrated to multiserrated. Xylem fibers was thick walled and lignified. The phloem was narrow. In transverse section of leaf, the epidermis layer was made up parenchymatous cells and the epidermal layer was covered with thick cuticle. Mesophyll cells (palisade and spongy parenchyma cells) were filled with chlorophylls. The large intercellular spaces occurred in the spongy parenchyma. *Phyllanthus niruri* L. showed paracytic stomata. The transverse section of root, it was encircled by cork cells. The cortical zone was narrow and the cortical cells intacted. The cortical regions arranged parenchymatous cells. The xylem was lignified and phloem was narrow and medullary rays were present.

**Key words:** Anatomy, Euphorbiaceae, Leaf, *Phyllanthus niruri*, Root, Stem

### CORRESPONDENCE

\* [sultanaru@yahoo.com](mailto:sultanaru@yahoo.com)

Department of Botany, University of Rajshahi, Rajshahi-6205, Bangladesh

### 1. INTRODUCTION

*Phyllanthus niruri* L. is belonging to the family Euphorbiaceae, is originated in India. It is widely spread throughout the tropical and subtropical countries. It is widely found in both cultivated and inferior land of Bangladesh. It is an annual, herb; height varies between 30-60 cm; main stem simple or branched, terrete smooth or scabridulous in younger parts (Morton, 1981).

Although considered a problematic weed for farmers, it is a valuable medicinal plant for herbalists (Oudhia & Tripathi, 2002) and holds a reputed position in both Ayurvedic and Unani systems of medicine. The use of *P.*

*niruri* is gaining momentum because of its novel antiviral activity against hepatitis B virus and for several other biological activities such as kidney stones, gallbladder stones, cold, flu, tuberculosis, liver diseases and disorders including hepatitis, jaundice and liver cancer (Unander et al., 1993). It also acts against liver cell toxicity and improves the immune system of patients and has been found effective against hepatitis A. *P. niruri* L. is often used in the traditional system of medicine for a variety of ailments including dropsy, diabetes, jaundice, asthma and bronchial infection (Foo & Wong, 1992). In the Ayurvedic system of medication, it is used in problems of stomach, genitourinary system, liver, kidney and spleen. It is bitter, astringent, stomachic, diuretic, febrifuge and antiseptic. The whole

plant is used in gonorrhoea, menorrhagia and other genital affections. It is useful in gastropathy, diarrhoea, dysentery, intermittent fevers, ophthalmopathy, scabies, ulcers and wounds. It is also used as a good tonic. In Suriname (Northeastern part of South America), this plant is always sold as fresh and dry plant material in the herb markets. Decoctions are used in herbal baths and after labor, cramps, asthma, uterus complaints and to treat stomachache (May et al., 1982).

Plant anatomy is the study for the internal structure of plant. It is now frequently investigated at the cellular level, and often involves the sections of tissue and microscopy. Anatomical study has much significant in different sectors of investigation. Studies on anatomy of plant can explain where, what, when and how chemical component are produced. The anatomical studies can be clarified the quality of wood properties. It can apply mainly when there is no reproductive organ in the investigated sample (Metcalfe & Chalk, 1950).

Study on anatomy of *Phyllanthus niruri* L. was very limited. So far one anatomical study has reported on the stem of this plant (Tuhin & Limon, 2019) with other 7 species namely, *Phyllanthus amarus*, *P. reticulatus*, *P. acidus*, *P. emblica*, *P. debilis*, *P. urinaria* and *P. vibratus*. The report on leaf and stem anatomy of *phyllanthus* spp. was published by Lee et al. (2005). They reported on *P. emblica* and *P. myrtifolius* and possessed the common features like calcium oxalate crystals distributed in leaf, well represented vascular bundles in petiole, leaf, stem and root. Their distinct features like vasculature in petiole, sclerenchyma and lactifers were observed. Leaf lamina showed loosely arranged spongy mesophyll and palisade mesophyll.

In other species of *Phyllanthus* genus, *P. amarus* and *P. odontadenius* were reported on the stem, leaf and root anatomical traits by Awomukwu et al. (2015). They observed that the epidermis of the leaf was one layer thick. Large intercellular spaces occurred in the spongy parenchyma of *P. odontadenius* while *P. amarus* had small intercellular spaces. The cross-section of *P. odontadenius* showed the presence of ridges and furrows while *P. amarus* possessed none. The roots of those two species were composed of an outer piliferous layer that was one layer thick. The vascular bundles contained both xylem and phloem with xylem being more prominent. Anatomy of stem, root, node and stomata were observed using compound microscope in *Phyllanthus amarus* Schum. & Thonn. and *P. urinaria* L. (James et al., 2018). The leaf anatomy of ten plant species in the family Euphorbiaceae from three sites with different pollution levels in Southwestern Nigeria were carried out (Ekpemerechi et al., 2017). The leaf, stem and root anatomy described in *Phyllanthus amarus* Schum. and Thonn. (Siva & Subramaniam, 2016). Stem and leaf anatomy of *Euphorbia hirta* belongs to the family Euphorbiaceae has been carried out (Sultana, 2017).

An anatomical report on stem was published in *P. niruri* L. for one time that was very poor investigation. Till now leaf, root or other parts of the investigated plant were not

studied anatomically. However, it is spelled out that more anatomical research is needed to detail describe anatomical feature of *P. niruri* L. In the present study, a detail anatomical feature of stem, leaf and root of *P. niruri* L. was studied.

## 2. MATERIALS AND METHODS

*Phyllanthus niruri* L. is naturally grown plant, found in inferior lands. The plants were collected from near Stadium, Botanical Garden, Mulberry Garden, inside the 4<sup>th</sup> Science Building, the back side of 3<sup>rd</sup> Science and 2<sup>nd</sup> Science Building of Rajshahi University Campus, Rajshahi, Bangladesh. As plant materials, disease free healthy plants were selected at the research sites and then whole plants were collected. Stems, leaves and root were used as plant materials for this research (Fig. 1). Free hand transverse sections were prepared from stems, leaves and roots. Stem and root sections prepared by cutting into thin sections with stainless steel razor blade followed by leaf sections with assist of potato block. The sections were then placed into water in a petri dish. Thin and uniformed sections were separated carefully with small headed hair brush.



Fig. 1 A plant of *Phyllanthus niruri* L. grown in the inferior land of Botanical Garden, Rajshahi Unuversity, Bangladesh.

Well-expanded leaves were used for stomatal observation. Epidermal peels of lower surface were made and placing on

a clean glass slide, with the surfaces to be studied facing down. The specimens were irrigated with water holding and it downwards from one end and then the epidermis above the desired surface was scraped-off carefully with a sharp razor blade. The loose cells were washed away from the epidermal peels with the aid of hair brush and the epidermal peels were stained.

Thin and uniform fresh sections of stem, leaf, root and epidermal peels were stained with freshly prepared 1% (w/v) aqueous solution of Safranin for 4-5 minutes, rinsed carefully by water to remove excess stain and mounted in 10% glycerol. The sections of stem, leaf and root as well as epidermal peels were observed under light microscope fitted with digital camera. Keen and minute observation was made for each of the 15 slides made. Microphotographs were taken with digital camera. Diagram of cells ( $\mu\text{m}$ ) and stomata measured in 50 samples.

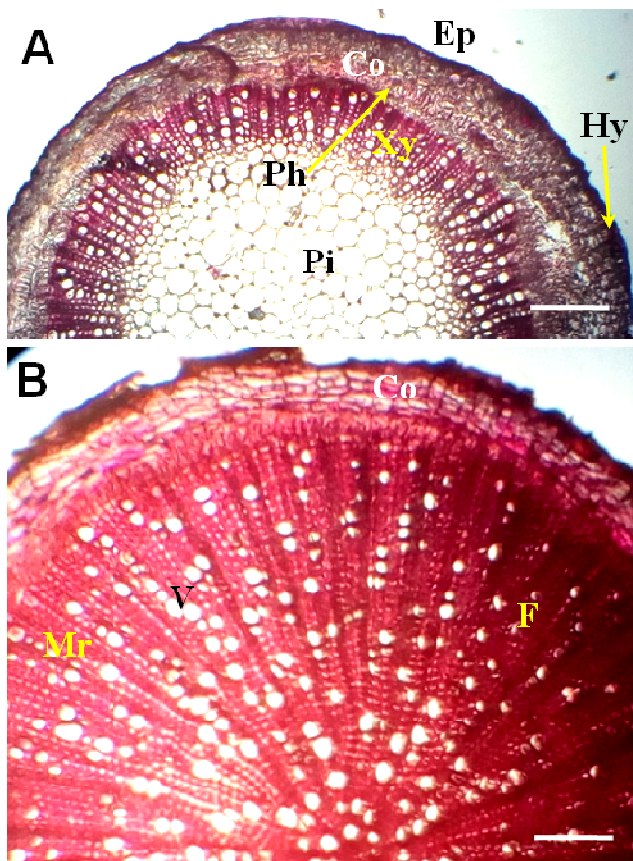


Fig. 1 Stem and root anatomy of *Phyllanthus niruri*. A) T.S of Stem, B) T.S of Root. Ep: Epidermis, Hy: Hypodermis, Co: Cortex, Ph: Phloem, Pi: Pith, V: Vessel, F: Fiber, Mr: Medullary ray. Bar: 100  $\mu\text{m}$ .

### 3. RESULTS AND DISCUSSION

#### 3.1. Stem Anatomy

The stem of *P. niruri* was circular in cut view. One layer epidermis at the inner side of cuticular layer was composed of oblong and flattened formed cells (Fig.2A). The diameter

of epidermis cell was measured at  $28.5 \pm 1.4 \mu\text{m}$  (Fig. 1). The hypodermis laid externally, ranged 2-3 layers.

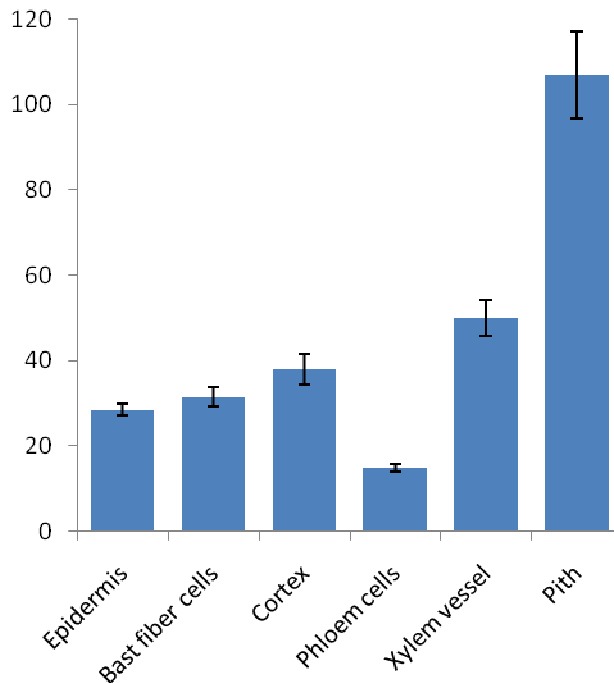


Fig. 2 Different cell diameter ( $\mu\text{m}$ ) in stem of *Phyllanthus niruri* L.

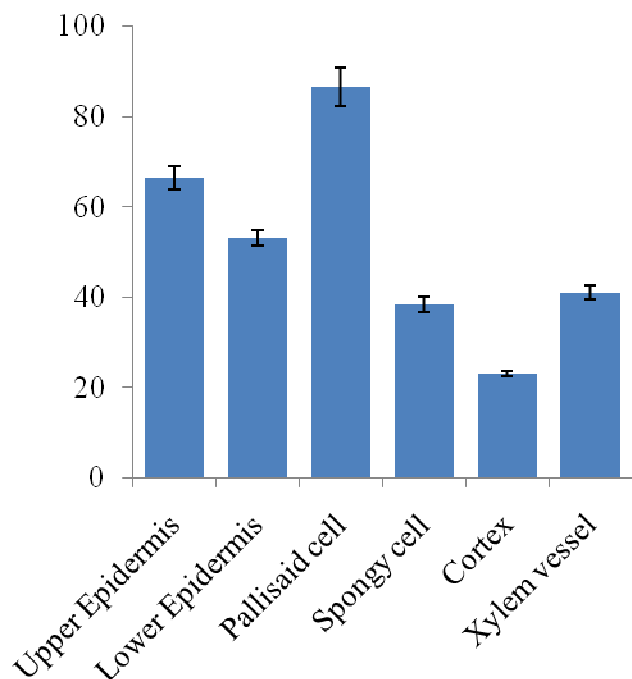


Fig. 3. Different cell diameter ( $\mu\text{m}$ ) in leaf of *Phyllanthus niruri* L.

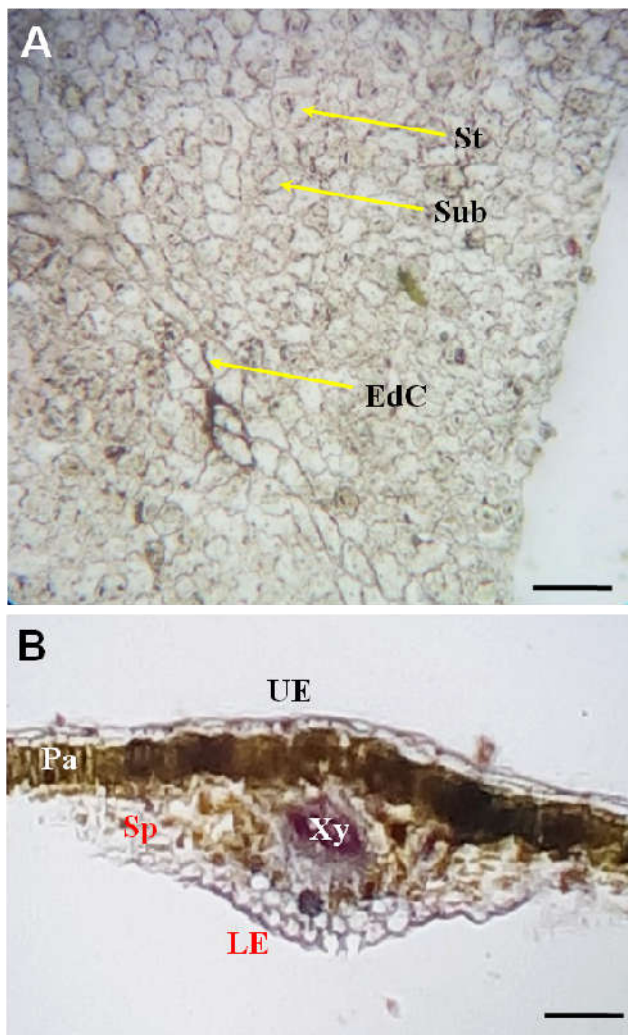


Fig. 4 Leaf anatomy of *Phyllanthus niruri*. A) Peel of lower surface of leaf, B) T.S of leaf. St: Stomata, Sub: Subsidiary cell, EdC: Epidermal cell, UE: Upper epidermis, Pa: Palisade, Sp: Spongy, Xy: Xylem, LE: Lower epidermis. Bar: 100  $\mu\text{m}$

Generally, the cortex ( $38.0 \pm 3.7 \mu\text{m}$ ) was placed in the inner side of the hypodermis (Fig. 1), was composed of 6-10 layers of oblong, round, ellipse parenchyma and having numerous intercellular spaces (Fig. 2A). The reddish ring near outer edge of the cortex consisted of bast fiber cells, measured at  $31.5 \pm 2.3 \mu\text{m}$  (Fig. 1). The bast fiber cells showed thick walls. The phloem cells were in irregular shape and measured in diameter at  $15.0 \pm 0.9 \mu\text{m}$  (Fig. 1). Cambium was composed of flattened square shape cells. The xylem vessel was largest cell, measured at  $50.0 \pm 4.3 \mu\text{m}$  (Fig. 1). Xylem fiber, wood parenchyma and medullary ray were also present. The pith cells closed to the center, were larger and round-like in shape with rather thin walls and measured at  $107.0 \pm 10.2 \mu\text{m}$  (Fig. 1). Intercellular spaces were present abundantly in pith region (Fig. 2A). Tuhin & Limon (2019) had reported for the first time on the stem anatomy of *P. niruri*, but it was not detail. The phenomenon

of the present study is agreed with the report of Awomukwu et al. (2015). The report on stem anatomy of *Phyllanthus* genus was published by Lee et al. (2005) in *P. emblica* and *P. myrtifolius*. In other species of Euphorbiaceae family, *Euphorbia hirta* L. and *Euphorbia thymifolia* were reported with the stem anatomical traits by Sereena & Shahida (2015).

### 3.2. Leaf Anatomy

The transverse section of leaf showed epidermis (both upper and lower surfaces) cover the section both in lamina and midrib portion. The upper epidermis was covered with cuticle especially in lamina portion (Fig. 2C). The lamina portion exposed that palisade cells, covers half to one third portion of lamina. The palisade parenchyma was one layer thick that measured at  $86.5 \pm 4.3 \mu\text{m}$  in diameter. Large intercellular spaces occurred in the spongy parenchyma at  $38.5 \pm 1.7 \mu\text{m}$  (Fig. 2C). The distribution of the collenchyma cells within the midrib of the studied species. Collenchyma cells were present in the adaxial and abaxial portions of the leaf. Mesophyll cells (palisade and spongy parenchyma cells) were filled with chlorophylls and at places are interrupted with vascular cells. The vascular bundle strand was single, showed in the transverse section at the mid rib of leaf. The vascular bundle was consisting of the xylem and phloem (Fig. 2C). The result showed that the vascular bundle was surrounded by cortex at  $23.0 \pm 0.6 \mu\text{m}$  in diameter (Fig. 2C). The lower surface of leaf preparation showed the paracytic type of stomata (Fig. 2B). In 100  $\mu\text{m}$  leaf area,  $18 \pm 3.5$  stomata were present.

The findings of leaf anatomy in the study are agreed with the outcome of Awomukwu et al. (2015). According to Elumalai et al. (2014), the leaf section of *Ricinus communis* L. (Euphorbiaceae) showed epidermis multilayered (two to three layer), prominent mesophyll tissue divided into equally palisade and spongy parenchyma region, was contrary to the present study. Villa (2017) reported that the transverse section of leaf of *Euphorbia hirta* L. epidermis was uniseriate with regularly thin walled cells. The presence of paracytic stomata in the tribe Euphorbieae were reported by Metcalfe & Chalk (1950). Raju & Rao (1977) reported presence of paracytic stomata as most common. Some studies recorded anisocytic, anomocytic and diacytic stomata in 50 species belonging to 17 different tribes of the Euphorbiaceae. In their opinions the paracytic type forms the basic stomatal type for the family Euphorbiaceae because of common occurrence in majority of tribes studied. The findings in the present report are in similar to the report of Raju & Rao (1977). Other species of Euphorbiaceae, *Euphorbia hirta* possessed amphistomatic leaves bearing anomocytic and anisocytic stomatal complexes, as well as paracytic stomatal complexes seen on adaxial side. Lee et al. (2005) was published leaf anatomy of *Phyllanthus* genus. They reported that *P. emblica*, *P. embergeri*, *P. hookeri*, *P. urinaria* L. subsp. *nudicarpus*, *P. urinaria* L., *P. ussuriensis* and *P. virgatus* had non-glandular hairs.

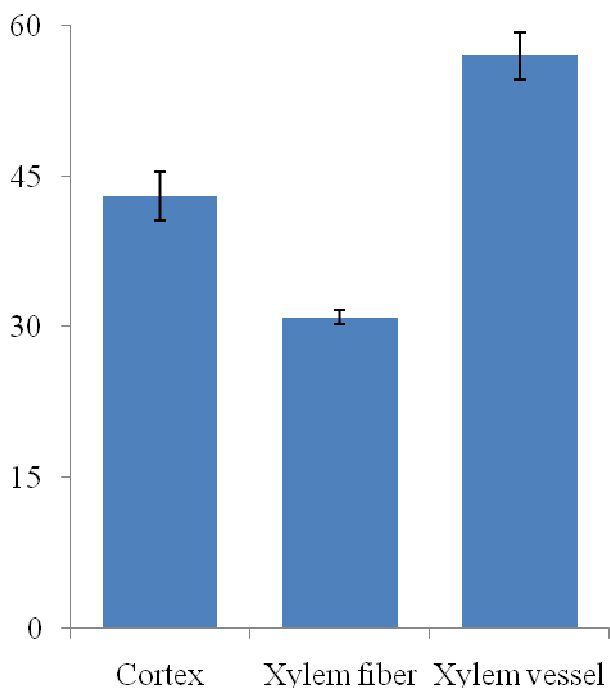


Fig. 5 Different cell diameter ( $\mu\text{m}$ ) in root of *Phyllanthus niruri* L.

### 3.3. Root Anatomy

The Transverse section of the root was composed of the outer epidermis which was made of a few layers of irregular. The root was rough with irregular fissures on the surface (Fig. 2D). The cortical zone was narrow and the cortical cells intact at certain regions and collapsed in other regions (Fig. 2D). The cortical region comprising of randomly arranged parenchymatous cells ( $43.0 \pm 2.4 \mu\text{m}$ ), was lying below the epidermal. The innermost layer of the cortex which was the endodermis, was one layer thick and prominent.

The vascular bundles contained both xylem and phloem where xylem being more prominent. The vascular bundle was collateral in nature with phloem arranged outwards and the xylem situated inwards occupying the whole area of the section (Fig. 2D). The secondary phloem consisted of small groups of sieve elements which were intervened by the medullary rays. The phloem elements were wide and thick walled. The central cylindrical secondary xylem was lignified and consisted of xylem elements such as parenchyma, vessels and fiber. The vessels in the central part of the root were narrow (Fig. 2D). The vessels were solitary, circular and thick walled. The diameter of vessel measured at  $46.0 \pm 0.3 \mu\text{m}$ . Medullary rays were found to be penetrating throughout the whole length of the xylem. The central pith was found to be absent (Fig. 2D). The findings in the present report are in similar more or less to the report of Awomukwu et al. (2015) in *Phyllanthus* genus, *P. amarus* and *P. odontadenius*.

## 4. CONCLUSION

In the present study, stem, leaf and root anatomy of *Phyllanthus niruri* was studied. Previously one study has been carried out on stem anatomy of *Phyllanthus niruri*. Thus leaf and root anatomy has first study for this plant. This study will help for the higher research on *Phyllanthus niruri*.

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