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# **Morphological and Anatomical Characters of *Piper betle* L. from Madaya Township, Mandalay Region**

Win Win Khaing<sup>1</sup> & Thein Kywe<sup>2</sup>

## **Abstract**

In this research, the species of *Piper betle* L. belonging to family Piperaceae were studied. The specimens were collected from Madaya Township, Mandalay Region from January to October 2015. The morphological and anatomical characters of leaves, stems and roots were studied, identified and photomicrographs were recorded. Cyclocytic type of stomata was observed in adaxial surface of leaves. In anatomical characters of stems, anomalous structure of vascular arrangement and central mucilage canal were observed. The vascular strand of roots showed triarch.

Key word: *Piper betle* L., Morphology, Anatomy

## **Introduction**

*Piper betle* L. was native to Malay Archipelago (Chibber 1912). It was commonly grown throughout the Eastern Tropics but not known as a wild plant (Dassanayake 1985). It is extensively cultivated in India, Bangladeshi, Pakistan, Malaysia, Indonesia, Vietnam, Laos, Thailand, Myanmar and Singapore, etc. for its leaves which are used as masticatory (Ravindran 2000 and Kumar *et al.* 2010). As a masticatory, it is credited with many properties: it is aromatic, digestive, stimulant and carminative (Anonymous 1969 and Nianhe *et al.* 1999).

*Piper betle* L. is a perennial creeper belonging to the family Piperaceae, cultivated in Myanmar for its leaves, used mainly for chewing and medicinal uses. It is a perennial dioecious, semiwoody climber. Stems were strongly swollen at the nodes, and papillose when young. Leaves were alternate, simple and yellowish green to bright green in colour.

Hooker (1885) stated that *P. betle* L. have large coriaceous leaves, blade ovate-oblong or rounded ovate-cordate, base often unequal, 5 to 7-

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nerved, petiole 1.0-3.5cm long, male spikes 7.0-15.0cm long; female spike long peduncled, pendulous.

*Piper betle* L. is known as 'kun' in Myanmar (Hundley & Chit Ko Ko 1987, Kress *et al.* 2003 and Arambewela & Alagiyawanna 2006). It is generally known as 'pann' in the Indian subcontinent and different names in the Asiatic region (Kumar *et al.* 2010). In Thailand, *P. betle* L. is commonly known by its Thai name of “Phlu” (Chahal *et al.* 2011).

The chief constituent of the leaves of this plant is a volatile oil known as betel oil. The volatile oil is bright yellow to dark brown liquid possessing a clove like flavor and consists of terpenes and phenols. Leaves of *Piper betle* L. possess several bioactivities and are used in traditional medicinal systems. Betel leaf phytochemicals are useful in prevention of gastric cancer (Chahal *et al.* 2011 and Baliga *et al.* 2011).

In India, Myanmar, Nepal, Srilanka and other parts of South Asia, the leaves of *Piper betle* L. are chewed together in a wrapped package along with the areca nut (which, by association is often called the betel nut) and mineral slaked lime (calcium hydroxide) (Bhalerao *et al.* 2013).

Analysis of a sample of flesh leaves contain moisture, 85.4; protein, 3.1; fat, 0.8; carbohydrates, 6.1; fibre, 2.3; and mineral matter, 2.3%; calcium, 230 mg; phosphorus, 40mg; iron, 7mg; ionisable iron, 3.5 mg; carotene, 9 600 I.U; thiamine, 70 µg; riboflavin, 30 µg; nicotinic acid, 0.7 µg; and vitamin C, 5 mg/100 g; iodine, 3.4 µg/100 g; vitamin B (Anonymous 1969).

Betel leaves contained about 2.0%, 1.7% and 0.8% essential oil respectively, on dry weight basis. In fact, these constituents are the sources of the medicinal, aromatic, stimulant, tonic and various other useful properties found in the leaves. Such useful properties of the oil indicate a promising industrial future for it as a raw material for manufacturing skin emollients, tooth-pastes, tooth-powders, perfumes, room fresheners, deodorants, soaps, face creams, antiseptic creams and lotions, cold drinks, chocolates, incense sticks, carminative mixtures, digestive agents, tonics, medicines etc (Guha 2006).

*Piper betle* L. is a very important and profitable crop of West Bengal. Betel leaves are mainly used for mastication. Betel leaves have some medicinal values. Medicinal properties of betel leaves were shown

to have effect of indigestion, stomach ache and diarrhea, wounds eczema, lice infestation sore of throat, respiratory disorder, scanty or obstructed urination (Sengupta 2014).

In anatomical characters, upper epidermis of leaf of *Piper betel* L. is many-layered. The usual number of layers is three. The cells are isodiametric in surface view, lateral walls are quite straight. Stomata are absent. The hypodermic is diversified by the presence of secretory cells. These are spherical in outline. In lower epidermis, the hypodermal layer is only one layered, lateral walls are straight or slightly curved and stomata are present. Midribs have three large and a variable number (0 to 4) of smaller vascular bundles, all arranged in a crescent. A large mucilage canal present above the central bundle. The upper border of petiole is either flat or slightly notched, or deeply grooved along the middle. The centre of petiole in transverse section is occupied by a single large mucilage canal. Vascular bundles are crescent shaped. They are graded in size. The phloem is characterized by the presence of the secretory cells. The transverse section of stems show a central canal and about a dozen peripheral canals situated in the ground tissue just outside the medullary bundles. Vascular bundles are collateral type. The transverse section of root can be seen six primary medullary rays. The intervening xylem plates have bifurcated once or twice. The centre of the section is occupied by ligneous elements. The medullary rays are about twelve cells broad (Chibber 1912).

Metcalf and Chalk (1950) mentioned that stem and petiole of *Piper betle* L. show spherical secretory cells with suberized walls, containing mucilage. Several species of *Piper* (e.g. *P. betle* L. and *P. nigrum* L.) are distinguished by mucilage canals in petiole and axis. Hairs are mostly uniseriate. Small, spherical, shortly stalked glands, and sometimes deciduous peral glands, also occur. Stomata, nearly always confined to the lower surface, are often surrounded by a rosette of numerous epidermal cells, whilst others are cruciferous. Petiole of *Piper betle* L., in transverse section, vascular system consisting of a crescent or almost closed circle of separate bundles, the centre of the petiole being occupied by a large mucilage canal. Crystals are mostly in the form of small needles in the petiolar ground-tissue. Pith includes scattered stone cells. Secretory cells present in the soft tissues. A peripheral ring of vascular bundles are present and bounded internally and separated from

the pith by a sinuous zone of sclerenchyma. Outer margin of the phloem strands are also accompanied by sclerenchyma. Remaining vascular strands more or less scattered and embedded in the thin-walled pith. A central, and up to about 12 peripheral, mucilage canals are present. Root of *Piper betle* L. shows 6 xylem plates, bifurcated once or twice and radiating from the centre, the plates being separated by broad, sometimes as much as 12-seriate, rays. Cork and cortex well developed. Mucilage canal absents in the root.

Morphological and medicinal uses of betel leaves, and planting techniques and disease symptoms of betel vine were studied by other researchers. However, the anatomical study of *Piper betle* L. is scanty. It is for this reasons, it is needed to study morphological and anatomical characters of *Piper betle* L.

The aims and objectives of this research are to study and describe their morphological and anatomical characters of leaves, stems and roots of *Piper betle* L. from Madaya Township, Mandalay Region, to verify the morphological and anatomical features for species confirmation and to fulfill the need of this species.

### **Materials and Methods**

The specimens were collected from Madaya Township, Mandalay Region from January 2015 to October 2015. The collected specimens were studied and identified in Department of Botany, University of Mandalay with the help of literatures (Hooker 1885; Hutchinson 1923; Backer 1963; Dassanayake 1987).

After the collection, some of the specimens were studied in fresh stage and others were pressed and dried at room temperature as well as preserved in 50% ethyl alcohol for further study. The leaves and stems of the specimens, as well as the plants in natural habits were studied and recorded.

After killing and fixation, the small portions of leaves, stems and roots were thoroughly washed in running water for overnight and dehydrated through a series of tertiary butyl alcohol solutions. Then, they were infiltrated in a series of paraffin wax of which melting points were 49°C, 54.5°C, 55 °C and 60°C respectively. They were kept in oven for

overnight or 24 hours. The specimens were then embedded in the 60°C paraffin wax and cut into 15-25 µm thick sections by using a rotary microtome. These thin sections were double-stained with the combination of safranin and fast green solutions. After staining, the sections were permanently mounted under a cover slip with a Canada Balsam in xylol and kept dried for a few weeks on slide tray. The dehydration, infiltration, embedding, staining and mounting were made according to Johansen's method (1940).

The parts of specimens were macerated by warming in equal volume of 50% hydrogen peroxide solution and 50% glacial acetic acid solution according to Jeffery's method (1917). The vessels elements, tracheids and fibers were studied and measured. The photomicrographs were also presented.

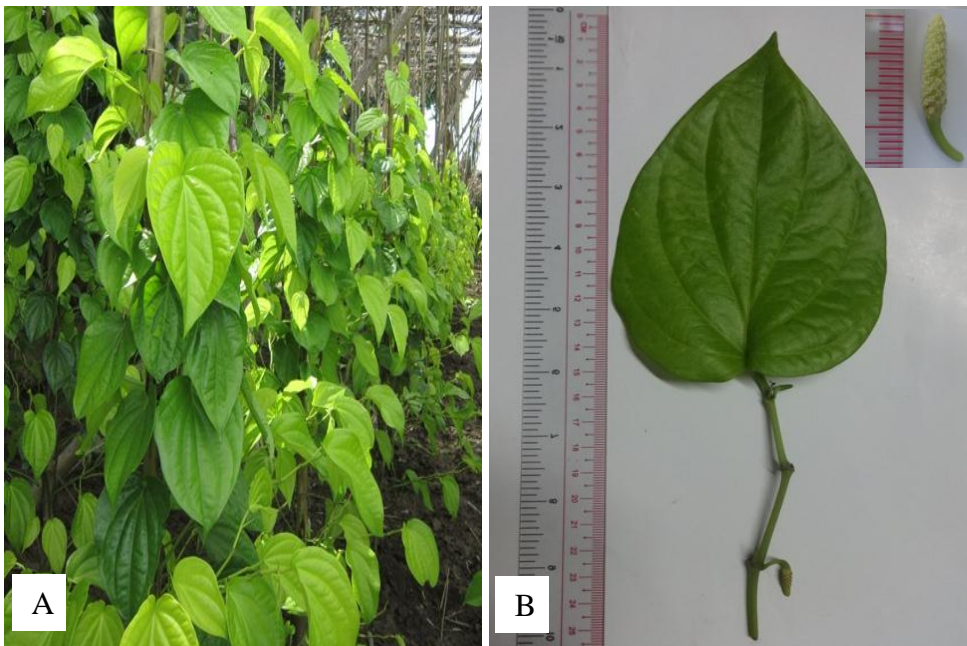
### **Morphological studies**

#### ***Piper betle* L. Sp. Pl. 28. 1753. (Figure 1)**

Family : Piperaceae  
 Local name : Kun nuwar  
 Common name: Betel

Perennial herbs, 2.0-6.0 m in height; root climbers; older stems ridged the younger stems smooth, tumid at the nodes, adventitious roots present. Leaves simple, alternate, petiolate, stipulate, reticulate venation; blades ovate, 8.0-18.0 cm by 6.0-12.0 cm, cordate at the base, entire along the margin, acuminate at the apex, glossy upper surface and coriaceous, yellowish green, 7- to 9- veined; petiole 2.0-6.0 cm by 0.2-0.3 cm, slightly canaliculate above; stipules bud scale, lanceolate, creamy, 2.2-4.7 cm long, adnate to the petiole. Inflorescences axillary spike; pistillate spikes 1.2-1.5 cm long, pendulous; peduncle 0.7-1.5 cm long. Flowers unisexual. Pistillate flowers pale yellow, minute, apetalous; ovary globoid, unilocular with one ovule in the locule on the pendulous placenta, sunken in fleshy receptacle; style short; stigma 4 to 5 - fid, pale yellow.

**Specimen Examined** : Mandalay Region, Madaya Township, Myokone village; 22 October, 2015; Win Win Khaing, collection no.1.



**Figure 1** Morphological Characters of *Piper betle* L.

- A. Plants in natural habit
- B. Branch with inflorescence

## Anatomical Studies

### Internal structure of the leaves of *Piper betle* L. (Figure 2)

#### Petiole

In transverse section, petiole (Figure 2 A) was heart shaped in outline, 2.0 - 3.0 mm in length and 2.5 - 3.0 mm in breadth, distinguishable into dermal, ground and vascular tissue systems.

**Dermal Tissue System:** In surface view, the cuticle smooth and the epidermal cells parenchymatous, rectangular in shape, anticlinal walls straight, cells 10.0 - 25.0  $\mu\text{m}$  in length and 10.0 - 35.0  $\mu\text{m}$  in breadth;

trichomes uniseriate, 1 to 3 - celled, 10.0 - 75.0  $\mu\text{m}$  in length, secretory cells present.

**Ground Tissue System:** Differentiated into collenchymatous and parenchymatous tissues, collenchymatous tissue composed of 7 to 10 - layered, the layers 125.0 - 175.0  $\mu\text{m}$  in thickness, angular type, cells polygonal in shape, 10.0 - 25.0  $\mu\text{m}$  in diameter. Parenchymatous tissue composed of 30 to 37 - layered and 2062.5  $\mu\text{m}$  in thickness, cells polygonal in shape, 30.0 - 75.0  $\mu\text{m}$  in diameter. A large mucilage canal 100.0 - 175.0  $\mu\text{m}$  in length and 100.0 - 200.0  $\mu\text{m}$  in breadth embedded in the center of parenchymatous tissues and 3 - 6 mucilage canals arranged in a ring. Each canal 60.0 - 125.0  $\mu\text{m}$  in diameter, situated opposite to the vascular bundles.

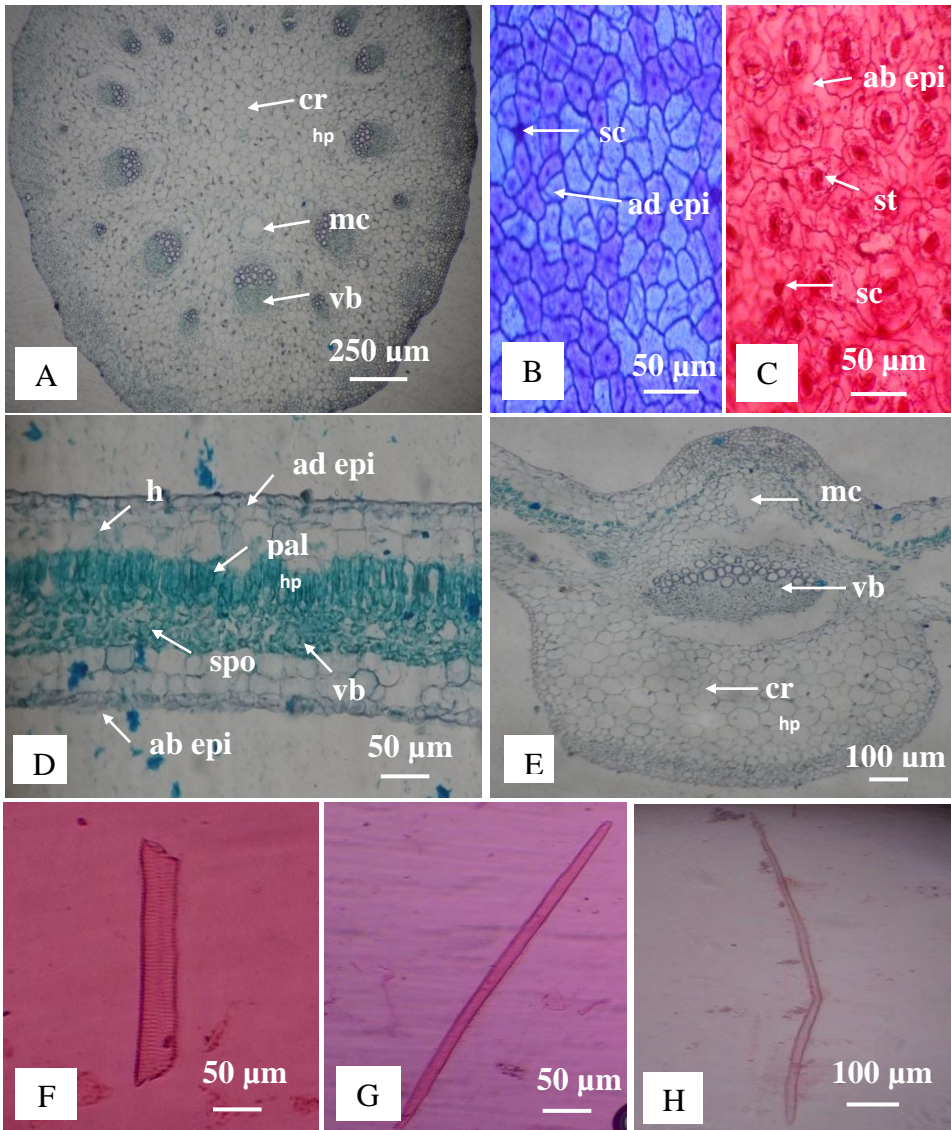
**Vascular Tissue System:** Vascular bundles arranged in a ring, embedded in the ground tissue, circular or oval shaped in transverse section, colletral type, each bundle consisted of 11 to 12 - inner large bundles, 172.8 - 384.0  $\mu\text{m}$  in length and 120.0 - 288.0  $\mu\text{m}$  in breadth, alternating with 3 to 5 - small accessory bundles, 96.0 - 168.0  $\mu\text{m}$  in length and 72.0 - 124.8  $\mu\text{m}$  in breadth, each bundle surrounded by continuous ring of 2 to 3 - layers of parenchymatous bundle sheaths. Phloem tissues 50.0 - 175.0  $\mu\text{m}$  in thickness and composed of sieve tube elements and companion cells; xylem tissues 50.0 - 200.0  $\mu\text{m}$  in thickness and composed of scalariform and spirally thickened vessels, tracheids, fibers and xylem parenchyma cells. Vessels were measured 100 - 600  $\mu\text{m}$  (mean 266.3  $\mu\text{m}$ ) in length and 10 - 40  $\mu\text{m}$  (mean 23.7  $\mu\text{m}$ ) in breadth. Tracheids were measured 120 - 500  $\mu\text{m}$  (mean 303.0  $\mu\text{m}$ ) in length, 10 - 25  $\mu\text{m}$  (mean 15.7  $\mu\text{m}$ ) in breadth. Fibers were measured 250 - 1090  $\mu\text{m}$  (mean 476.5  $\mu\text{m}$ ) in length, 5 - 15  $\mu\text{m}$  (mean 10.9  $\mu\text{m}$ ) in breadth (Figure 2).

### **Lamina**

In transverse section, lamina (Figure 2 D) was dorsiventral with reticulate venation, 125.0 - 215.0  $\mu\text{m}$  thick, distinguishable into dermal, ground and vascular tissue systems.

**Dermal Tissue System:** In surface view, the cuticle was smooth, epidermal cells of both surfaces were parenchymatous, polygonal, anticlinal walls wavy and thin walled. Adaxial epidermal cells 15.0 - 75.0  $\mu\text{m}$  in length, 15.0 - 50.0  $\mu\text{m}$  in breadth, secretory cells present. Stomata present on the abaxial surface, anisocytic type, oval shaped with reniform





**Figure 2 Internal structure of leaf of *Piper betle* L.**

A. Transverse section of petiole

B. Adaxial surface view of lamina

C. Abaxial surface view of lamina

D. Transverse section of lamina

(ab epi = abaxial epidermal cell, ad epi = adaxial epidermal cell, cr = cortex,

h = hypodermis cell, mc = mucilage canal, pal = palisade parenchyma cell,

sc = secretory cell, spo = spongy parenchyma cell, st = stoma, vb = vascular bundle)

E. Transverse section of midrib

F. Vessel element

G. Tracheary element

H. Fiber

shaped guard cells. The guard cells 20.0 - 25.0  $\mu\text{m}$  in length and 5.0 - 10.0  $\mu\text{m}$  in breath. Abaxial cells 20.0 - 50.0  $\mu\text{m}$  in length, 15.0 - 50.0  $\mu\text{m}$  in breadth, absence of stomata and presence of secretory cells. In transverse section, adaxial epidermis composed of 1 outer epidermal layer and 2 to 3 - inner hypodermal layers, and abaxial epidermis composed of 1 outer epidermal layer and 1 to 2 - inner hypodermal layer; adaxial epidermal cells 10.0 - 15.0  $\mu\text{m}$  in length, 10.0 - 30.0  $\mu\text{m}$  in breadth; abaxial epidermal cells 5.0 - 15.0  $\mu\text{m}$  in length, 10.0 - 25.0  $\mu\text{m}$  in breadth; both epidermal cells rectangular or barrel in shape, outer wall convex; adaxial hypodermal layers 50.0 - 65.0  $\mu\text{m}$  thick, cells 15.0 - 35.0  $\mu\text{m}$  in length and 15.0 - 40.0  $\mu\text{m}$  in breath, rectangular in shape, thin and straight or slightly curved anticlinal walls. Lower hypodermal cells mostly similar to those of the adaxial hypodermal cells. Secretory cells present in hypodermal layer. Cuticle thin on both surfaces.

**Ground Tissue System:** Mesophyll differentiated into palisade parenchyma at upper side and spongy parenchyma at the lower side; palisade cells 1 - layered, 25.0 - 35.0  $\mu\text{m}$  in thickness, the cells elongated in shape; cells compactly arranged; the spongy parenchyma cells 3 to 5 - layered, the layers 50.0 - 65.0  $\mu\text{m}$  in thickness, rounded to oval in shape, the cells 5.0 - 10.0  $\mu\text{m}$  in diameter.

**Vascular Tissue System:** Vascular bundles of lateral veins were embedded in the mesophyll tissues. They were collateral type and different in size according to their position; bundle sheath distinct and composed of parenchymatous cells, rounded or oval in shape. Phloem composed of sieve tubes, companion cells and phloem parenchyma. Xylem composed of scalariform and spirally thickened vessels, tracheids, fibers and xylem parenchyma. Vessels were measured 100 - 800  $\mu\text{m}$  (mean 370.0  $\mu\text{m}$ ) in length and 10 - 30  $\mu\text{m}$  (mean 19.8  $\mu\text{m}$ ) in breadth. Tracheids were measured 50 - 450  $\mu\text{m}$  (mean 194.8  $\mu\text{m}$ ) in length, 5 - 20  $\mu\text{m}$  (mean 9.3  $\mu\text{m}$ ) in breadth. Fibers were measured 275 - 1200  $\mu\text{m}$  (mean 565.7  $\mu\text{m}$ ) in length, 5 - 20  $\mu\text{m}$  (mean 10.9  $\mu\text{m}$ ) in breadth (Figure 2).

## Midrib

In transverse section, midrib (Figure 2 E) was subcircular, 850.0 - 1500.0  $\mu\text{m}$  in length and 875.0 - 1175.0  $\mu\text{m}$  in breadth, distinguishable into dermal, ground and vascular tissue systems.

**Dermal Tissue System:** In surface view, the epidermal cells parenchymatous, rectangular in shape, cell walls thin, anticlinal walls straight, cells 15.0-35.0  $\mu\text{m}$  in length and 15.0-50.0  $\mu\text{m}$  in breadth; the trichomes uniseriate, 10.0-35.0  $\mu\text{m}$  in length, secretory cells present. In transverse section, both adaxial and abaxial epidermal cells 1- layered, oval to barrel in shape, 5.0-15.0  $\mu\text{m}$  in length and 10.0-25.0  $\mu\text{m}$  in breadth, outer wall convex, anticlinal walls straight; cuticle thin, smooth on both surfaces.

**Ground Tissue System :** Lying internal to the epidermis made up of two types of cells, collenchyma and parenchyma; adaxial side collenchymatous cells, 3 to 10 - layered, 50.0 - 110.0  $\mu\text{m}$  in thickness, angular type, rounded or polygonal in shape; parenchymatous cells 3 to 6 - layered, 48.0 - 96.0  $\mu\text{m}$  thick, cells 10.0 - 25.0  $\mu\text{m}$  in length and 10.0 - 35.0  $\mu\text{m}$  in breadth, oval, rounded or polygonal in shape; abaxial side collenchymatous cells 3 to 6 - layered, 15.0 - 30.0  $\mu\text{m}$  thick, cells 5.0 - 15.0  $\mu\text{m}$  in diameter, angular type, oval, rounded, irregular or polygonal in shape; parenchymatous cells 6 to 12 - layered, 85.0 - 325.0  $\mu\text{m}$  thick, cells 15.0 - 55.0  $\mu\text{m}$  in length and 15.0 - 50.0  $\mu\text{m}$  in breadth, oval, polygonal in shape; intercellular space absent. A single mucilage canal 50.0 - 75.0  $\mu\text{m}$  in diameter embedded in adaxial side of parenchymatous cells.

**Vascular Tissue System:** A large vascular bundle is embedded in the ground tissue, circular or oval shape, 250.0 - 350.0  $\mu\text{m}$  in length and 300.0 - 350.0  $\mu\text{m}$  in breadth, colletral type, surrounded by continuous or discontinuous ring of pericyclic parenchymatous cells, 2 to 3 - layered, polygonal in shape. Phloem tissues were 75.0 - 100.0  $\mu\text{m}$  in thickness and composed of sieve tube elements and companion cells; xylem tissues 75.0 - 125.0  $\mu\text{m}$  in thickness and composed of scalariform and spirally thickened vessels, tracheids, fibers and xylem parenchyma. Vessels were measured 100 - 500  $\mu\text{m}$  (mean 253.1  $\mu\text{m}$ ) in length and 10 - 30  $\mu\text{m}$  (mean 19.6  $\mu\text{m}$ ) in breadth. Tracheids were measured 35 - 1050  $\mu\text{m}$  (mean 539.4  $\mu\text{m}$ ) in length, 5 - 25  $\mu\text{m}$  (mean 16.3  $\mu\text{m}$ ) in breadth. Fibers were measured 200 - 1700  $\mu\text{m}$  (mean 820.4  $\mu\text{m}$ ) in length, 5 - 15  $\mu\text{m}$  (mean 11.3  $\mu\text{m}$ ) in breadth (Figure 2).

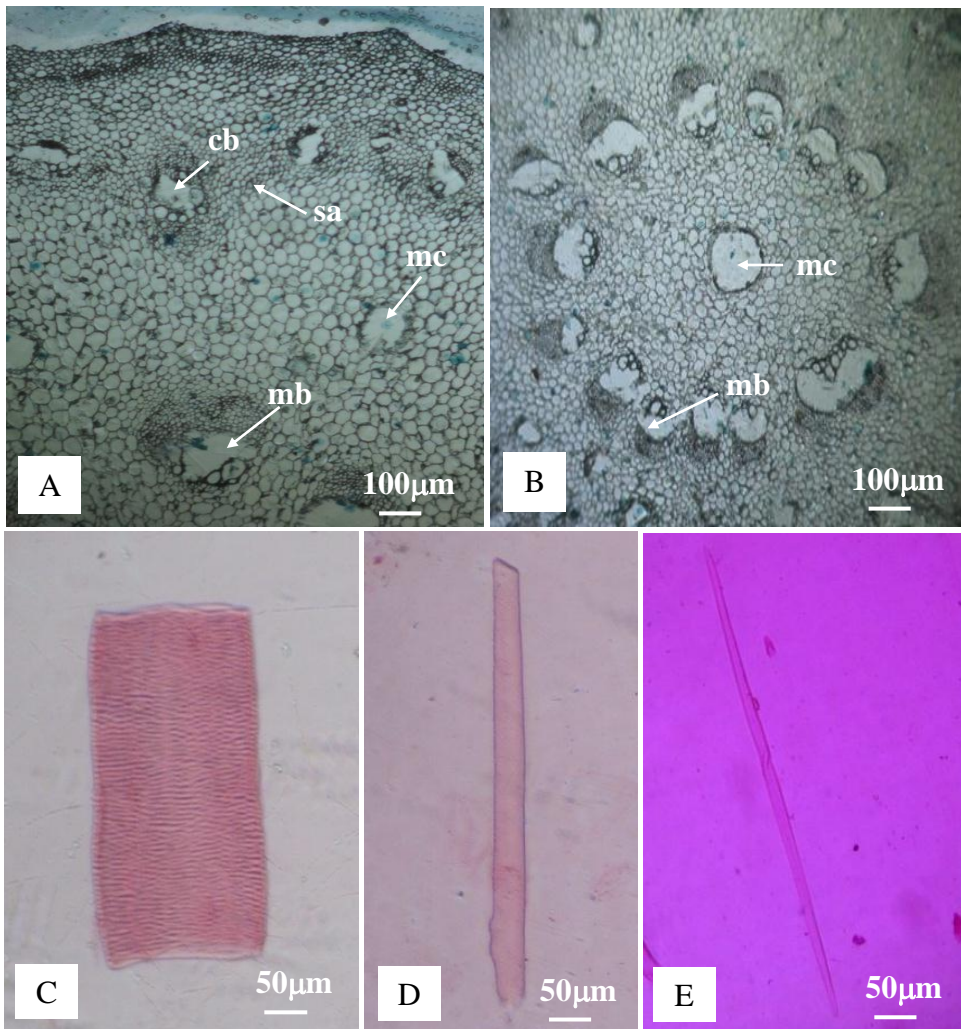
### **Internal structure of the stem of *Piper betle* L. (Figure 3)**

In transverse section, stem (Figure 3) was oval shaped in outline, 3.0 - 4.0 mm in tangential diameter and 3.0 - 3.5 mm in radial diameter, distinguishable into dermal, ground and vascular tissue systems.

**Dermal Tissue System:** In transverse section, epidermal cells 1-layered, oval to barrel in shape, 5.0 - 15.0  $\mu\text{m}$  in length and 5.0 - 10.0  $\mu\text{m}$  in breadth, outer wall convex, anticlinal walls straight; cuticle thin.

**Ground Tissue System:** Differentiated into collenchymatous and parenchymatous tissues, the outer cortex collenchymatous tissue composed of 5 to 8 - layered, the layers 50.0 - 90.0  $\mu\text{m}$  in thickness, angular type, cells polygonal in shape, 5.0 - 25.0  $\mu\text{m}$  in diameter, forming a continuous ring beneath the hypodermis, if interrupted them the patches are separated by the extension of inner cortex. The inner cortex parenchymatous tissue composed of 6 to 10 - layered and 75.0 - 155.0  $\mu\text{m}$  in thickness, cells polygonal in shape, 10.0 - 27.5  $\mu\text{m}$  in diameter. Endodermis and pericycle were indistinct. Pith parenchymatous tissue composed of 16 to 34 - layered, cells circular in shape, 15.0 - 55.0  $\mu\text{m}$  in diameter. A large mucilage canal 165.0 - 190.0  $\mu\text{m}$  in length and 100.0 - 150.0  $\mu\text{m}$  in breadth was embedded in center of pith parenchymatous tissues, minute prismatic crystals of calcium oxalate were seen in the mucilage duct. In peripheral region, a ring of 9 to 12 - mucilage canals were present 60.0 - 95.0  $\mu\text{m}$  in length and 45.0 - 80.0  $\mu\text{m}$  in breadth; circular secretory cells present in both outer and inner cortex.

**Vascular Tissue System:** Vascular bundles embedded in the ground tissue, oval shape in transverse section, occurred in a ring of two rows, cortical and medullary bundles, the inner medullary bundles are larger than the outer cortical bundles; cortical bundles small and many, one third of them are larger than the others, all embedded in a wavy ring of 5 to 9 - cells thick sclerenchymatous tissues, cells polygonal, 10.0 - 20.0  $\mu\text{m}$  in diameter with thickened walls; medullary bundles were 16 - 20, all bundles 190.0 - 275.0  $\mu\text{m}$  in radial diameter and 115.0 - 200.0  $\mu\text{m}$  in tangential diameter. Both of vascular bundles were collateral with phloem above and xylem below, separated by cambium; cambium composed of 4 - 5 layers of tangentially elongated rectangular cells; bundle sheath present and composed of parenchymatous cells, round or oval in shape. Phloem tissues



**Figure 3** Internal structure of stem of *Piper betle* L.

A. T.S of a portion of stem

B. T.S of stem showing inner cortex with a ring of vascular bundles

C. Vessel element

D. Tracheary element

E. Fiber

(cb=cortical bundle, mb=medullary bundle, mc=mucilagenous canal, sa=sclerenchyma)

75.0 - 137.5  $\mu\text{m}$  in thickness and composed of sieve tube elements and companion cells; xylem tissues 70.0 - 150.0  $\mu\text{m}$  in thickness and composed of scalariform and spirally thickened vessels, tracheids, fibers and xylem parenchyma cells. Vessels were measured 50 - 875  $\mu\text{m}$  (mean 378.7  $\mu\text{m}$ ) in length and 10 - 175  $\mu\text{m}$  (mean 89.8  $\mu\text{m}$ ) in breadth. Tracheids were measured 150 - 600  $\mu\text{m}$  (mean 315.2  $\mu\text{m}$ ) in length, 5 - 35  $\mu\text{m}$  (mean 20.6  $\mu\text{m}$ ) in breadth. Fibers were measured 250 - 1485  $\mu\text{m}$  (mean 829.8  $\mu\text{m}$ ) in length, 5 - 25  $\mu\text{m}$  (mean 13.0  $\mu\text{m}$ ) in breadth (Figure 3).

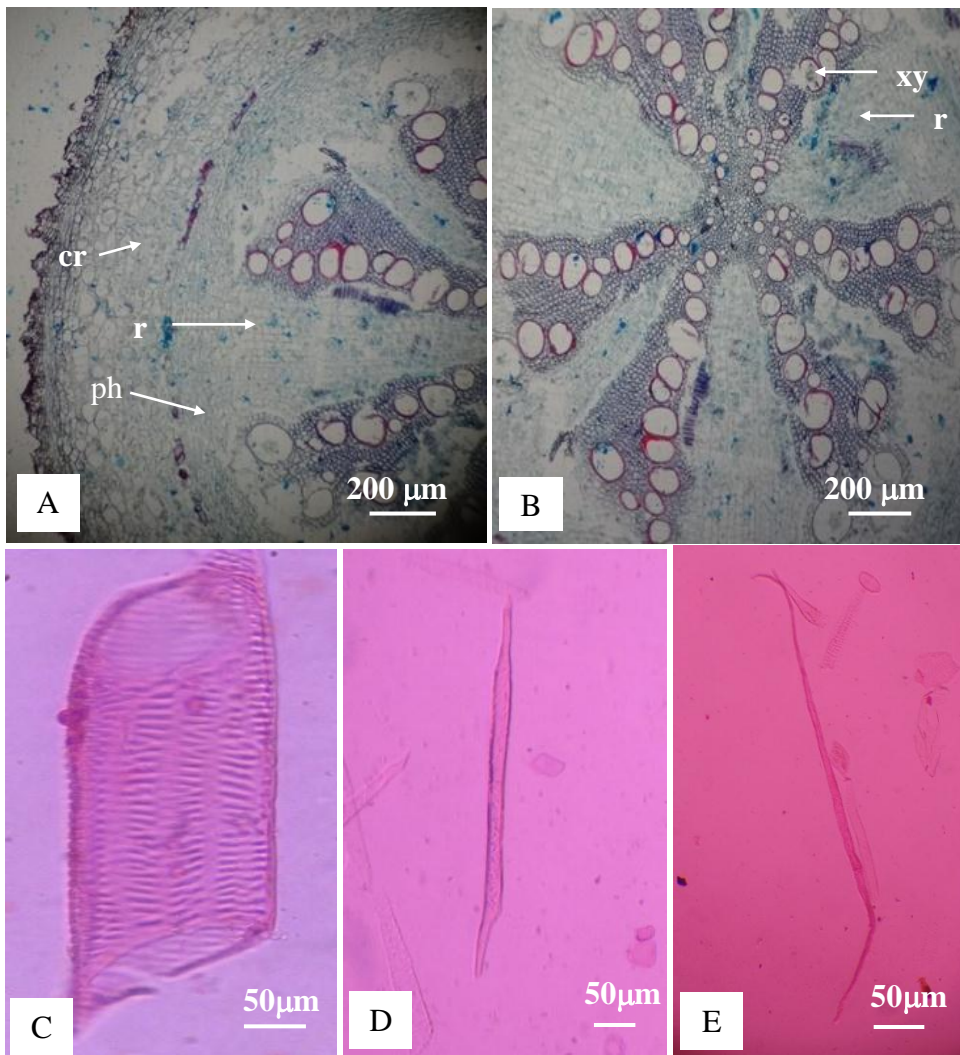
#### **Internal structure of the root of *Piper betle* L. (Figure 4)**

In transverse section, root of *Piper betle* L. (Figure 4) was circular in outline, 2.5 - 3.5 mm in diameter. Distinguishable into dermal, ground and vascular tissue systems.

**Dermal Tissue System:** The root epidermis 1 - layered, parenchymatous cells, the cells 15.0 - 35.0  $\mu\text{m}$  in length, 20.0 - 50.0  $\mu\text{m}$  in breadth.

**Ground Tissue System:** Composed of cortex, endodermis and pericycle. Cortex homogenous parenchymatous cells, 11 to 14 - layered, the layers 400.0 - 500.0  $\mu\text{m}$  in thickness, the cells 25.0 - 60.0  $\mu\text{m}$  in tangential diameter, 15.0 - 60.0  $\mu\text{m}$  in radial diameter, polygonal or oval in shape, many circular or rhomboidal crystals occurred throughout the cortical parenchyma cells. Endodermis 1- layered continuous, the cell 10.0 - 40.0  $\mu\text{m}$  in tangential diameter, 25.0 - 60.0  $\mu\text{m}$  in radial diameter, parenchymatous barrel shaped; pericycle uniseriate, discontinuous, cells 10.0 - 25.0  $\mu\text{m}$  in tangential diameter and, 25.0 - 60.0  $\mu\text{m}$  in radial diameter, barrel shaped. Pith absent; central part of root occupied by xylem parenchyma. Mucilage canal absent.

**Vascular Tissue System:** In transverse section, vascular cylinder triarch, 3 xylem plates, bifurcated once or twice and radiating from the center, the plates being separated by rays, rays 7 - 19 cells broad and 350.0 - 450.0  $\mu\text{m}$  thick, the bundles 1150.0 - 1375.0  $\mu\text{m}$  in thickness, phloem distributed above the xylem beneath the pericycle; phloem 9 to 12 - layered, the layers 150.0 - 200.0  $\mu\text{m}$  thick, cells 10.0 - 15.0  $\mu\text{m}$  in tangential diameter, 20.0 - 35.0  $\mu\text{m}$  in radial diameter; phloem composed of sieve tubes and companion cells; xylem towards the centre, 975.0 - 1150.0  $\mu\text{m}$  thick, cells 25.0 - 190.0  $\mu\text{m}$  in tangential diameter, 30.0 - 175.0  $\mu\text{m}$  in radial diameter,



**Figure 4** Internal structure of root of *Piper betle* L.

- A. T.S of a portion of root
  - B. T.S of root showing close up view of vascular bundle
  - C. Vessel element
  - D. Tracheary elements
  - E. Fiber
- (cr = cortex, ph = phloem, r = ray, xy = xylem)

xylem composed of vessel elements, tracheids, xylem fibres and xylem parenchyma. Vessels were measured 100 - 300  $\mu\text{m}$  (mean 175.2  $\mu\text{m}$ ) in length and 35 - 175  $\mu\text{m}$  (mean 115.7  $\mu\text{m}$ ) in breadth. Tracheids were measured 150 - 390  $\mu\text{m}$  (mean 271.7  $\mu\text{m}$ ) in length, 5 - 30  $\mu\text{m}$  (mean 16.9  $\mu\text{m}$ ) in breadth. Fibers were measured 275 - 700  $\mu\text{m}$  (mean 461.1  $\mu\text{m}$ ) in length, 5 - 15  $\mu\text{m}$  (mean 10.4  $\mu\text{m}$ ) in breadth (Figure 4).

### Discussion and Conclusion

The morphological and anatomical characters of *Piper betle* L. belong to family Piperaceae were studied in this research. The betel vines from Madaya were perennial herbs, root climbers, tumid at the nodes, adventitious roots present. The plants were cultivated plants for harvesting green leaves. The maximum length of leaves was 18.0 cm and 12.0 cm in maximum breadth.

In the present study, The leaves were found to be simple, alternate, petiolate, stipulate, coriaceous, ovate, acuminate apex, entire margin which were agreed with Chibber 1912; Dassamayake 1987; Nianhe *et al.* 1999; Ravindran 2000; Lakshmi & Naidu 2010; Chahal *et al.* 2011; Selvadurai *et al.* 2011; Periyanyagam *et al.* 2011.

The female inflorescence of *Piper betle* L. was pendulous and 3.0-5.0 cm long (Nianhe *et al.* 1999), 3.5-5.0 cm long (Dassanayake, 1987). In the present study, the size of inflorescences was different from previous researchers.

In this research, transverse sections of laminae, midribs, petioles, stems and roots of *Piper betle* L. showed the three tissue systems viz. dermal tissue system, ground tissue system and vascular tissues system. Dermal tissues composed of one layer of epidermal cells on both surfaces and thin cuticle layer. Anisocytic type of stomata was found in surface layers of lower epidermis and they were in an agreement with Metacalfe & Chalk 1950. In transverse section, adaxial epidermis was composed of an outer epidermal layer and 2 to 3 inner hypodermal layers, and abaxial epidermis was composed of an outer epidermal layer and 1 to 2 inner hypodermal layers. All of the epidermal cells were rectangular or barrel in shape.



The ground tissue of lamina was differentiated into upper palisade and lower spongy parenchyma. Palisades were one-layered and composed of elongated cells, cells were compactly arranged. The spongy layers were 3- to 4- layered, cells were rounded to oval in shape, these characters were in agreement with Chibber 1912, Lakshmi & Naidu 2010, Selvadurai *et al.* 2011 and Raman *et al.* 2012.

The transverse section of midrib was subcircular or oval-shaped. Dermal tissue system was composed of epidermal cells and trichomes, epidermal cells were oval to barrel in shape, cuticle smooth on both surfaces. The ground tissue of midrib was differentiated into two types of tissues, collenchymatous and parenchymatous, collenchymatous cells below the adaxial epidermis were 3- to 10- layered and above the abaxial epidermis were 3- to 6- layered, the main mass of midrib was thin-walled parenchymatous cells. A large vascular bundle was present, collateral type and circular or oval in shape, a single mucilage canal was found the adaxial side of vascular bundle; these characters were in agreement with Lakshmi & Naidu 2010, Selvadurai *et al.* 2011 and Mubeen *et al.* 2014.

The transverse section of petioles was found to be heart-shaped, composed of dermal tissue, ground tissue and vascular tissue system. Dermal tissue system composed of epidermal cells and trichomes. In surface view, anticlinal walls were straight. Trichomes are uniseriate and 1 to 3-celled, 10.0-75.0  $\mu\text{m}$  long. The ground tissue of petioles differentiated into collenchymatous and parenchymatous cells. Collenchyma was next to epidermis. A large mucilage canal was observed in the centre of petioles and 4-5 mucilage canals were also observed in the parenchyma tissues. Vascular tissues were separated, variable number of large bundles alternating with small bundles, collateral type, circular or oval in shape. These were agreed with Metcalfe & Chalk 1950, Lakshmi & Naidu 2010, Selvadurai *et al.* 2011 and Raman *et al.* 2012.

The transverse section of stems of *Piper betle* L. was oval in shape, composed of dermal tissue, ground tissue and vascular tissue system. Epidermal cells were rectangular or polygonal in shape, in surface view anticlinal wall straight. In the ground tissue system, cortex is composed of outer collenchyma and inner parenchyma cells. Pith was composed of many layers of parenchymatous cells; cells were circular or oval in shape. A large mucilage canal was observed in the centre of pith, minute

prismatic crystals of calcium oxalate were seen in the mucilage duct. A ring of variable number of mucilage canal was also observed in the peripheral region. Endodermis and pericycle were indistinct. Vascular bundles of stems were two concentric rings. They were collateral type and variable number. These characters were agreement with Metcalfe & Chalk 1950 and Roman *et al.* 2012.

In transverse section of roots of *Piper betle* L. were circular in outline. The root epiblema were uniseriate parenchymatous cells. Cortex layers were composed of only parenchymatous cells. Many circular or rhomboidal secretory cells were observed in the cortical layers. The vascular bundles were observed in triarch. Pith absent. These characters were similar as mentioned by Metcalfe & Chalk 1950.

In conclusion, the present research can provide the information of morphological and anatomical characters of *Piper betle* L. from Mandalay Region. It is hoped that these finding were useful in species confirmation.

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