



## MORPHO-ANATOMICAL AND PHYTOCHEMICAL STUDIES OF *COMMENLINA BENGHALENSIS* L. OF *COMMENLIACEAE*

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### ABSTRACT

The bioactive substances or chemical constituents naturally occur in the organ of medicinal plants and they have therapeutic importance. On this basic information, the perennial monocotyledon herb *Commelina benghalensis* L. of *Commeliaceae* was examined by morpho-anatomical and phytochemical studies (Qualitative and Quantitative) were made. The most important bioactive compounds such as alkaloids, catechin, flavonoids, phenols, tannins, steroids, glycosides etc were presents in the entire dried sample. Quantification of physicochemical, biochemical and photosynthetic pigments were notably presents in the dried shoot and root of *C. benghalensis*.

**Key words:** *Commelina benghalensis*, Morpho - anatomy, Phytochemicals and Quantification.

### INTRODUCTION

Medicinal plants are the major sources of medicines in Ayurvedha, Siddha, and Folk medicine systems. In India about 95% of all modern drugs are derived from medicinal plants and very likely most of these medicines are used by people to cure many ailments. The medicinal plants are rich in secondary metabolites and essential oil which are of therapeutic importance. The presence study deals with morpho-anatomical characters, phytochemical studies such as physicochemical and biochemical estimation of medicinal plant of *C. benghalensis*. L. It is a benghal day flower and also known as tropical spiderwort, is an herbaceous perennial and a troublesome weed, native to Africa and tropical Asia. They belong to this family *Commeliaceae* and genus *Commelina*. Whole part of the plant is medicinally useful. Plant is bitter, emollient, demulcent, refrigerant, laxative and beneficial in leprosy treatment. The plant is used medicinally but as a laxative to cure

inflammations of the skin as well as leprosy (Kaiser *et al.*, 1975). The people of Nepal eat the young leaves as a vegetable, use a paste from the plant to treat burns and the fluid from the roots is used to treat indigestion (Manandhar *et al.*, 2002).

### MATERIALS AND METHODS

#### Collection of plant materials

*Commelina benghalensis* L. was collected from near costal area of Tiruchendur Taluk in Tuticorin district, Tamil Nadu, India. Herbarium specimen was prepared and identified based on the keys given by (Oladipo *et al.*, 2014). The collected plant was authenticated and voucher specimen (Voucher No. ACBH34) was lodged in the Botany Research Laboratory, Aditanar College of Arts and Science, Virapandianpatnam, Tiruchendur, Tamil Nadu, India.

#### Morpho-anatomical observations

##### Macroscopic studies

The fresh plant was used for the study of macroscopic characters such as habit, stem type, leaf type, phyllotaxy, leaf shape, leaf blade, leaf apex, leaf surface,

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leaf base, leaf margin, leaf venation, stem colour, root colour and flower colour, flower shape, length of plant, length of leaf, width of leaf, length of flower, width of flower, width of stem etc, as described by (Brain and Turner, 1975; Wallis 1985).

### Microscopic study

Anatomical study was carried out by taking free hand section of fresh leaves, stem and root of *C. benghalensis* L. Toluidine blue O, Eosin blue and Crystal violet was used to stain the sections. Photomicrographs were taken by using compound binocular microscope (Olympus CH20i) with built in analogue camera and the help of Adobe Photoshop version 11.0. of Computer.

### Preparation of plant materials

The whole plant was cleaned thoroughly in order to free it from dust, soils and other unwanted materials that may adhere to it. The plant was shade dried and then ground to fine powder using mortar and pestle and then stored in an appropriate container until required for further analysis.

### Physicochemical analysis

Physicochemical parameters such as total ash value, acid insoluble ash value, water soluble ash value and moisture content were determined as per method described Anonymous, 1996.

### Phytochemical screening

Two gm of powdered sample was taken and extracted by cooling percolation method using different solvents (Benzene, Petroleum ether, Chloroform, Acetone, Methanol and Water) at room temperature. Then the extract was collected and subjected for phytochemical analysis and identification of various phytochemical constituents as per standard procedures (Harborne, 1973; Brinda *et al.*, 1981 and Lala, 1993)

### Quantitative estimation

For biochemical analysis such as estimation of carbohydrate, protein, amino acid, total free phenols, tannins, organic carbon, chlorophyll *a*, *b*, total chlorophyll and carotenoids were carried out using standard methods. The total carbohydrate was estimated by anthrone method of Sheifter *et al.*, (1950). Protein was estimated by Lowry's method (Lowry *et al.*, 1951). Amino acid was estimated by ninhydrin method as suggested by Rosen, (1957). Other biochemical's like estimations of total free phenols by Folin-Ciocalteu's method by Sadasivam and Manickam (1992), tannin was vanillin-HCl method by Burns (1971), Vitamin A by (Eitenmiller and Landenm, 1998), Organic carbon by (Walkley and Black, 1943), and photosynthetic pigments by Arnon (1949) as modified by Harborne (1973) methods are followed.

## RESULTS AND DISCUSSION

### Morphological character

Morphological observations of *C. benghalensis* L. was presented in table 1. *C. benghalensis* L (Benghal dayflower) is a perennial herb 20-25 cm long, stem are round and hairy. Width of stem was 0.3-0.4 cm, stems often root at the nodes and purple-blue aerial flowers are funnel-shaped about 2 cm long 1.2 cm width. Roots are white and brown and fibrous. Underground stolons can produce subterranean flowers are cleistogamous reported by (Ferrell *et al.*, 2004). Stipules are absent. Leaves evenly distributed (alternately) on the stem, simple, 5cm length and 3-3.5 cm width, sub-sessile, leaf blade curved, leaf surface hairy and leaf sheath are present, leaves and above ground stems have short hairs (pubescent) and longer red hairs on the leaf sheath and petiole margins, margin is entire, apex acute and parallel-veined.

### Microscopic characters

#### Stem

Transverse section of *C. benghalensis* stem is circular in outline with trichomes emerging all over the surface. For microscopic observation, it consists of epidermis, hypodermis, ground tissue and vascular bundles. The epidermis is a single outer layer rectangular in shape with thick cuticle covering and the presence of hairs or trichomes are emerged from the epidermal cells. Next to epidermis constitute the hypodermis. It was 3 to 5 layers of thick walled cells collenchymatous followed by few layers of chlorenchymatous cells are present. The cells of the ground tissue are smaller in size, compactly arranged polygonal in shape. The vascular bundles are scattered in ground tissue, closed, collateral, enclosed by sclerenchymatous sheath was present. Starch grains, scleroides, and raphides (crystals) are present in the cortical cells. Spiral thickening of xylem vessels also present. These observations were confirmed early by (Tomlinson, 1969; Kausch and Horner, 1982, Madhavan *et al.*, 2010). (fig 1)

#### Root

Single outermost layer rhizodermis is thick walled cells and unicellular root hairs are present. Next to the rhizodermis followed by hypodermis one or two layers of thick walled parenchymatous cells were present followed by thin walled parenchymatous cells are present. Stone cells and intercellular space are present in the hypodermis. Broad parenchymatous ground tissues are smaller in size, compactly arranged polygonal shaped cells. The vascular bundles are scattered in ground tissue, closed, collateral and enclosed by sclerenchymatous sheath was present. Crystals were found in phloem parenchymatous cells and spirals of vessels and sieve plates also observed in xylem elements. (fig 2).

## Leaves

Transverse section of *C. benghalensis* leaves consists of an upper epidermis and lower epidermis. The epidermal cells are polygonal in shape. Amphistomatic types of stomata were present and abaxial side of the epidermis is more frequently. The presence of simple bicellular hooked non glandular trichomes on both surface of leaves. The epidermal morphology of *C. benghalensis* was also reported by Oladipo and Ayo-Ayinde (2014). The ground tissue system of leaf is known as mesophyll tissue. The presences of mesophyll tissue have lot of air spaces. The vascular bundles are collateral and closed. Kidney shaped, hexacytic type stomata were present. This observation was agreed in earlier work of Chandurkar (1971). Xylem vessels and trachids are also present in the vascular bundle. Stone cells of astrosclereids and osteosclereids were present in the mesophyll tissue. Crystals and starch grains are present in the mesophyll tissue. (fig 3).

## Physicochemical analysis

The dried root and shoot of *C. benghalensis* L plant powder was carried out to the physicochemical analysis. In this study moisture content, total ash value, acid insoluble ash value, and water soluble ash value were determined is presented in Table - 2. The moisture content of shoot ( $13.047 \pm 0.904\%$ ) and root ( $14.307 \pm 0.142\%$ ) were recorded. Total ash, acid insoluble and water soluble ash value of the shoot via  $16.33 \pm 0.163\%$ ,  $1.267 \pm 0.147\%$ ,  $11.250 \pm 1.97\%$  and root via  $17.987 \pm 0.131\%$ ,  $7.533 \pm 0.178\%$ , and  $10.170 \pm 0.111\%$  were respectively noted. The amount of composition of ash remaining after combustion of plant material varies considerably according to the part of the plant, age, treatment etc. the constituents of the ash also vary with time and from organ to organ. Ash usually represents the inorganic part of the plant (Vermani Archa *et al.*, 2010). Ash value is useful in determination authenticity and purity of sample and also these values are of important qualitative standards. When the plant is consumed higher value of acid insoluble ash indicates the higher digestability (Lethika Nair *et al.*, 2012). The amount of inorganic matter present in the sample and the acid insoluble ash almost with in 1.5% which expresses low siliceous matter present in the sample (Kumar S *et al.*, 2014).

## Phytochemical screening

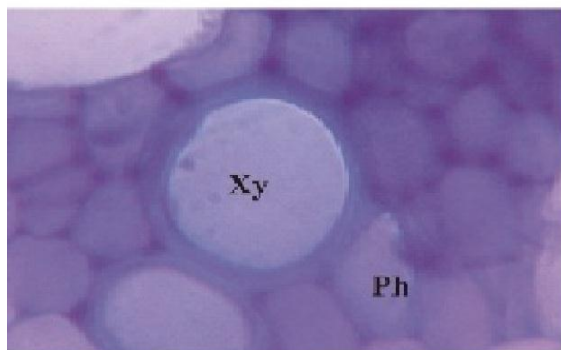
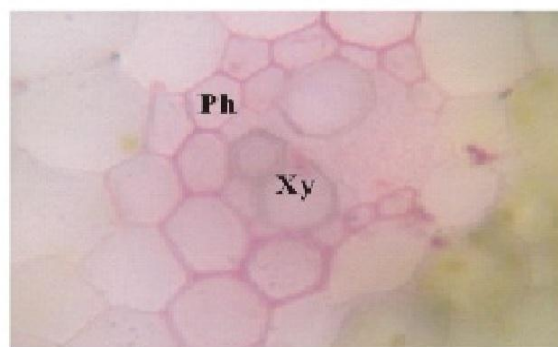
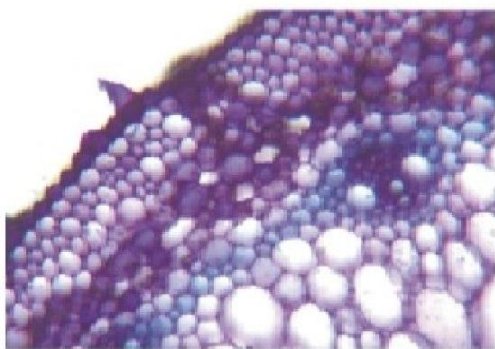
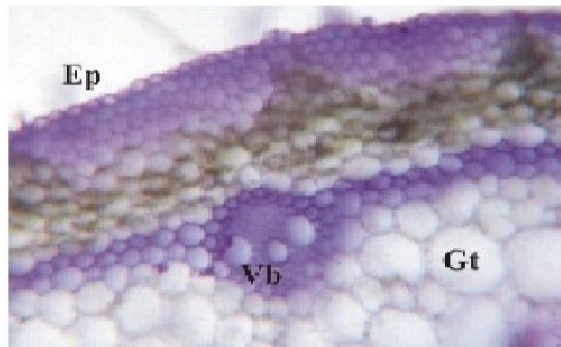
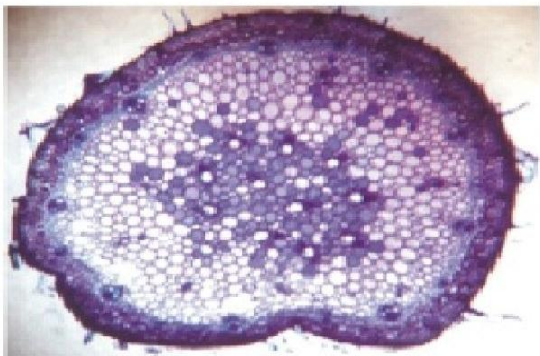
The phytochemical screening of *Commelina benghalensis* L. was presented in Table-3. The bioactive compound like alkaloids, catechin, flavonoids, phenols, quinones, saponins, tannins, sugar, glycosides, protein and amino acids were presents. The compound like anthraquinones, coumarin, and xanthoprotein are absent. The traditional uses of this plant *C. benghalensis* reacted against infection by wounds or burns (Mohammad A A Khan *et al.*, 2011) and also this medicinal plant has

antibacterial activity. Phytochemical screening is one of the techniques to identify new sources of therapeutically and industrially important compounds present in the plant extract. The chemical constituents of plants are desirable the synthesis of new bioactive compounds for treating the specific disease such phytochemical screening in various plants is reported by many workers. The plant is also suggested to have diuretic properties (Jayvir *et al.*, 2002). The presence of tannin shows that the plant is astringent as documented and suggests that it might have antiviral and antibacterial activity and can aid in wound healing and burns (Haslem, 1989; Ibrahim *et al.*, 2010). The saponins and glycoside are also very important classes of secondary metabolites as some are cardio-active and used in treatment of heart conditions (Oloyode, 2005). The plant body of *C. benghalensis* L is used in the treatment of leprosy and leucoderma (Mukerjee, 2006).

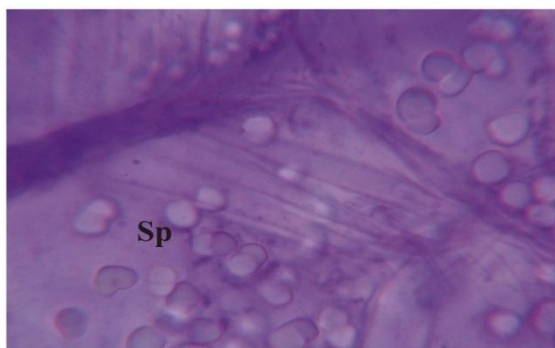
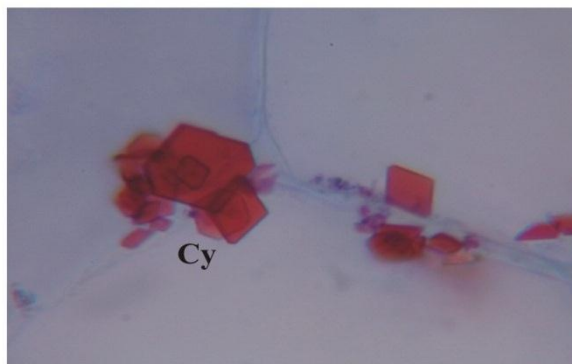
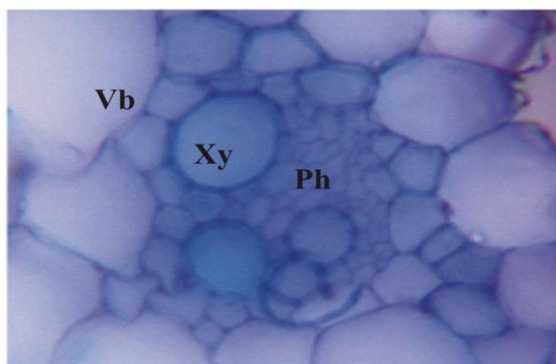
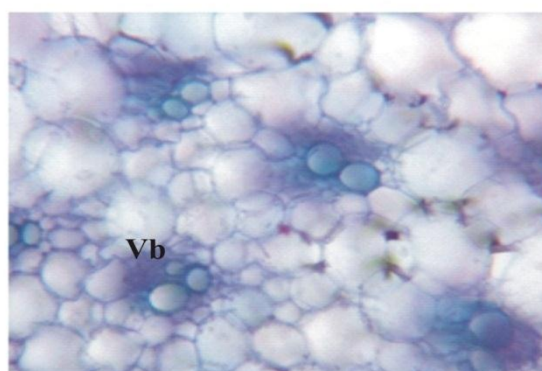
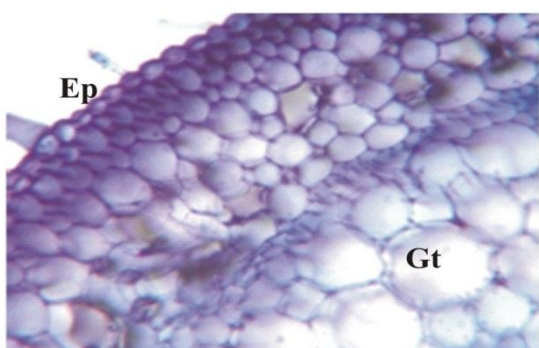
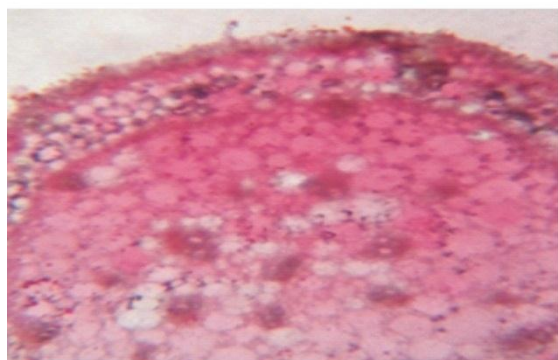
## Biochemical Analysis

Biochemical constituents such as carbohydrate, protein, amino acid, vitamin A and organic carbon of dried shoots and roots *C. benghalensis* L. were presented in Table 4. The primary metabolites of carbohydrate ( $0.582 \pm 0.009$  mg/g and  $0.005 \pm 0.0004$  mg/g), protein ( $15.62 \pm 0.134$  and  $1.08 \pm 0.025$  mg/g), and amino acid ( $1.191 \pm 0.013$  and  $0.269 \pm 0.010$  mg/g) were presented in the shoot and root system respectively. The presence of higher protein level in the plant towards their possible increase in food value and that a protein base bioactive compound could also be isolated in future (Thomsan *et al.*, 1991). The vitamin A content of shoot ( $17.48 \pm 0.205$ ) and root ( $13.6 \pm 0.277$  mg/g), organic carbon content shoot ( $22.6 \pm 0.145$ ) and root ( $15.2 \pm 0.119\%$ ) were presented. The secondary metabolites like total free phenols ( $44.8 \pm 0.438$  mg/g) and ( $12.8 \pm 0.118$  mg/g), tannins in ( $10.7 \pm 0.26$  and  $1.3 \pm 0.159$  mg/g) were presented in the dried shoot and root of *C. benghalensis* L. respectively. Medicinal plants have provided a good source of a wide variety of compounds, such as phenolic compound vitamins and some other secondary metabolites with are rich in valuable bioactivities such as anti-inflammatory agent, antibacterial, antifungal and antiviral activities (Lai, 2014, Tapseell, 2006 and Soni *et al.*, 2013).

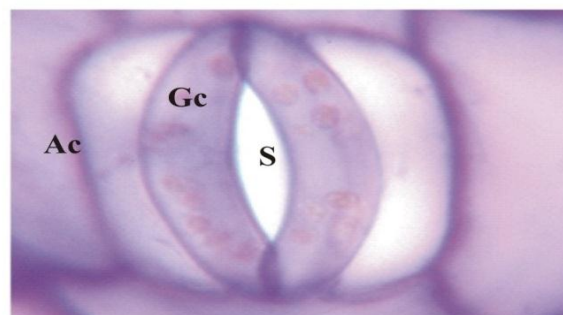
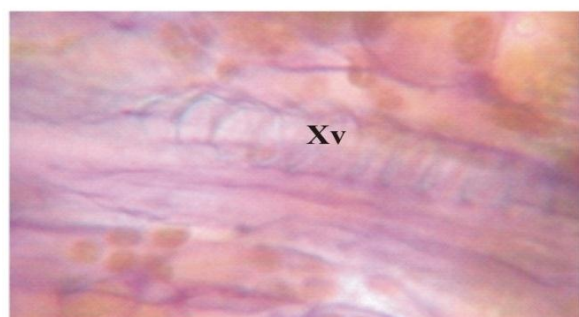
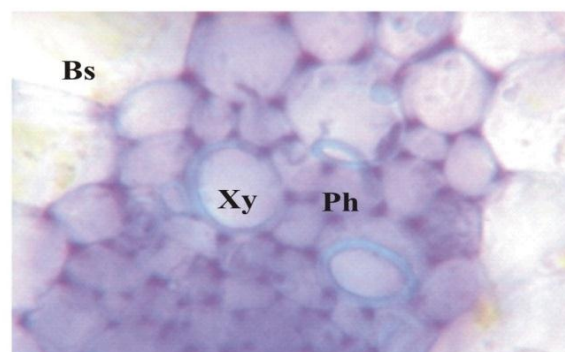
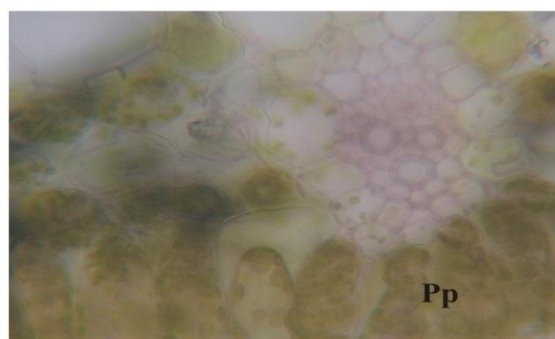
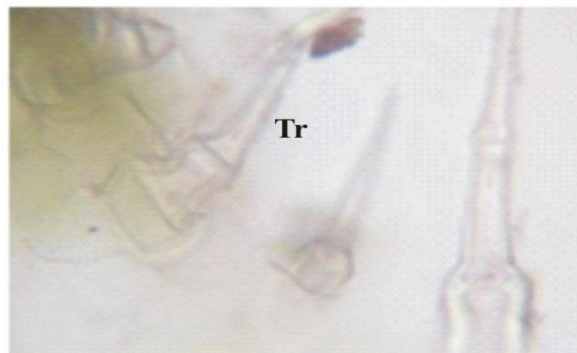
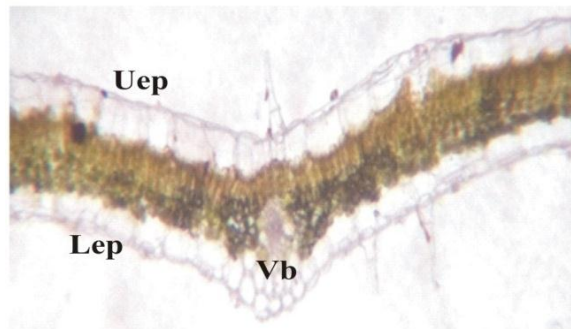
The photosynthetic pigments like chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoids content of fresh shoot and root were estimated and presented in Table 5. The pigment chlorophyll *a*, chlorophyll *b*, total chlorophyll and carotenoids content of fresh shoot;  $0.397 \pm 0.015$ ,  $0.157 \pm 0.018$ ,  $0.543 \pm 0.008$ ,  $104.86 \pm 1.98$  mg/g, and root;  $0.007 \pm 0.0007$ ,  $0.012 \pm 0.0014$ ,  $0.022 \pm 0.0014$  and  $7.833 \pm 0.540$  mg/g were respectively. The considerable amount of photosynthetic pigments to enhance the metabolic activity of *C. benghalensis* and also the carotenoids are functioned as provitamin A activity.

Fig 1. Photomicroscopy of *C. benghalensis* stem

Ep= Epidermis; Gt = Ground tissues; Vb = Vascular bundle; Xy = Xylem  
Ph = Phloem; Xv = Xylem vessels; Rh = Raphides; Sc = Sclereides

Fig 2. Photomicroscopy of *C. benghalensis* root

Rt = Root hairs; Ep = Epidermis; Gt = Ground tissues; Vb = Vascular bundles;  
 Xy = Xylem; Ph = phloem; Xv = Xylem vessels; Sp = Sieve plate; Cy = Crystals

**Fig 3. Photomicroscopy of *C. benghalensis* leaves**

Uep = Upper epidermis; Lep = Lower epidermis; Vb = Vascular bundles; St = Stomata;  
 Sp = Spongy parenchyma; Pp; Palisade parenchyma Xy = Xylem; Ph = Phloem;  
 Xv = Xylem vessels; Gc = Guard cells Ac = Accessory cells; S = Stoma  
 Xy = Xylem; Ph = phloem; Xv = Xylem vessels; Sp = Sieve plate; Cy = Crystals

**Table 1. Morphological observations of *Commelina benghalensis*. L**

Sl. no.	Morphological observation	<i>C. benghalensis</i> . L.
1.	Habit	Herb
2.	Stem type	Round with nodes and hairy
3.	Leaf type	Simple
4.	Phyllotaxy	Alternate
5.	Leaf shape	Ovate
6.	Leaf blade	Curved
7.	Leaf apex	Acute
8.	Leaf surface	Hairy (white and brown coloured hairs)
9.	Leaf base	Sheathing
10.	Leaf margin	Entire
11.	Leaf venation	Parallel
12.	Stem colour	Green
13.	Root colour	White and Brown
14.	Root flower	subterranean flowers (Cleistogamous)
15.	Flower colour	Purple blue
16.	Flower shape	Funnel shape
17.	Length of plant	20-25 cm
18.	Length of leaf	5 cm
19.	Width of leaf	3-3.5 cm
20.	Length of flower	2 cm
21.	Width of flower	1.3 cm
22.	Width of stem	0.3-0.4cm

**Table 2. Physicochemical analysis of *Commelina benghalensis* L. dried shoot and root system (n=3; means  $\pm$  SE)**

Parameter	Shoot	Root
Moisture content % (w/w)	13.047 $\pm$ 0.904	14.307 $\pm$ 0.142
Total ash % (w/w)	16.327 $\pm$ 0.163	17.987 $\pm$ 0.131
Acid insoluble ash % (w/w)	1.267 $\pm$ 0.147	7.533 $\pm$ 0.178
Water soluble ash % (w/w)	11.250 $\pm$ 0.197	10.170 $\pm$ 0.111

**Table 3. Preliminary phytochemical screening of *C. benghalensis* L. of Commeliaceae**

Phytoconstituents	Solvents					
	Benzene	Petroleum Ether	Chloroform	Acetone	Methanol	Water
Alkaloids						
I. Mayer's test	-	-	-	+	+	-
II. Wagner's test	-	-	-	-	+	-
Anthraquinones (Borntrager's test)						
Catechin	-	-	-	+	+	+
Coumarin	-	-	-	-	-	-
Flavonoids	+	-	+	+	+	+
Phenols	-	+	+	+	+	+
Quinones	-	-	-	+	+	+
Saponin (Foam test)	+	+	+	+	-	-
Steroids	-	-	-	-	+	-
Tannins	+	-	+	+	+	+
Sugar I. Benedict's test	-	-	+	+	+	+
II. Fehling's test	-	-	+	+	+	+
Glycosides						
I. Anthrone test	+	+	-	-	-	-
II. Borntrager's test	-	-	-	-	-	-
Amino acids						
Ninhydrin test	-	-	-	+	+	+
Xanthoprotein	-	-	-	-	-	-
Protein	+	-	+	+	+	+

('+' present, '-' absent)

6 **Table 4. Biochemical composition of *C. benghalensis* dried shoots and roots (n=3; means  $\pm$  SE)**

Parameter	Shoot	Root
Carbohydrate mg/g	0.582 $\pm$ 0.009	0.005 $\pm$ 0.00038
Protein mg/g	15.62 $\pm$ 0.134	1.08 $\pm$ 0.025
Amino Acid mg/g	1.191 $\pm$ 0.013	0.269 $\pm$ 0.010
Vitamins A Mg/100g	17.48 $\pm$ 0.205	13.6 $\pm$ 0.277
Total free Phenols (mg/g)	44.8 $\pm$ 0.438	12.8 $\pm$ 0.118
Tannins (mg/g)	10.7 $\pm$ 0.262	1.3 $\pm$ 0.159
Organic Carbon %	22.6 $\pm$ 0.145	15.2 $\pm$ 0.119

7  
8 **Table 5. Photosynthetic pigments of *C. benghalensis* fresh shoot and roots (n=3; means  $\pm$  SE)**

Parameter	Shoot	Root
Chlorophyll a (mg g <sup>-1</sup> )	0.397 $\pm$ 0.015	0.007 $\pm$ 0.0007
Chlorophyll b (mg g <sup>-1</sup> )	0.157 $\pm$ 0.018	0.012 $\pm$ 0.0014
Total Chlorophyll (mg g <sup>-1</sup> )	0.543 $\pm$ 0.008	0.022 $\pm$ 0.0014
Carotenoids (mg g <sup>-1</sup> )	104.866 $\pm$ 1.982	7.833 $\pm$ 0.540

9  
10 **CONCLUSION**

11 The present study could be used to standardize the bioactive substance of *C. benghalensis* L. in future.

12  
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