

## MORPHOLOGICAL AND ANATOMICAL INVESTIGATION AMONG SIX VARIANTS OF *CANNA INDICA* L.

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### Abstract

The present study explores detailed morphological and anatomical features of six variants of *Canna indica* L. Noticeable variations have been recorded in the morphology of six variants of *C. indica* especially on the basis of their different colours of leaves, flowers, staminodes and fruits. Root, stem and leaf anatomy of *C. indica* revealed little variation among the variants employed in the study. Phloem is 5-6 layered in the variant 2 (small red), 4-5 layered in the variant 4 (orange), and 3-4 layered in the remaining variants. The variant 4 (orange) can be distinguished from other five variants anatomically, by presence of 5-7 layers of schlerenchymatous tissue, and morphologically by its green pseudostem, ovate to obovate leaves, orange flowers, green bracts with maroon edges and orange-red staminodes. However, based on morphological and anatomical discrepancies the variants of *Canna indica* cannot be assigned to discrete taxonomic variety.

### Introduction

*Canna* L. (Cannaceae) is popularly known as an ornamental and medicinal plant which is cultivated extensively for its beautiful, decorative and widely varying flower colours. The commonly cultivated garden Cannas are mostly of hybrid origin, with *Canna indica* L. as a principal parent (Cronquist, 1981). It is generally native to tropical America, Mexico, the Caribbean and tropical South America, Central America and West Indies, now naturalised in many parts of eastern and south-eastern Australia. The genus *Canna* is under the Order Zingiberales which consists of 8 families (*i.e.* Zingiberaceae, Strelitziaceae, Marantaceae, Musaceae, Costaceae, Cannaceae, Heliconiaceae and Lowiaceae) with about 1,800 species (Cronquist, 1981).

*Canna* species are economically and medicinally important throughout the world. The rhizomes of *Canna* are edible and are usually consumed after boiling or cooking in various ways. The black and hard-coated seeds used as beads or made into rosaries. A decoction of root of *Canna indica* is applied for treatment of gonorrhoea and amenorrhoea. The powdered root is used as diaphoretic, diuretic, and demulcent and is administered in fevers and dropsy (de Kramer and Mass, 2008). In Java, pounded seeds are used in a poultice to relieve headache. Juice extracted from grated rhizomes is used against diarrhoea. In Hong Kong, a decoction of fresh rhizomes is prescribed in acute hepatitis. In Indo-China, crushed fresh rhizomes are applied topically for traumatic injuries in traditional medicine. In the Philippines, a decoction of the rhizomes is used as a diuretic and macerated rhizomes in water are applied to alleviate nose bleeding (Ong and Siemonsma, 1996). Leaves of *Canna indica* are applied for treating malaria and fumigated leaves are reported to have insecticidal properties (de Kamer and Maas, 2008).

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Taxonomic relevance of vegetative anatomy in delimitation of taxa and establishment of intergeneric or interspecific relationships is well reported (Tomlinson, 1961; Stace, 1965; Kotresha and Seetharam, 2000; Tschan and Denk, 2012). Foliar anatomical features play an important role in distinguishing different groups of plants. Leaf is considered as the most varied organ anatomically in angiosperm which provides a variety of anatomical features that can be employed as useful taxonomic characters (Metcalf and Chalk, 1950; Metcalfe, 1968; Stace, 1984). Taxonomically *Canna indica* is a complex and variable species as it possesses different colours of flowers. Variations among different hybrids or variants of *Canna indica* has never been studied based on morphology and anatomy in Bangladesh. Despite its ornamental, economical and medicinal importance, *Canna indica* did not receive much attention. Considering this fact the present study has been undertaken with a view to characterize *Canna indica* employing taxonomical and anatomical approaches.

## Materials and Methods

### *Plant material*

The taxonomical and anatomical studies were performed based on living materials of *Canna indica* collected from different areas of Bangladesh as well as herbarium specimens. The collected specimens were maintained under the controlled climatic condition and planted in the Botanical Garden of Jagannath University, Dhaka. These materials were supplemented by the herbarium specimens examined at Dhaka University Salar Khan Herbarium (DUSH), Jagannath University Herbarium (JnUH) and Bangladesh National Herbarium (DACB).

### *Taxonomic study*

Plant samples were collected from the Botanical Garden, Department of Botany, Jagannath University, Dhaka. The comprehensive taxonomical study of the six variants of *Canna indica* was carried out and relevant literatures were consulted (Baker, 1892; Wu and Kress, 2000; Hassan, 2007; de Kamer and Maas, 2008). Under each variant at least five specimens were investigated. The herbarium specimens deposited at DUSH and DACB were also studied critically.

### *Anatomical study*

For anatomical analysis free hand sections of stems, leaves and roots were performed following Shethi *et al.* (2017). The sections were stained in safranin and mounted in 20% glycerin. Afterwards, permanent slides were prepared from the selected sections, and studied with the help of OPTIKA microscope. Micro-photographs of the sections were taken using Euromax Camera attached with computer through Image Focus 4 software.

## Results and Discussion

### *Taxonomic treatment*

***Canna indica*** L., Sp. Pl.: 1 (1753); Baker in Hook. f., Fl. Brit. Ind. 6: 260 (1892); Prain, Beng. Pl.: 1047 (1903); Kränzlin in Engl., Das Pflanzenr. 56 (IV. 47): 1-77 (1912); Anglade, Tab. Ined. : t. 267 (1918); Stewart, Ann. Cat. Vasc. Pl. W. Pak. & Kashm.: 66 (1972); Kazmi & A. Jehan, Sultania 3: 61 (1977); Ghazanfar, Fl. Pak. 145: 1-3 (1982); Matthew, Fl. Tamilnadu Carnatic 3: 1621 (1983); Deb, Fl. Tri. State 2: 381 (1983); Ratter, J.A. Europ. Gard. Fl. 2: 130 (1984); Matthew, Suppl. III. Fl. Palni Hills : t. 1158 (1998). *Canna orientalis* Rosc., Trans. Linn. Soc. 8: 338 (1807). *Canna chinensis* Willd. in Ges. Naturf. Fr. Berl. Mag. 2: 170 (1808). *Canna indica* var. *orientalis* Rosc., Scit. Pl. : t. 12 (1824); Baker in Hook. f., Fl. Brit. Ind. 6: 260 (1892); Prain, Beng. Pl.: 1047 (1903). *Canna coccinea* Mill., Gard. Dict. ed. 8. No. 3 (1768); Balak., Fl. Jowai 2: 532 (1983). *Canna edulis* Ker-Gawler in Edw., Bot. Regist. 9: t. 775 (1824). **(Fig. 1).**

*English names:* Canna, African arrowroot, Indian canna, Purple arrowroot, Queensland arrowroot, Sierra Leon arrowroot.

*Bengali names:* Kolabati, Sarbajoya.

A long-lived, large, perennial herb, growing up to 2 m tall, spreading laterally by means of fleshy underground stems. Pseudostem erect, 1-2 m long, green or greenish maroon, cylindrical, unbranched, up to 120 × 4 cm, glabrous. Leaves simple, spirally arranged, 12.5-34.0 × 7-14 cm, greenish-maroon or green, ovate-lanceolate, acute, entire, base attenuate, pinnately veined with a distinct midrib, exstipulate, petiolate, petiole 7-11 cm long, with a short open sheath gradually passing into the petiole, upper surface glabrous. Inflorescence racemose, raceme simple, 8.5-9.0 cm long. Flowers complete, bisexual, irregular, zygomorphic, epigynous, fundamentally trimerous but with a modified androecium; bract 1, c. 11.0-11.5 × 3.0-3.5 cm, upper portion maroon-chocolate or green, lower portion maroon or light-green, ovate-lanceolate; bracteoles 3, unequal, outer one largest, c. 2.5-3.0 × 1.5-2.0 cm, other two c. 1.0-1.5 × 0.5-1.0 and 1.2-1.4 × 0.7-0.9 cm, ovate, maroon-chocolate; pedicel short, 0.2-0.3 cm long. Sepals 3, maroon, not petaloid, polysepalous, unequal, c. 1.5-1.7 × 0.7-1.0 cm, imbricate, persistent in fruits, glabrous. Petals 3, connate in a cylindrical corolla tube, lanceolate, red, equal, c. 6.0-6.5 × 1.0-1.5 cm in each flower, imbricate, glabrous. Stamens 5, 1 partially perfect, all petaloid, connate below in a cylindrical



Fig. 1. *Canna indica* L.; A. Habit; B. Flower; C. Rhizome; D. Fruit.

corolla-like tube, one with a 1-celled anther, adnate to one of its edge, this stamen rolled, c. 4.0-4.5 × 0.7-0.9 cm; anthers 1.0-1.5 × 0.1-0.2 cm, yellow, the others staminode, unequal, c. 6.2-6.6 × 1.4-1.6 cm, spathulate, emarginate to apiculate, bright red, glabrous. Carpels 3, syncarpous; ovary 3-celled, inferior, 0.5-0.6 cm in diameter, margin with soft spines, ovules many in each cell; style 1, petaloid, adnate below to the staminal tube, c. 5.4-5.5 cm long; stigma 1, flat, c. 0.5-0.7 cm in diameter, reddish-yellow, glossy. Placentation axile. Fruit a warty capsule, ovoid, c. 1.8-2.0 × 2.5-2.7 cm, maroon, outside with soft spines.

*Specimens examined:* **Chattogram:** Hazarikhil (West), 31.10.1987, A.M. Huq and M.K. Mia H. 8622 (DACB). **Dhaka:** Shahbagh, Ramna Park, 04.09.2019, Sabiha Sultana Akhi 15 (JnUH); Shahbagh, Ramna Park, 22.07.2006, Nahid Sultana 2 (DUSH); Dhaka University Botanical Garden, 20.02.1980, Mahbuba Halim 738 (DACB); Baldha Garden, 23.01.1980, Mahbuba, N. Zaman and Mia M. 469 (DACB); Ramna Park, 19.02.1980, Mahbuba Halim 678 (DACB). **Mymensingh:** Kishorgonj to Katabaira, 13.01.1979, Mahbuba Halim 364 (DACB). **Patuakhali:** Galachipa to Ulania, 12.03.1982, Rahman and Mia R. 1292 (DACB). **Sylhet:** Bahamura, 11.10.1973, Khan *et al.* K. 3225 (DACB).

### Morphological variations

Morphological variations based on size, shape and colour of pseudostem, leaves, bracts, flowers, staminodes and fruits have been found among the six variants of *Canna indica* employed in this study. Variations observed in the variants are outlined as follows: Variant 1: Pseudostem greenish-maroon; leaves obovate-ovate, greenish-maroon; flowers large red; bracts maroon; sepals maroon; petals maroon; staminodes red, c. 11-14 × 3.5-5 cm; fruits maroon (Fig. 2A). Variant 2: Pseudostem green; leaves ovate, green; flowers red and small; bracts green; sepals light green with light red tip; petals light red; staminodes bright red, c. 3.0-5.5 × 0.3-0.7 cm; fruits green (Fig. 2B). Variant 3: Pseudostem green; leaves lanceolate-ovate, green; flowers pink; bract chocolate-maroon; sepals maroon; petals maroon; staminodes pinkish-white, c. 11-14 × 3.5-6.5 cm; fruits maroon (Fig. 2C). Variant 4: Pseudostem green; leaves ovate to obovate, green; flowers orange; bracts green with maroon edges; sepals green; petals green; staminodes orange-red, one orange-red with yellow centre, c. 10-13 × 3-5 cm; fruits green (Fig. 2D). Variant 5: Pseudostem green; leaves lanceolate-ovate, green; flowers yellow; bracts green, sepals green; petals light red; staminodes yellow, c. 4.6-7.1 × 1.5-2.0 cm; fruits green (Fig. 2E). Variant 6: Pseudostem green; leaves broadly ovate-lanceolate, green; flowers yellow with red spots; bracts green with maroon tip; sepals light green-maroon; petals reddish-yellow; staminodes reddish-orange with yellow edges, c. 11.5-15.0 × 3.5-5.5 cm; fruits green (Fig. 2F).

### Anatomical investigation

#### *Root anatomy*

The epidermis of the root is composed of tabular cells with no intercellular spaces. Root hairs were also absent in this layer. In all the variants' roots, immediately beneath the epidermis a massive cortex lies consisting of thin-walled parenchyma cells with sufficiently developed intercellular spaces among them. The endodermis consists of a single layer of barrel-shaped compact cells having no intercellular spaces among them. The pericycle has been found to be uniseriate and composed of thin walled parenchymatous cells. In all the variants vascular bundles are radial, the xylem strands are found to be alternate with the phloem ones, xylem is exarch, and pith is large (Fig. 3).



Fig. 2. Six variants of *Canna indica* L.; A. Variant 1 - Local red; B. Variant 2 - Small red; C. Variant 3 - Pink; D. Variant 4 - Orange; E. Variant 5 - Yellow; F. Variant 6 - Yellow with red spots.

#### *Stem anatomy*

A single layered epidermis is found in all the six variants of *Canna indica* with a very thin layer of cuticle on the outside (Fig. 4). The epidermal cells are rectangular. No any trichome is observed on epidermis. Cortex cells are sufficiently large, 2-layered and polygonal.

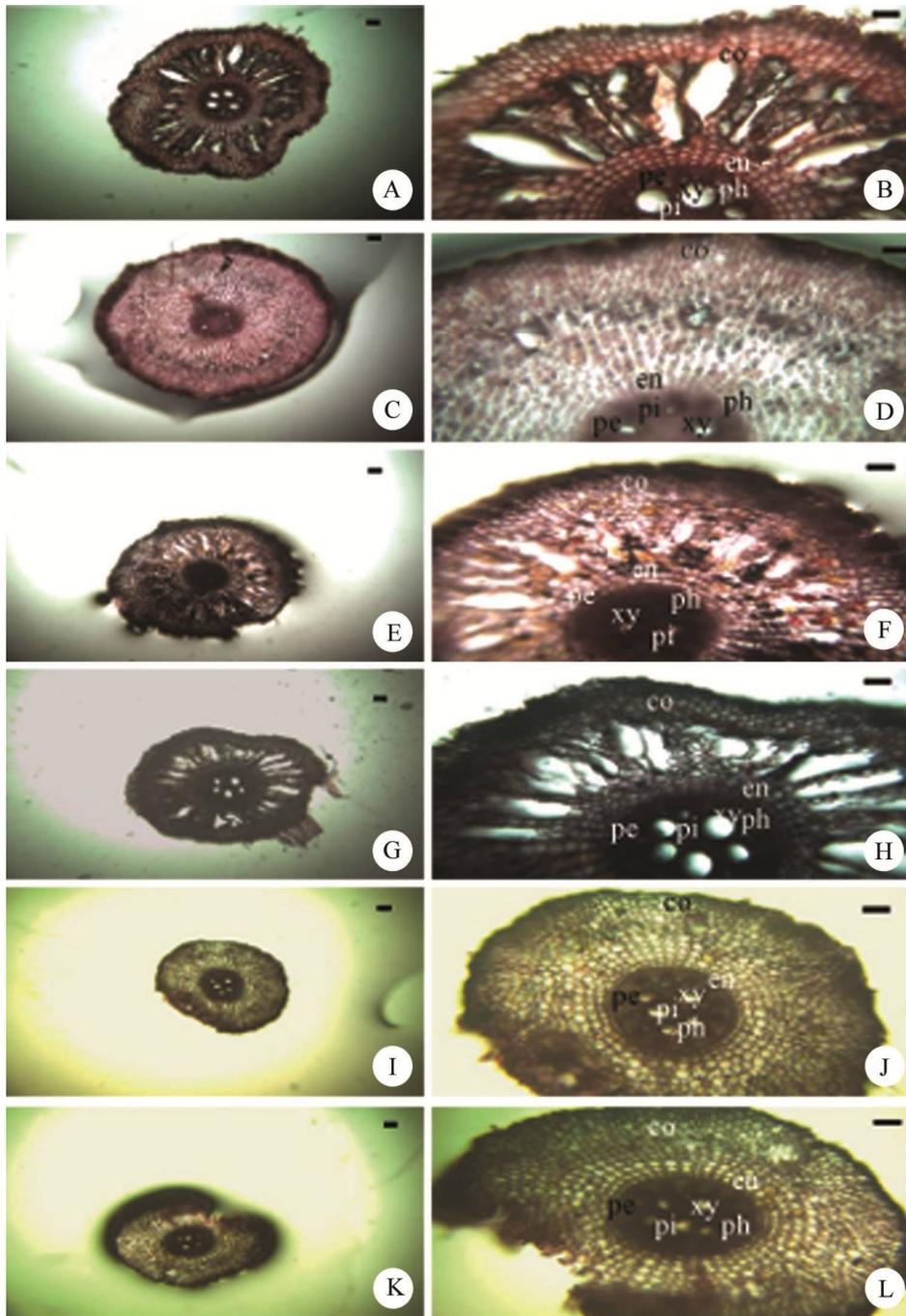


Fig. 3. Transverse section of roots of six variants of *Canna indica* L.; A-B. Variant 1 - Local red; C-D. Variant 2 - Small red; E-F. Variant 3 - Pink; G-H. Variant 4 - Orange; I-J. Variant 5 - Yellow; K-L. Variant 6 - Yellow with red spots. ep: epidermis, co: cortex, pe: pericycle, en: endodermis, ph: phloem, xy: xylem, pi: pith. (Bar = 100  $\mu$ m; A,C,E,G,I&K = 4X; B,D,F,H,J&L = 10X).

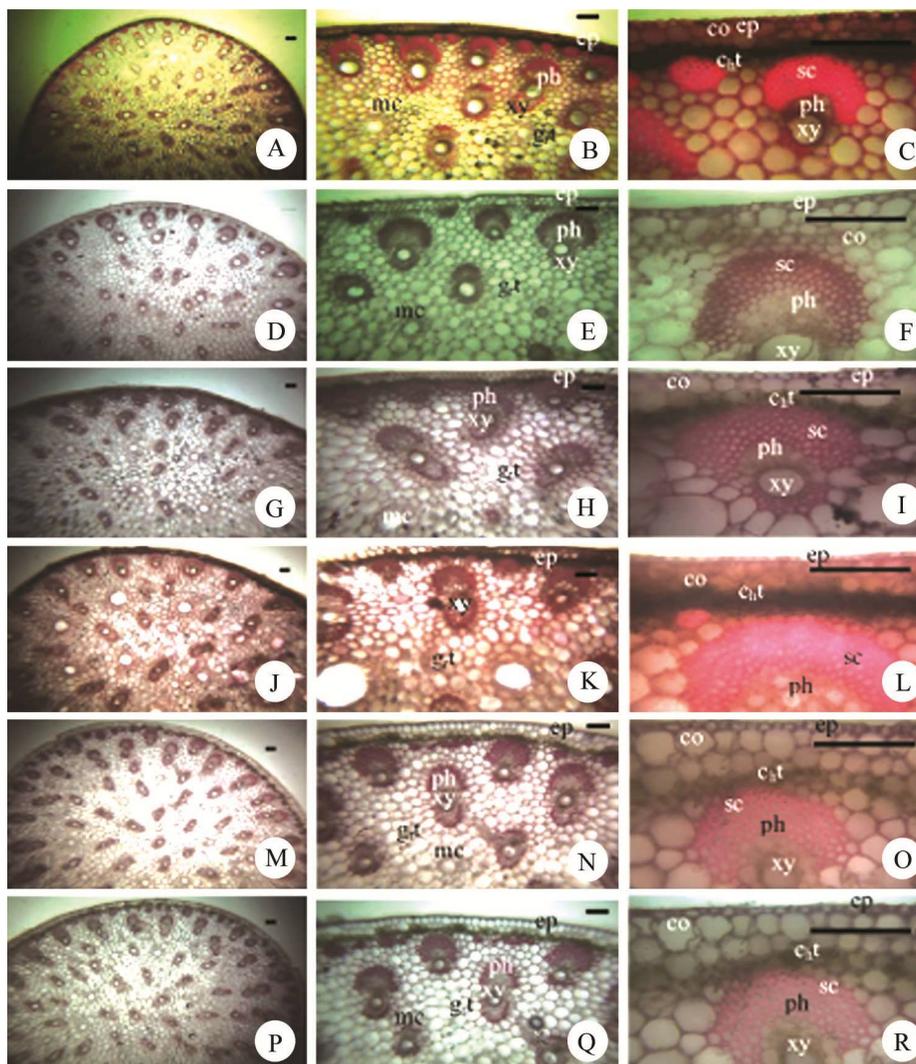


Fig. 4. Transverse section of stem of six variants of *Canna indica* L.; A-C. Variant 1 - Local red; D-F. Variant 2 - Small red; G-I. Variant 3 - Pink; J-L. Variant 4 - Orange; M-O. Variant 5 - Yellow; P-R. Variant 6 - Yellow with red spots. ep: epidermis, co: cortex, c,t: chlorophyllous tissue, g,t: ground tissue, sc: sclerenchymatous cells, mc: mucilage canals, ph: phloem, xy: xylem. (Bar = 100  $\mu$ m; A,D,G,J,M&P = 4X; B,E,H,K,N&Q = 10X; C,F,I,L,O&R = 40X).

A single layered chlorenchyma is found beneath the cortex consisting of chloroplast bearing cells. The sclerenchyma patches with 5-7 layers also remain attached to the chlorenchyma tissue. In the variant 4 of *Canna indica* (orange), the sclerenchyma is 5-7 layered but the remaining five variants contain 6-7 layers of sclerenchyma. The variant 2 of *C. indica* (small red) has 5-6 layered phloem and the variant 4 of *C. indica* (orange) contains 4-5 layers of phloem, and other four variants have 3-4 layers of phloem (Table 1). The remaining consists of large thin-walled, parenchymatous cells with developed intercellular spaces (ground tissue) cannot be divided into distinct regions. All the six variants possess numerous wide mucilage canals.

**Table 1. Stem anatomical characters of six variants of *Canna indica* L.**

Name of the variants	Trichome	Cortex	Sclerenchyma	Phloem
1. <i>Canna indica</i> L. (local red)	Absent	2-layered	6-7 layered	3-4 layered
2. <i>C. indica</i> L. (small red)	Absent	2-layered	6-7 layered	5-6 layered
3. <i>C. indica</i> L. (pink)	Absent	2-layered	6-7 layered	3-4 layered
4. <i>C. indica</i> L. (orange)	Absent	2-layered	5-7 layered	4-5 layered
5. <i>C. indica</i> L. (yellow)	Absent	2-layered	6-7 layered	3-4 layered
6. <i>C. indica</i> L. (yellow with red spots)	Absent	2-layered	6-7 layered	3-4 layered

**Leaf anatomy**

Leaf anatomy of the six variants of *Canna indica* revealed upper and lower epidermis with thin cuticle made up of a single layer of thin walled cells (Fig. 5). Both upper and lower epidermal cells are rectangular or cubic in shape. The stomata found on both surfaces are almost equal in number. In all the variants the mesophyll is not differentiated into palisade and spongy parenchyma. All the mesophyll cells are nearly isodiametric and thin-walled as the leaf is isobilateral, and they are compactly arranged with limited intercellular spaces containing numerous chloroplast. Trichome is absent in all the variants.

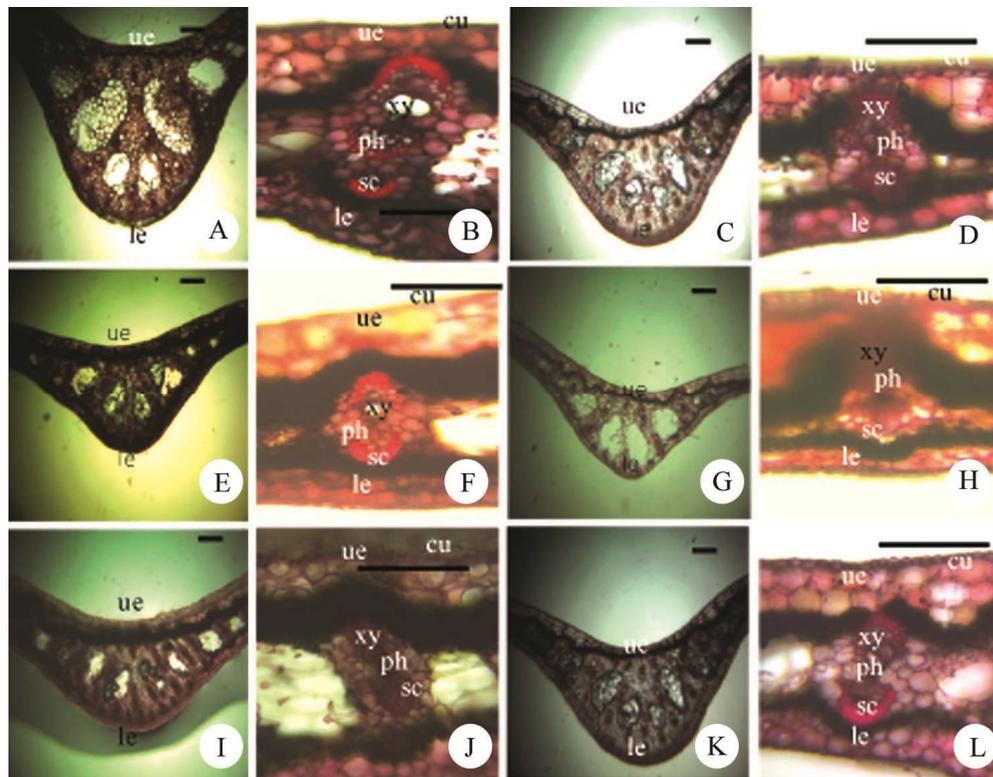


Fig. 5. Transverse section of leaf of six variants of *Canna indica* L.; A-B. Variant 1 - Local red; C-D. Variant 2 - Small red; E-F. Variant 3 - Pink; G-H. Variant 4 - Orange; I-J. Variant 5 - Yellow; K-L. Variant 6 - Yellow with red spots. ue: upper epidermis, cu: cuticle, sc: sclerenchymatous cells, ph: phloem, xy: xylem, le: lower epidermis. (Bar = 100  $\mu$ m; A,C,E,G,I&K = 10X; B,D,F,H,J&L = 40X).

In the bundle sheath of all variants sclerenchyma cells are observed. Vascular bundles are embedded in mesophyll and are of rectangular type. In the investigated variants, xylem faces upper surface as phloem faces the lower surface (Fig. 6). In all the studied variants of *Canna indica* stomata is surrounded by two subsidiary cells which are parallel to the long axis of the pore and guard cells, and the stomata are of Paracytic or Rubiaceous type (Fig. 7).

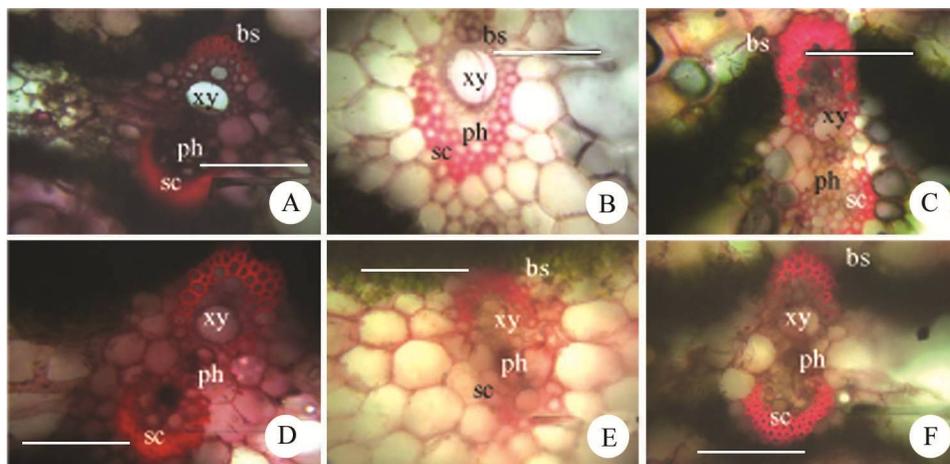


Fig. 6. Middle vascular bundles of six variants of *Canna indica* L.; A. Variant 1 - Local red; B. Variant 2 - Small red; C. Variant 3 - Pink; D. Variant 4 - Orange; E. Variant 5 - Yellow; F. Variant 6 - Yellow with red spots. bs: bundle sheath, sc: sclerenchyma, ph: phloem, xy: xylem. (Bar = 100  $\mu$ m at 40X).

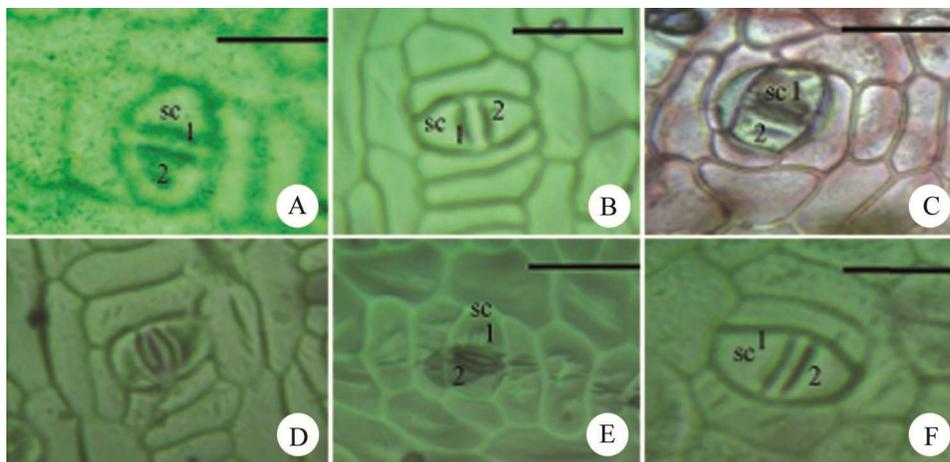


Fig. 7. Types of stomata in six variants of *Canna indica* L.. A. Variant 1 - Local red; B. Variant 2 - Small red; C. Variant 3 - Pink; D. Variant 4 - Orange; E. Variant 5 - Yellow; F. Variant 6 - Yellow with red spots. sc: subsidiary cells. (Bar = 100  $\mu$ m at 40X).

### Discussion

In the present investigation, detailed taxonomic account of *Canna indica* has been provided. Based on variations in colours of different flowers as well as other characters *Canna indica* has been described under six variants. Aiton (1789) recognized 4 varieties of *Canna indica* on the

basis of leaf and flower characters. Eichler (1875) stated that 3 sepals of *Canna indica* are slightly unequal, mostly glaucous green, erect, narrowly obovate, narrowly triangular, acute to obtuse and persistent. Roscoe (1826) pointed out that the 3 petals of *Canna indica* are mostly erect and narrowly ovate-triangular with acute to acuminate apex. They are basally connate and unequal, one being always smaller than the other two, and the aestivation is imbricate. Roscoe (1807) considered the construction of the stamen to be the main character. In the present study, all 5 stamens are petaloid and connate below in a cylindrical corolla-like tube, one with a 1-celled anther adnate to one of its edge ( $4.0 \times 0.7$  cm), yellow in colour, the other staminodes unequal in size. According to Kirchoff (1983) and Kress (1990), the androecium of *Canna indica* is composed of 2 essentially trimerous whorl, the outer whorl being episealous and the inner one epipetalous. The style is petaloid, firm and fleshy and for most of its length connate with the stamen. The styler canal forms a 3-pointed star in cross section (Kunze, 1984). The ovary in *Canna* is inferior and 3-locular with axillary placentation and each locule contains 2 alternating rows of ovules; 3-sepals nectarises are filling the upper part of the ovary and opening at the top, the lower part of the ovary being the fertile part (Vogel, 1969). The present study has been found concordant with those of previous studies. We have found that fruits are warty capsule, ovoid, seeds are numerous, round, up to 8 mm in diameter, black and hard. Grootjen and Bouman (1988) reported the mature fruit of *Canna indica* as a dry capsule crowned by persistent sepals, and mature seeds are numerous, shiny brown to black,  $3.5-10.0 \times 2-8$  mm, globose to narrowly ellipsoid which is supported by the present study. The study has revealed noticeable variation among the six variants of *Canna indica*, more particularly in colour of leaves, flowers, fruits, staminodes and the size of staminodes per flower, though all of the variants show affinity in terms of pseudostem and leaf shape.

Anatomical investigation in six variants of *Canna indica* provided little variation. Previous studies on some other monocotyledonous plants showed that root cortex contained dense paranchymatous cells (Bibi *et al.*, 2014). Solereder and Meyer (1930) reported compact peripheral layers of small, hexagonal cells in root cortex of *C. indica*. Inner cortex is very uniform and cells are radially arranged including very uniform intercellular spaces. Endodermis is slightly U-shaped, and is uniseriate in young roots, while the pericycle is single layered. Very recently, Gayatri *et al.* (2018) observed parenchymatous cortex with intercellular spaces in the root of a monocotyledonous *Triticum aestivum* which is in agreement with our investigation as observed in *Canna indica*. Tomlinson (1961) found that epidermis of stem of *Canna indica* is thinly cutinized, cells are rectangular to elongated, and walls are slightly thickened. Chlorenchyma present as 1-2 layers of small cells separated from the epidermis by 1-2 layers of colourless hypodermal cells. The sclerenchyma patches also remain attached to the chlorenchyma. Vascular bundles are irregularly scattered, without sheathing fibres but sometimes with a little thick-walled prosenchyma adjacent to xylem and phloem. Mucilage canals apparently restricted to the periphery of the central cylinder are wider and more irregular (Solereder and Meyer, 1930).

Leaf epidermis of *Canna indica* contains a very thin cuticle, and the lower epidermis is found to be somewhat irregular with many frequent narrow, costal bands than that of upper epidermis, and the cells of both upper and lower epidermis are rectangular (Tomlinson, 1961), and this is supported by the present study. A single layered hypodermis has been observed below the epidermis. Upper hypodermal cells serve as expansion cells (Löv, 1926). The leaf contains two layers of pallisade parenchyma and spongy parenchyma, and characteristically oblique cells are present with round ends obliquely to the long axis. However, in our experiment we found that mesophyll was not differentiated into pallisade and spongy parenchyma. Stomata is occasionally found in the upper surface of leaves, and the type of stomata remains uncertain (Tomlinson, 1961). Most of the anatomical features of *Canna indica* as revealed from this investigation have been

found consistent with those of earlier studies (Solereeder and Meyer, 1930; Tomlinson, 1961; Bibi *et al.*, 2014).

In conclusion, noticeable morphological as well as anatomical variations have been observed among the six variants of *Canna indica*. The variant 2 (small red) and variant 4 (orange) can be differentiated on the basis of schlerenchyma and phloem characters. However, these variations are not sufficient enough to distinguish all the variants employed in the study. Based on morphological and anatomical variations as revealed from the present study the variants of *Canna indica* cannot be assigned to distinct taxonomic variety. Therefore, more studies employing additional tools, *viz.* cytological, palynological and molecular approaches could be undertaken for better understanding of systematics of *Canna indica* and its different forms.

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