



Research article

Morphological and anatomical characterization of two species of *Crinum* L.

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Abstract: *Crinum*, a genus under Amaryllidaceae, is having around 180 species all over the world, and are mainly distributed in the tropical and subtropical region of the world. Taxonomical treatment of Indian species of *Crinum* was done by William Roxburgh and incorporated in the Flora of India. The perennial geophytes of *Crinum* spp. generally show morphological diversities in respect of the underground and above-ground parts of them. The different *Crinum* species have been used in different parts of the world to treat various health problems in the form of tonics, laxatives and expectorants, kidney pains and in urinary troubles etc. and some of such biomolecules are used to treat various health problems in the form of tonics, laxatives and expectorants, kidney pains and in urinary troubles etc. The present study is aimed at characterizing two species of the genus concerning the details of morphological and anatomical features and to contrast them to delimit the species more vividly. Two species under study are *Crinum asiaticum* and *Crinum latifolium* morphological characterization of the species has been carried out in terms of the general habits of each species, features of bulb, leaf, inflorescence and floral parts. The anatomical features have dealt with the sectional view of the bulb and lamina. The phenology of these two species has been determined by studying the individuals from different locations at least thrice in a month. The provenances have been noted to differ considerably in their morphological details e.g. gross habit, leaf length, width, margin, floral characters. Plant height, leaf length and margin, the appearance of bulbs, floral morphology along with the anatomical features of leaves and roots has been presented here. Phenological variation has also been noted at the intraspecific level, even when the plants from different localities are grown in the same spot under the same environment, a fact pointing to the inherent difference in the individuals in this regard. Along with the understanding of the range of characters for each species, the study highlights the interspecific diversities for the two species under study.

Keywords: *Crinum asiaticum* - *Crinum latifolium* - Diversity - Taxonomy.

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INTRODUCTION

Crinum L. is a pan tropical genus belonging to the family Amaryllidaceae with 180 species. Originally, the genus is originated from two main centers - South America and South Africa. In India, only 13 species, 1 variety and 1 form have been reported so far and among them, 5 are endemic to India (Govaerts *et al.* 2012).

The plants are geophytes due to their propagation by means of bulb or rhizome (Bjora *et al.* 2009). The *Crinum* species have importance as sources of medicinally useful biomolecules. Many species of the genus have ornamental value for their large showy and attractive flowers. A large number of natural products like alkaloids, phenol compounds, flavonoids, glycosides etc. have so far been recorded in these plants. Different species of the genus have not only been used in pharmaceutical industries, but also extensively as folk herbal medicines for the remedy of various diseases in several countries. Plant anatomy played a very important role in identifying proper species to avoid adulterants and also assure the quality of drugs (Mandal & Nandi 2012). The utility of morpho-anatomical studies of medicinal plants for the quality control of the drugs has been proved through the

works of earlier workers (Banerjee & Mookherjee 2001, Gupta *et al.* 2001). World Health Organization has also emphasized the microscopic and macroscopic studies of medicinal plants as an important measure for the proper identification and assurance of the quality of crude drugs (Anonymous 1996). The present study displays the details of morphological and anatomical features of two different species of *Crinum* L. In this study an account of different morphological, phenological and anatomical features of both the species has been carried out for their ready and easy recognition, which may help in proper identification of the whole plant in the field as well as the supplied plant parts in use in laboratories.

MATERIALS AND METHODS

Material

The species *Crinum asiaticum* L. and *Crinum latifolium* L. both have been collected from Midnapore forest range of West Bengal.

Name of Species	Location	Latitude (°N)	Longitude(°E)
<i>Crinum asiaticum</i> L.	Midnapore forest area of West Bengal	22.430889	87.321491
<i>Crinum latifolium</i> L.	Midnapore forest area of West Bengal	22.430889	87.321491

Morphological study

External morphological studies of both the species of even-aged plants were carried out taking into account plant height, phyllotaxy, leaf length and width, leaf margin, the appearance of bulbs, basal part of bulbs, end part of bulbs, inflorescence type, flower colour, perianth, stamens, carpels, fruits and seeds.

Anatomical study

Anatomy of the roots and the lamina of both species were worked out with their respective transverse sections (T. S.) and observed under the *Leica DM 1000* microscope. These studies incorporated the type of stomata, nature of the vascular bundle, number of xylem strand etc. Plants of equal ages were taken to avoid the chance of different impacts on the features due to unequal age. The tissue sections were mounted in glycerol (10%) and photographs were taken under the microscope.

Phenological study

The phenological survey was conducted during the year 2018–19 in both the species from the districts of Paschim Medinipur of West Bengal. The phenology of two medicinal and ornamental plants *Crinum asiaticum* L. and *Crinum latifolium* L. had been observed in three widely separated study sites following standard protocols (Fig. 1). Phenological observation had been at least thrice in a month by visiting the respective field from the above-mentioned study areas.

The periods of starting of flowering were measured when the inflorescences were completely open for 3 spots and blossomed flowers were observed from at least 3 of same spots. Blossomed flowers have been considered to those where petals have been completely opened. The leaf unfolding was considered as unfolded herb with more than 3 spots looking like leaves and the fall foliage was considered with more than 3 foliage spots on the total leaves. The start of leaf falling was when fallen leaves were observed from 3 spots. All the data were recorded as a percentage. The growth started when the buds were observed above the soil surface and the unfolding started when leaves completely open was observed for the leaf unfolding period. The fallen blossom was considered when 90–100% of leaves were withered or dead.

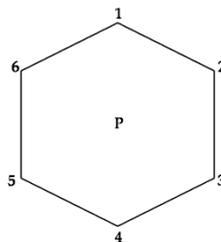


Figure 1. Phenological clock. [A phenological clock with different phenological stages were shown where number 1 is to indicate sprouting of buds in rhizomes; 2. Vegetative phase; 3. Flowering phase; 4. Fruiting phase; 5. Seed maturation; 6. dyeing or death of species and P-perennation for phenological observation]

Statistical analysis

The numerical data were statistically analyzed with SPSS software. The level of significance used in F test was $P = 0.05$.

RESULTS AND DISCUSSION

The study showed worthwhile differences in morphology and anatomy between the species in their height, the appearance of bulb, leaf length, leaf margin, perianth and androecium and in anatomy in regard of number of the vascular bundle of leaf, layers of the lower epidermis, number of xylem strand in roots, size of the cortex and stele etc. Some basic characters common for the species taken here are as follows. Leaves are lanceolate or oblanceolate, arranged in a whorl from the bulb and true stem is absent. Flowers are bisexual, white and with six lobed perianths. Stamens are six and gynoecium is with 3 carpels and 3 lobed inferior. Ovary is syncarpous with axile placentation with multiple ovules. Fruits are three-lobed capsules. Seeds are flattened and blackish. The findings of phenological observations have been presented in table 1.

Table 1. Phenological clock shows appearance of different features of both the species round the year.

Name of species	Jan	Feb	Mar	Apr	May	June	July	Aug	Sep	Oct	Nov	Dec
<i>Crinum asiaticum</i> L.												
<i>Crinum latifolium</i> L.												

While the gross morphological resemblance of two species, that makes confusion in easy recognition of these plants under study, scrutiny of details morphological, phenological and anatomical features help discriminate them. They have shown differences in gross morphology like plant height, plant growth diameter, bulb shape, scape height, number of flower per scape, flower colour, type of perianth, shape of perianth, venation of tepal, aestivation type, length of anther, length of filament and seed colour (Figs. 2 & 3; Table 2).



Figure 2. Whole plant: A, *Crinum asiaticum* L.; B, *Crinum latifolium* L.



Figure 3. Flowera: A, *Crinum asiaticum* L.; B, *Crinum latifolium* L.

Table 2. Morphological features of *Crinum asiaticum* L. and *Crinum latifolium* L.

Parmeter	<i>Crinum asiaticum</i> L.	<i>Crinum latifolium</i> L.
Habit	Leafy herb	Leafy herb
Habitat	Terrestrial	Terrestrial
Pl. height (cm)	197.00±12.97	84.40±08.75
Plant growth diameter(cm)	193.60±09.70	168.88±06.28
Phyllotaxy	Spiral	Spiral
Shape of bulb	Ovate	Spherical
Neck of bulb	Large rounded neck	Neck absent
Bulb length (cm)	18.11±00.65	06.00±00.11
Bulb diameter (cm)	05.10±00.24	18.97±00.21
Leaf length (cm)	95.28±05.50	84.56±08.79
Leaf width (cm)	11.31±00.87	02.10±00.15
Leaf margin	Plane	Plane
Inflorescence	Umbel	Umbel
Scape height (cm)	55.00±05.24	37.04±04.48
Scape colour	Greenish	Green
Scape diameter (cm)	02.18±00.24	01.10±00.08
Number of flower per scape	21.64±01.60	04.40±00.76
No. of bract	02.24±00.59	02.24±00.59
Length of bract (cm)	08.40±00.50	05.56±00.13
No. of bracteole	02.24±00.59	02.24±00.59
Length of bracteole (cm)	07.97±00.57	03.55±00.11
Flower colour	White	Red tinted white
Perianth type	Salver- shaped	Funnel shaped
Shape of perianth	Tube-linear	Lance-ovate
Length of petiole (cm)	01.01±00.07	06.23±00.32
Width of petiole (cm)	00.76±00.10	00.67±00.06
Length of tepal (cm)	12.54±00.87	11.68±00.35
Venation of tepal	07.84±00.55	17.92±06.81
Aestivation	Imbricate	Quincuntial
Androecium	Epiphyllous	Epiphyllous
No. of stamen	06.00±00.00	06.00±00.00
Colour of stamen	Yellow	Yellow
Type of anther	Bilocular	Bilocular
Length of anther (cm)	05.90±00.28	00.59±00.09
Length of filament (cm)	01.90±00.23	08.40±05.13
Attachment of anther	Dorsifixed	Dorsifixed
Gynoecium	Syncarpous	Syncarpous
Length of style (cm)	18.14±00.49	11.70±00.34
Length of stigma (cm)	00.28±00.04	00.30±00.03
Placentation	Axile	Axile
Type of ovary	Inferior	Inferior
Length of ovary (cm)	02.00±00.14	01.50±00.02
Width of ovary (cm)	01.40±00.08	00.60±00.01
Fruit	Capsule	Capsule
Seed	Endosperm fleashy	Albuminous
Seed colour	Grey	Black
Seed Shape	Uneven rounded	Uneven

Plants of these two species can be identified right on the field based on their respective floral morphology. Plant morphology and anatomy play very important role to identify the right species in their live forms and also the parts thereof, used as raw materials for drugs, as effective in other species (Mandal & Nandi 2012). Previous workers have also revealed the morphological differentiation among the various species of this genus (Arroyo & Cutler 1984, Fangan & Nordal 1993, Snijman & Linder 1996, Meerow & Snijman 2001, Kwembeya & Stedje

2007, Bjora *et al.* 2009). The significance of anatomical information in identifying plant species as claimed by earlier workers Eltahir & AbuReish (2010), Kakrani *et al.* (2011), Mandal & Nandi (2013) has been proved to be true for the species studied here. Some anatomical parameters of both leaves and roots of the studied species have been noted to be effective in delimiting them. Leaf anatomy reveals differences only in the shape and number of the vascular bundle of both species (Fig. 4; Table 3).

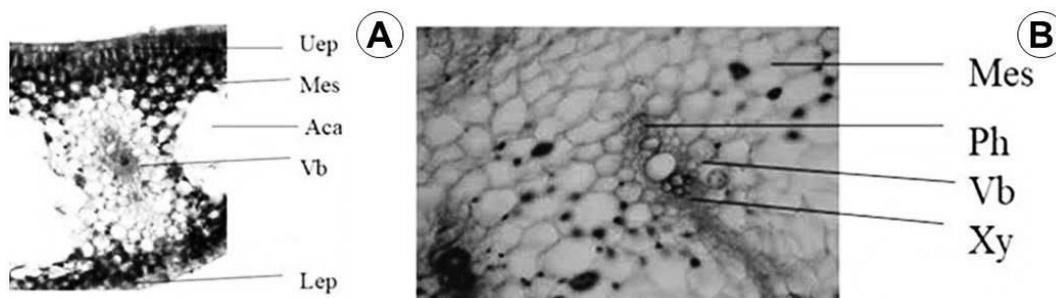


Figure 4. Transverse section of Leaf: **A**, *Crinum asiaticum* L.; **B**, *Crinum latifolium* L. [Uep = Upper epidermis, Mes = Mesophyll tissue, Aca = Air cavity, Vb = Vascular bundle, Lep = Lower epidermis]

Table 3. Leaf anatomical features of *Crinum asiaticum* L. and *Crinum latifolium* L.

Parameter	<i>Crinum asiaticum</i> L.	<i>Crinum latifolium</i> L.
Stomata	Diacytic	Diacytic
Vascular bundle type	Closed collateral	Closed collateral
Vascular bundle Shape	Ovoid	Elliptical
No. of vascular bundle	17.92±00.49	35.48±01.32
No. upper epidermal cells	01.00±00.00	01.00±00.00
Shape of upper epidermal layer	Even	Even

Root anatomy of both species has been found to slightly differ in respect of the number of epidermis cell layers, size of cortex, number of xylem strand and size of stele (Fig. 5; Table 4). The details of anatomical features have not revealed previously for both of these plants. The worked out differences in various traits of these congeneric species of *Crinum* will enable sorting out the right species to meet the need of specific use.

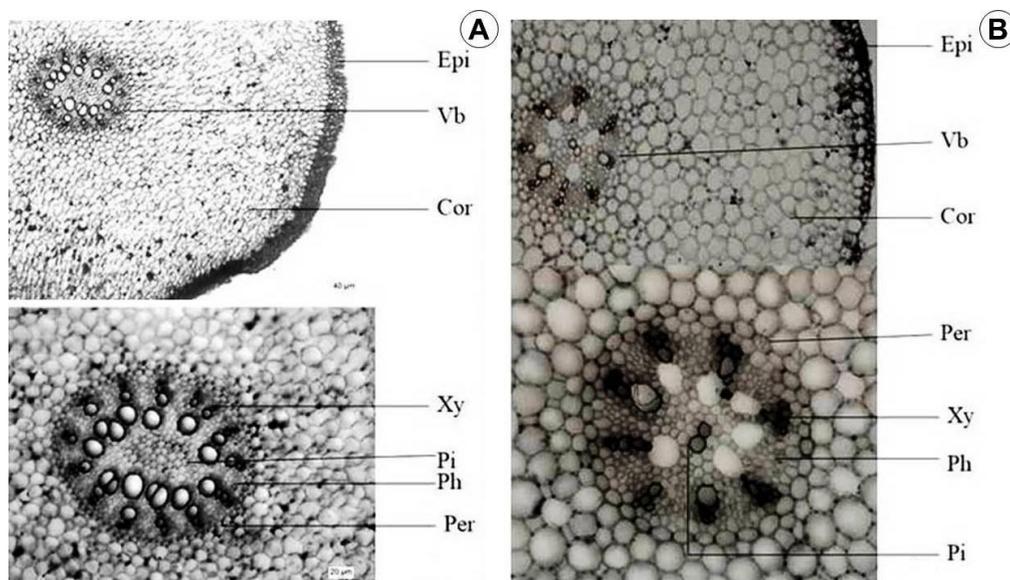


Figure 5. Transverse section of root: **A**, *Crinum asiaticum* L.; **B**, *Crinum latifolium* L. [Epi = Epidermis, Vb = Vascular bundle, Cor = Cortex, Xy = Xylem, Ph = Phloem, Per = Pericycle layer]

Table 4. Root anatomical features of *Crinum asiaticum* L. and *Crinum latifolium* L.

Parameter	<i>Crinum asiaticum</i> L.	<i>Crinum latifolium</i> L.
Type of vascular bundle	Radial polyarch	Radial polyarch
No. of epidermis cells	02.00±00.00	01.00±00.00
Cortex region (µm)	94.96±00.93	59.44±01.93
No. of pericycle layer	01.00±00.00	01.00±00.00
No. of xylem strand	12.88±00.33	08.00±00.00
Size of pith cells	Uniform	Uniform
Diameter of stele (µm)	60.00±02.80	27.92±01.73

CONCLUSION

The striking resemblance in the outward appearance of *Crinum asiaticum* and *Crinum latifolium* poses a problem in discriminating them readily. To overcome the problem of right identification for both the species in live as well as dry plant parts, some morphological features like bulb appearance, floral morphology and in anatomical characteristics of leaf and root have been found to be very useful and effective for choosing the right species (Table 2–4). This is also helpful to discriminate the proper plants from the adulterants.

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