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Anatomical Protocol of *Begonia dipetala* Graham for the Specific Identity of the Plant

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Abstract The botanical identity of *Begonia dipetala* Graham remained controversial, since its time of discovery in 1826. It was considered to be synonymous with *B.malabarica* Lamarck which is endemic to Malabar of Kerala India. Irmcher (1960) declared that *D.dipetala* Graham is an independent taxon. Santhosh *et al.(*2005) provided morphological features of *B.dipetala* and confirmed its identity. The present study deals with the anatomical parameters of leaf, petiole, internode, rhizome and root of *B.dipetala*. The morphological features of flowers and leaves are described briefly. The results of the study are believed to supplement the external features and to circumscribe the taxonomic status of *B.dipetala*. The present investigation aims at comparison with those of other members of Begoniaceae published by Solereder (1908) and Metcalfe and chalk (1957). Significant differences were observed between *B.dipetala* and other species of *Begonia*. It is suggested that *B.dipetala* can be treated as an independent species.

Keywords *Begonia dipetala* Graham, *B. Malabarica* Lamarck, Identitiy, Anatomical Parameters

1. Introduction

Begonia of Begoniaceae includes **900** species at global level[1]. In India there are 45 species[2]. In Tamil Nadu eight species have been recorded[2]. In terms of number of species, *Begonia* may be an insignificant taxon; when its economic values are considered, it is of high horticultural importance. The species of *Begonia* bear foliage-leaves and flowers which are ethereal in architecture and will captivate any casual observer. Furthermore, certain species of *Begonia*, especially *B. malabarica*, are credited with medicinal properties[3]. The ethnic communities of Western Ghats of South India, attribute many pharmaceutical efficacies to *Begonia*.

B.dipetala Graham is an archaic species and its history of discovery dates back to early part of nineteenth century. It was first discovered in the Western Ghats of Bombay, India in 1826 by Johnstone[4] and was described by Graham in 1828. In 1859,Thwaites[5] described a new variety, *D. malabarica var dipetala,* Clarke (1879) continued to use *B.dipetala* as a variety of *B.malabarica*. Based on the examination of herbarium specimens of *B.dipetala,* de Candolle[6] described it as *B.malabarica*, which was considered to be an incorrect identification. Irmcher[7] proved that *B.dipetala* Graham is clearly distinct from *B.malabarica* Lamarck and therefore, cannot be a variety of it. This tenet was further strengthened by the findings of Santhosh *et al.*[8] who provided the taxonomic circumscription of *B.malabarica* collected from Kerala, India.

The present study focuses on the microscopic features of different organs of *B.dipetala* to expand the diagnostic dimensions of the taxon and to supplement the external characters with the internal features to confirm the identity.

2. Literature Review

Anatomical studies on *Begonia* in general, and *B.dipetala* in particular are very much lacking, barring a few fragmentary studies.

Rose and Hurd-Kareer[9] recognised two physiological ly different types of cells, sharply differentiated when the leaf sections were put in the indicator solutions for about 15 minutes. They observed that most of the cells lose their high acidity and the colour of the cells changed towards the alkaline form of the die. Other cells called "Specialised Cells" retained their acidity and the acid colour of the cells remained indefinitely. These specialised cells contain calcium oxalate crystals of druses. The authors confirmed the physiological differences between the two types of leaf, by immersing the sections in solution by the oxidase reagents benzidine and gum guaiacum; the crystal containing cells stain blue while, other cells do not stain.

Watson and **Dallwitz**[10] have given a brief account of anatomy of leaf and stem apart from external features of the species of Begoniaceae. The authors reported "pearl –

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glands", hydathodes and stomata confined to the lower surface and the stomatal types varied from anisocytic, paracytic, diacytic to cyclocytic with 3 – 6 subsidiary cells arranged in two rings. They have also mentioned that presence of one or more hypodermal layers on the adaxial side of the lamina. Cystoliths and sclerenchymatous idioblasts are present or absent; minor veins do not have phloem elements. The stem has superficial cork-cambium; the nodes are trilacunar and pentalacunar, cortical bundles and medullary bundles are present or absent.

A brief account on the structure of the midrib of *Begonia elatior x hybriden* was given by Anisoara Stratu *et al.*[11]. These authors observed that the lamina between the veins is thin. The mesophyll of the lamina has bifacial heterofacial structure. The adaxidal epidermal cells are tangentially elongated and thick. The midrib wall is raised on the adaxial side. In 1830, **Hooker[**12] described the characteristics epidermal hairs, glands and structure of stem of *Begonia longipes*.

Hildebrand[13] and **Fellerer[**14] published an outstandi ng work on the systematic anatomy of *Begonia*. These authors considered mainly the cystoliths and cystosphere formation as systematic characteristics for the analysis of *Begonia*. They contemplated that these characters were proof for relationships with Cucurbitaceae.

Haberlandt[15] described the sclerenchyma distribution in the leaves of *B.nelumbifolia, B.pustulata* and *B.violifolia*. **Hailler[**16] proposed phylogenetic relationships of the anchor-hairs of *Begonia* species with the hairs in the compositae member namely *Hypochoeris aeltinensis*. Pneumatothodes have been described by **Vouk[**17] in stem of *B.vitifolia*, which resemble and replace typical lenticels. **Fotsch[**18] provided detailed account of anatomical aspect of *Begonia*.

Fellerer[14] and **Boghdan**[19] studied and described multicellular nonglandular and captitate and noncapitates trichomes of *Begonia*. Multiple epidermis has been reported in *Begonia* by several workers (Boghdan and Barkely,[20]; Barkley and Hozid,[21]; Boghdan,[22]. The tribals, Paliyans and Pulayans of Palani Hills of Tamil Nadu seem to use the paste of the stem of *B.malabarica* to cure the pimples and for cooling effects (Ganesan *et al.*,[23]).

Santhosh *et al.*[8] traced the history of the discovery of *Begonia* in India, starting from the period of Rheede[24]. The authors elucidated the identity of Rheede's *Tsjeria-narinampuli* as *Begonia malabarica* Lamk. **Clarke**[25] while studying Begoniaceae of British India, included *B.malabarica* with two more varieties viz; *var.dipetala* and *var.hydrophila*. **Gamble**[26] followed **Clarke[**25] and described the species occurring in Madras Presidency as *B.malabarica*. After elaborate discussion of various reports on *Begonia* by different investigators, the authors proposed a dichotomous key to distinguish *B.dipetala*, *D.dipetala var. hydrophila, B.malabarica* and *B.fallax*. In *B.dipetala* the male and female flowers consistently have 2 perianth lobes; the fruits are attenuated at the apex; the wings are rounded obtuse at the base and wings are rounded along the margins. *B.dipetala var hydrophila* is characterized by obovate – rounded fruit which is obtuse at the base and the wings are obtuse or sub angulate.

Begonia malabarica differs from *Begonia dipetala* in having 4 perianth lobes in male flowers and 3 perianth lobes in the female flower; the perianth lobes and the capsule are pilose on the outer sides (Santhosh *et al.*,[8]).

Rajbhandary *et al.*[27] traced the origin and dispersal of Asian. According to the concept of the authors the genus *Begonia* began to diverge in Africa during the Oligocene and the current hotspot of diversity for the genus in China and South East Asia must therefore be the result of an eastward dispersal of or migration across the Asian continent. The authors considered the role of Himalayas as a mesic corridor facilitating this migration and constructed a time calibrated molecular phylogeny using ITS sequence data. According to the investigators the Himalayan species of *Begonia* fall under two groups: 1, an unresolved grade of tuberous, deciduous species of unknown geographic origin, with evidence of endemic radiations in the Himalayan region beginning c.7.4 Ma coinciding with the outset of the Asian monsoon. 2, a group of evergreen rhizomatous species with a probable origin in China, which has migrated to the Himalayan region c.5.1 Ma coinciding with an intensification of the monsoon.

Anisoara *et al.*[11] studied certain decorative flowering plants with respect to the physiological and anatomical aspects. The study includes *Begonia elatior X hybriden*. Their results showed that water content in flower buds and flowers is highest, low values in the leaf; cell juice concentration was low; **chlorophyll -a :chlorophyll -b content was in low ratio.** The thickness of the lamina is less, the mesophyll is bifacial – heliofacial. The adaxial epidermis has larger and slightly elongated cells, and abaxial epidermal cells are thin. The midrib is obviously prominent. Based on the observation, the authors believe that in *Begonia elatior X hybriden*, the internal factors influence the exchange of gas between the leaf and environment and exchange of substances within the tissues. The values of various parameters are specific physiological indicators of the plant.

McLellan[28] considered that many combinations of features did not occur at random either due to functional constraint or genetic correlation. The distribution of variation in leaf morphology in the highly variable *Begonia dregei* species complex was examined in natural population and in F2 offspring from a cross between plants from two populations. The author quantified leaf shape using several morphometric measures; trichomes on the leaves were counted and measured. Correlations between leaf shape and the numbers and size of the trichomes were examined. There were significant correlations between the shapes of the leaves and the presence of number and size of the trichomes among populations and in the hybrid plants. According to the author deeply incised leaves had longer and larger number of trichomes at the sinuses. Higher number of trichomes on the leaf surfaces occurred together with the trichomes at the petiole and on the abaxial surface. The potential for

independent evolution of the leaf shape and trichomes in this group was reported to be limited.

Santhosh[29] considered *B.dipetala var. hydrophila* is an independent taxon and it is not a variety of *B.malabarica* as per the description of Clarke[25]. The former species is endemic to the Western Ghats of India. It seems that *B.dipetala var hydrophila* is often confused with *B.fallax* de Candole which stimulates very much former species. However, the latter species has lax inflorescence, more robust habit, male flowers with 4 perianth and female with 3 perianth lobes.

Ku *et al*.[30] reported a new species from China which has been named as *B.pengii* S.M.Ku & Yan Lieu. The authors have provided somatic chromosome number and karyotype, detailed morphological descriptions with clear illustrations, leaf anatomy including trichome morphology and ecology of the species.

Begonia tenera Dryander is a species endemic to Sri Lanka. This species has been reported from India by Shaju *et al*.[31] *B. tenera* is a perennial acaulescent herb with ovate or rotundate leaves; inflorescence is racemose, umbellate; tepals 2+2, white with pinkish periphery; tepals of the female flower are 5 in two whorls.

Santhosh and **Roy**[32] have provided taxonomic circumscription for *Begonia aliciae* C.E.C. Fischer, a niche specific endemic species of the Western Ghats of Kerala and Tamil Nadu. Santhosh *et al*.[33] rediscovered this species after a lapse of 64 years from the type locality from where it was first collected by Edward Barnes and the specific epithet was provided by Fischer[34] *B.aliaceae* is scapigerous herb. The male flowers have four perianth lobes and female flowers have six perianth lobes. The ovary is 2 celled and two winged. Makarand et al.[38] reduced *B. aliciae* to *B. crenata* Dryand.

Yoo Sung lee[35] made elaborate studies of stem anatomy of about 44 species of *Begonia* collected from different localities. The author studied the first internodes or the fifth inter nodes and the tissues of the inter nodes; collenchymas, sclerenchyma, secondary vascular tissues, periderm and trichomes; the conditions of the vascular ring and vascular bundles were analyzed during the study. The observation of different species was compared.

3. Aim and Scope of the Present Study

Careful perusal of literature so far reviewed may indicate that the systematic circumscription of *Begonia dipetala* has remained enigmatic for a long time. After a series of morphological studies, some consensus have been reached for the identity of the specific name of *Begonia dipetala* Santhosh kumar,[29]. We believe that the microscopic features of *Begonia dipetala* may supplement the morphological parameters, especially when the specimen lacks the floral parts. It has been shown that certain qualitative microscopic data are less prone to changes due to environmental stress. We aimed such of those features that are constant and consistent for *Begonia dipetala* and to prepare a protocol of data for applying such data for botanical identification of species.

4. Materials and Methods

Materials for the present study were collected from Hills near Salem (Tamil Nadu; India) during different seasons. The species identity was confirmed by Dr. Santhosh Kumar, Jawaharlal Nehru Tropical Botanical Garden and Research Institute, Palode, Kerala and the identification was confirmed by Botanical Survey of India, Howrah. (CNH13/ 2013/Tech II/958). Herbaria of voucher specimens were prepared and the specimens are lodged in the Plant Anatomy Research Centre, West Tambaram, Chennai - 45.

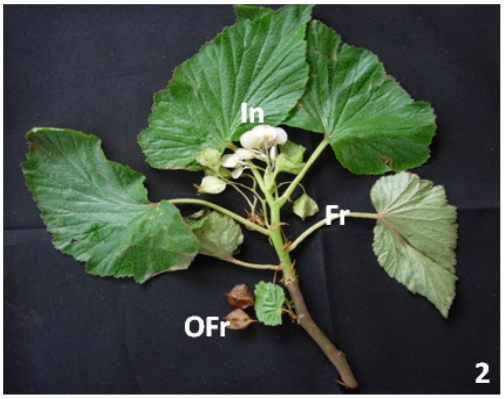
Different parts of the plant, such as leaf (midrib and lamina), petiole, stem, rhizome and roots were fixed in FAA (Formalin: Acetic acid: 70% Ethanol; 5:5:90) for 24 hrs. After fixation the specimens were washed, dehydrated by passing through Tertiary butyl alcohol (TBA) series following the procedure of Sass, 1940. After dehydration, the specimens were infiltrated with paraffin wax (melting point 56-58°C) ad embedded in the paraffin blocks. Sections were cut using Rotary microtome at a thickness of 10µm. Sections were stained with Toluidine blue (0.01%) aqueous solution. Photomicrographs were prepared with Nikon trinologular microscope and Nikon digital camera.

5. Observation

5.1. External Features

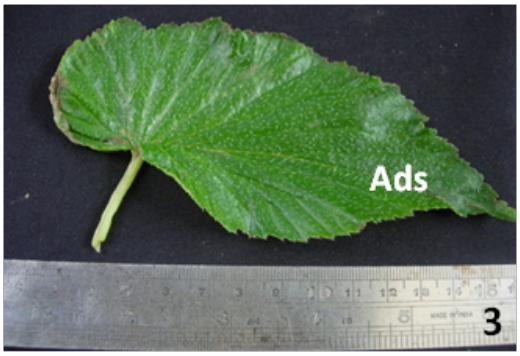
Begonia dipetala is a herb or sub - shrub growing predominantly in moist or water saturated habitats (Fig.1). It has thick stem which is pink or light red. The leaves are alternate (Fig. 2), distichous,obliquely ovate, semi cordate at base, apex acuminate (Fig.3&4). The inflorescence is an axillary raceme (Fig.2&5). The plant is monoecious with male and female flowers located on the same inflorescence; the male flowers are on the upper part of the peduncle, the female flowers being on the lower part (Fig.5&6). The flowers are white or pale pink. The flowers have two petals. The ovary is pink or red with truncate apex and three membranous wide semicircular wings (Fig 11). The stigmatic column bears spherical bodies (Fig.9&10). Ovary is tricarpellary, triangular in sectional view. Ovules are on axial placentation (Fig.12). The stamens are numerous forming a dense cluster located in between two petals (Fig.7&8). The seeds are elliptical, dark coloured, with reticulate, prominent thickenings on the surface (Fig.13).

Figure 1. *Begonia dipetala* in its natural habitat – Moist crevices of rocks



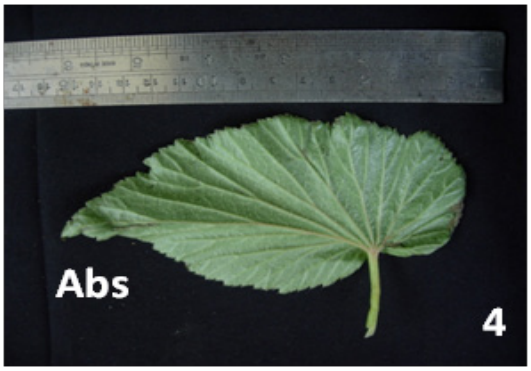
In – Inflorescenc, Fr – Fruit, OFr- Old Fruit

Figure 2. Flowering shoot system showing aphyllotaxy and inflorescence



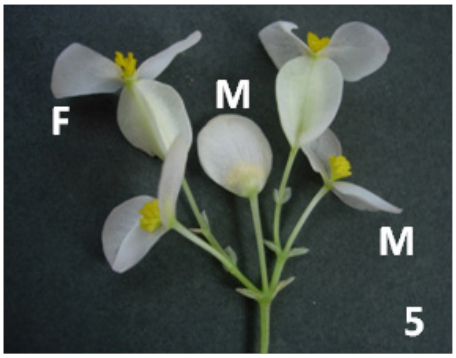
AdS – Adaxial Surface

Figure 3. Leaf showing adaxial view and shape



AbS – Abaxial Surface

Figure 4. Leaf in abaxial view



F – Female, M – Male

Figure 5. An inflorescence showing upper female flower and lower male flower

F – Female, M – Male

Figure 6. Female and male flowers – Enlarged

And – Androecium

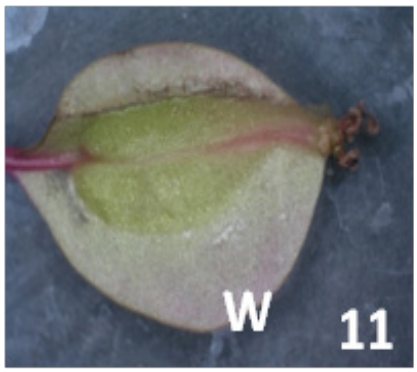
Figure 7. Two petals of the male flower open showing the cluster of stamens

Ant – Anther

Figure 8. Staminal cluster spread apart

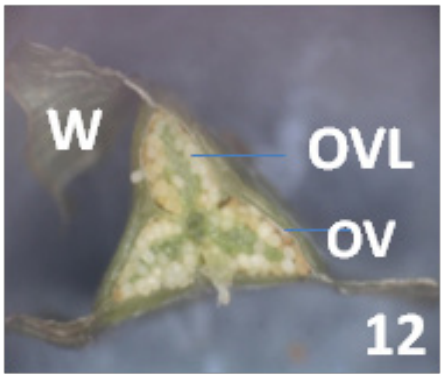
Figure 9. Female flower with two petals and stigmatic lobes

Figure 10. Stigmatic lobes – Enlarged



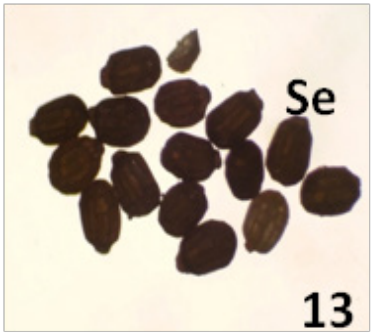
W – Wing

Figure 11. A mature winged fruit



W – Wing, OVL – Ovules, OV–Ovary

Figure 12. Cross section of the ovary showing the placentation and wings



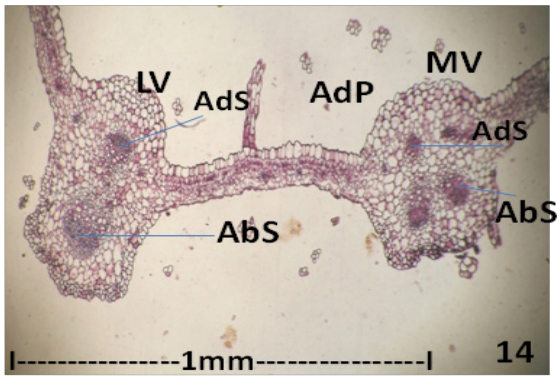
Se – Seeds

Figure 13. Surface features of the seeds

5.2. Anatomical Features

5.2.1. Leaf

The leaf is undulate in transectional view with ridges and furrows due to the prominent veins and thin lamina (Fig.14). The veins are multistranded. The number of vascular strands varies depending upon the size of the veins. The midrib has two larger abaxial vascular bundles and two smaller adaxial bundles (Fig.14). The bundles are collateral and the xylem units of the adaxial, abaxial bundles are juxtaposed (Fig.14). The smaller lateral veins have two vascular bundles which are adaxial and abaxial in position (Fig.14). The epidermal cells of the veins are thick with thin walls and less prominent cuticle. The ground tissue of the veins consists of polygonal thin walled, compact parenchyma cells.

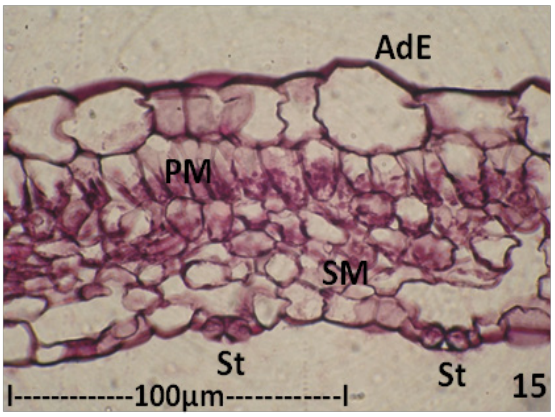


AbS – Abaxial Strand, AdP – Adaxial Part, AdS – Adaxial Strand, LV – Lateral Vein, MV – Mid Vein

Figure 14. Cross section of the leaf through mid vein and lateral vein

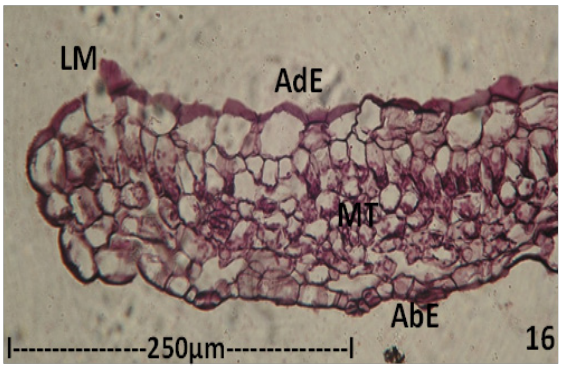
The lamina is 130-150µm thick. It is dorsiventral and heterofacial. The adaxial epidermis is uniseriate and the cells are 30-40µm thick. The abaxial epidermis is also uniseriate and thinner than the adaxial epidermis. The cells are narrow and cylindrical. It is stomatiferous. The stomata are raised above the level of epidermis and they have prominent stomatal ledges (Fig.15). The mesophyll includes adaxial band of palisade cells which are 30µm in height. The spongy parenchyma is four layered and the cells are small, spherical or lobed. The marginal part of the leaf is straight and semi circular; the epidermal cells of the margin are slightly larger

and thick walled (Fig.16). The mesophyll tissue remains unchanged (Fig.16).



AdE – Adaxial Epidermis, PM – Palisade Mesophyll
SM – Spongy Mesophyll

Figure 15. T.S. of lamina – Enlarged



LM – Leaf Margin, AdE – Adaxial Epidermis,
MT – Mesophyll Tissue, AbE – Abaxial Epidermis

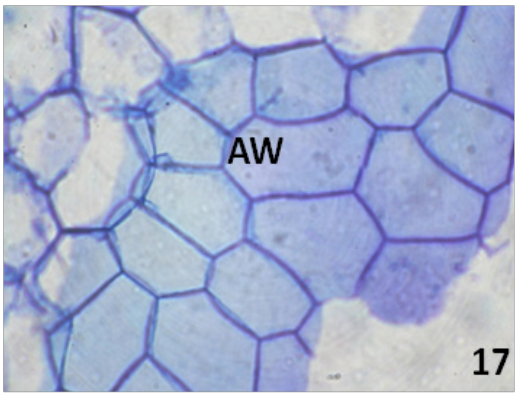
Figure 16. Marginal part of the lamina

5.2.2. Epidermal Trichomes

The epidermal trichomes are abundant on all parts of the leaf. Two types of trichomes are seen. Non-glandular trichomes are more frequent on the adaxial side. They are multicellular, and unbranched. The cells are vertically oriented and thin walled (Fig.19). The trichomes are emergences or shaggy type (Fig.19,30&31). The glandular trichomes are the second type which is more common on the abaxial side of the lamina. The glands are peltate type. It consists of a two celled short stalk and circular plate of 4 celled head, which is at right angles to the stalk. The head part of the gland is darkly stained and secretory in function (Fig.29). The gland is 60µm high and 70µm wide.

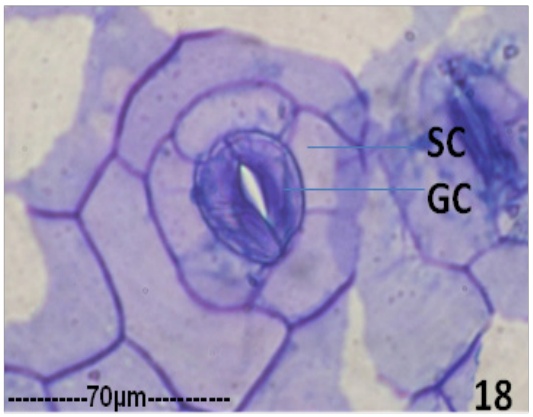
5.2.3. Epidermal Tissue

The adaxial epidermis, as seen in surface view, consists of polyhedral, angular, thin walled cells with straight walls. The epidermis is apostomatic (Fig.17). The abaxial epidermis is densely stomatiferous. The stomata are either **cyclocytic** with four subsidiary cells encircling the guard cells or **anisocytic** with three unequal subsidiary cells around a stoma (Fig.18).



AW – Anticlinal Wall

Figure 17. Surface view of the adaxial epidermal cells

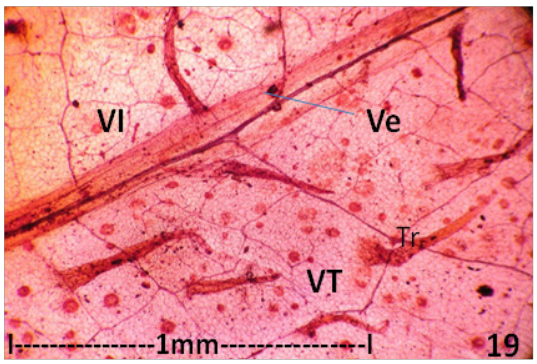


SC – Subsidiary Cells, GC – Guard Cells

Figure 18. Abaxial epidermis showing stomatal type

5.2.4. Venation Pattern

The lateral veins are thin and less prominent. They form fairly distinct rectangular and isodiametric vein-islets with long slender branched or unbranched vein-terminations (Fig.19).



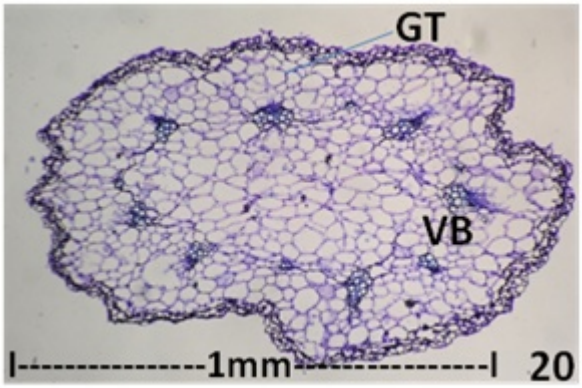
VI – Vein – Islet, VT – Vein Termination), Ve – Vein, Tr – Trichome

Figure 19. surface view of the lamina showing venation pattern and distribution of trichomes

5.2.5. Petiole

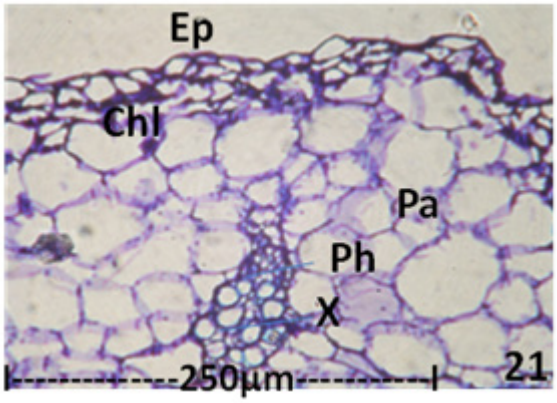
The petiole is 1.4mm thick and elliptical in outline; the

surface is uneven. The epidermal cells are conical and thin walled. There are two or three layers of chlorenchyma cells and the remaining ground tissue is thin walled parenchymatous. The vascular system is multistranded; there is a ring of 10 collateral vascular strands of different size and shape (Fig.20&21).



GT – Ground Tissue, VB – Vascular Bundle

Figure 20. T.S. of Petiole – Entire view



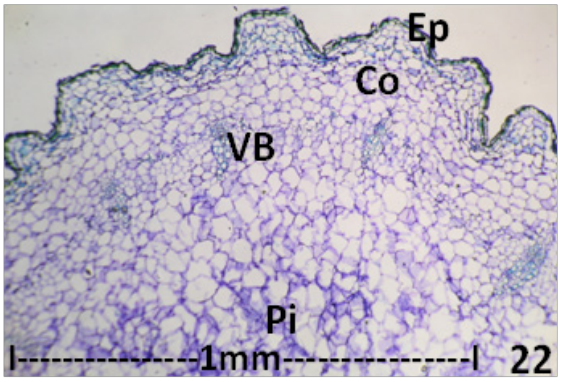
Ep – Epidermis, Chl – chlorenchyma, Pa – Parenchyma, Ph – Phloem, X- Xylem

Figure 21. T.S. of Petiole a sector showing vascular bundle

5.2.6. Stem

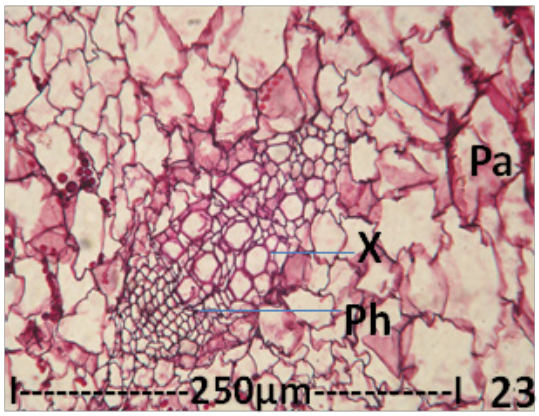
The young stem is circular in cross sectional view with irregular ridges and furrows. The epidermis has thin walled intact cells. The cortex consists of outer three or four layers of thick walled compact cells and inner thin walled parenchyma cells. The vascular system is **eustele** type with several discrete bundles and parenchymatous medullary rays. The vascular bundles are radially stretched and collateral. Xylem elements are wide, angular and thin walled; they occur in two or three rows of radial multiples (Fig.22&23). Sclerenchyma caps are seen on the outer end of the phloem units of each bundle. Starch grains are common in the cortex and pith cells.

Fairly thick stem has shallow ridges and furrows. Thin less prominent periderm with darkly stained two or three layers of cells occur on the surface of the stem. The cortex includes outer part of tangentially stretched cells and wide angular cells in the inner part. The vascular tissue exhibits initial stage of secondary growth. The primary vascular bundles are laterally interconnected by thin zone of secondary tissues in which the xylem elements have not yet mature. Phloem occurs on the outer part of the xylem which has sclerenchyma cap (Fig.24&25).



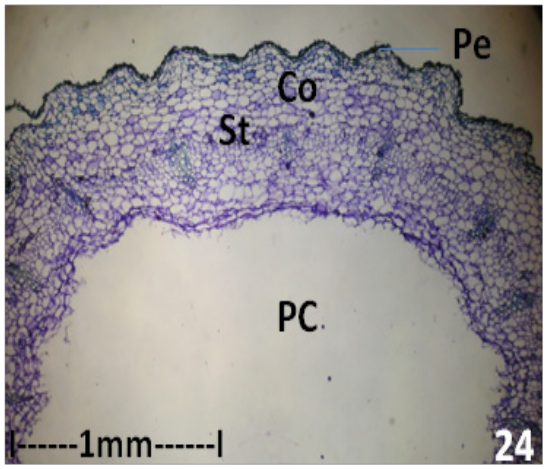
Ep – Epidermis, Co – Cortex, VB – Vascular Bundle, Pi – Pith

Figure 22. T.S. of Young stem



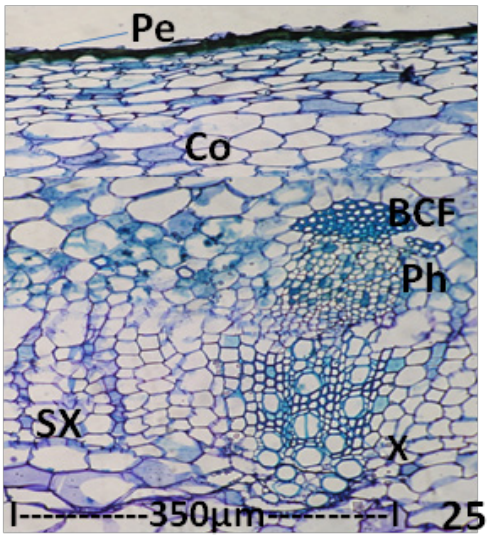
Pa – Parenchyma, Ph – Phloem, X- Xylem

Figure 23. A vascular bundle of the young stem



Pe – periderm, Co – Cortex, St – Secondary thickening, PC – Pith Canal

Figure 24. T.S. of Old stem



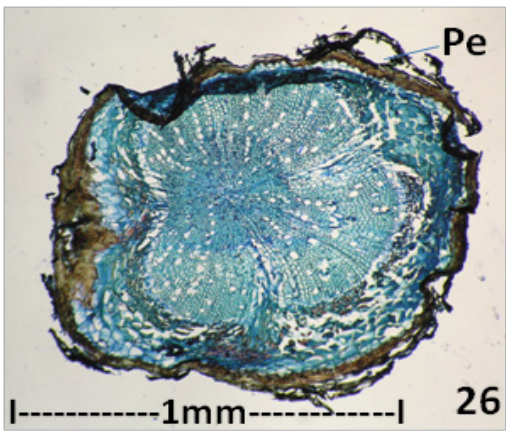
Pe – periderm, Co – Cortex, BCF – Bundle Cap Fibre,
Ph – Phloem, SX – Secondary Xylem, X- Xylem

Figure 25. Primary vascular bundle and interfasicular seondary xylem element

5.2.7. Rhizome

The rhizome is vertical thick cylinder of which gives rise to roots on all sides. It is deeply ridged with wide furrows. It consists of two or three layers of suberized periderm cells followed by rectangular or polygonal parenchymatous cortex (Fig.28). The vascular cylinder is thin and continuous, comprising inner part of conical primary xylem units and outer compact mass of phloem. Secondary xylem consists of thin walled radial rows of cells (Fig.28). Secondary phloem occurs in prominent circular masses on the outer part of the xylem cylinder.

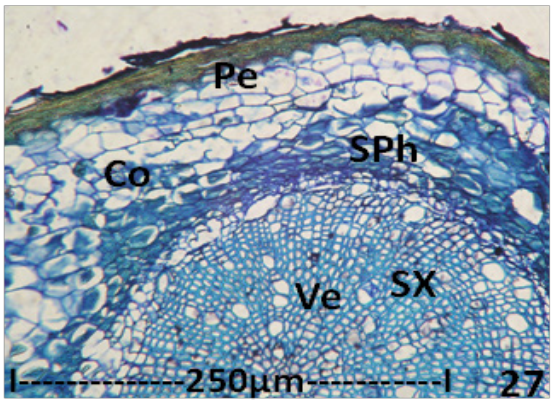
5.2.8. Root



Pe – periderm

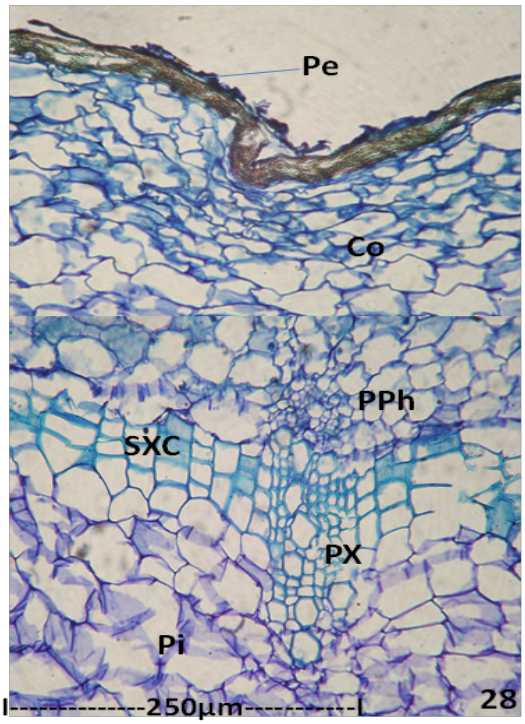
Figure 26. T.S. of Root – Entire view

A thick root measuring 1.8mm in diameter exhibits a thick periderm which is about five layered; the cells of the periderm are darkly stained and compact and cylindrical. The cortex has outer zone comprising radially oblong dilated cells. The root has large polygonal parenchyma cells most of them containing darkly stained tannin contents (Fig.26&27). The vascular cylinder is solid, dense and is wavy in outline. The secondary phloem is wide and continuous comprising clusters of sieve elements. The secondary xylem includes central part of darkly stained thick walled narrow fibers and a few diffusely distributed vessels. The outer part has fibres with wide lumen and narrow angular, mostly solitary vessels (Fig.27). The vessels are up to 40µm in diameter.



Pe – periderm, Co – Cortex, Ve – Vein, SX – Secondary Xylem,
SPh – Secondary Phloem

Figure 27. T.S. of Root – A sector enlarged



Pe – periderm, Co – Cortex, SXC – Secondary Xylem Cylinder,
PPh – Primary Phloem, PX – Primary Xylem, Pi – Pith

Figure 28. T.S. of Rhizome – A sector

6. Discussion

Begonia dipetala is established as an independent taxon.

The identity of the specimen is based on morphological criteria Santhosh,[29]. In the present study, microscopic features of vegetative organs of the plant are provided which are believed to throw more light on the diagnosis of the *Begonia dipetala*. In elaborate anatomical studies of the stems of *Begonia*, Lee[35] tried to correlate the stem anatomy and sectional classification of *Begonia*. Lee remarked that according to many investigators of *Begonia*, the epidermis of the lamina is multilayered. He disagreed with the observation of the previous authors and said in 44 species of *Begonia* which he studied have only unistratose epidermal layer. In the present study on *Begonia dipetala* also we could see only single layered epidermis both on the adaxial and abaxial sides of the lamina. According to Metcalfe and Chalk[36] the lamina in the members of Begoniaceae has hypodermal layer of parenchyma cells; they also reported that stone cells, idioblasts and similar mechanical elements are frequent in the mesophyll or around the veins; the palisade cells are locally replaced by stone cells; the vascular bundles are sometimes surrounded by collenchymas or elongated parenchyma cells.

In the present observation of *Begonia dipetala,* the structure of the lamina is simple and structure mentioned by Metcalfe and Chalk[36] is not evident. The adaxial epidermis consists of highly dilated cells measuring 40µm thick. Beneath epidermis is a compact layer of short wide palisade cells which are 20-40 µm in height. The stone cells and the idioblasts are absent; the vascular system of the midrib is multistranded comprising two pairs of vascular bundles in midrib and single pair in the lateral veins; the vascular bundles are adaxial and abaxial in position and they are juxtaposed with the xylem poles (Fig.14). This aspect of the vascular system of the veins of the leaf has been given due accent by early investigators. It is felt that vascular structure of the veins may serve as a valuable source of botanical diagnosis of the plant. The epidermal trichomes are said to be highly diversified and they have elaborately discussed by **Solereder**[37] based on the studies of **Fellerer [**14] on the hairy covering in Begoniaceae. **Solereder [**37] remarks that the epidermal hairy covering is exceptionally varied and is suitable for employment in specific diagnosis. Of different types of trichomes described by Solereder, we could recognize in *Begonia dipetala* the shaggy trichomes, capitate and peltate glandular trichomes. The shaggy trichomes are multiseriate with thick basal part which gradually become conical at the tip or uniseriate, one or two celled tip (Fig.30&31). A spherical darkly stained epidermal body is often seen at the terminal cell, which becomes shrunken deciduous at later stage. The shaggy trichomes do not form free mamilliform tips on the lateral sides as described by **Solereder[**37] (Fig.30&31). Apart from afore said trichomes, there are sessile, spherical dark brown unicellular shinning bodies, seen within the angles of the marginal serrations and elsewhere on the lamina (Fig.32). This type of glands have not been recorded by the previous investigators. The brown shining glandular bodies have a short, broad, bowl shaped stalk cell and spherical body with thick walls and dark contents.

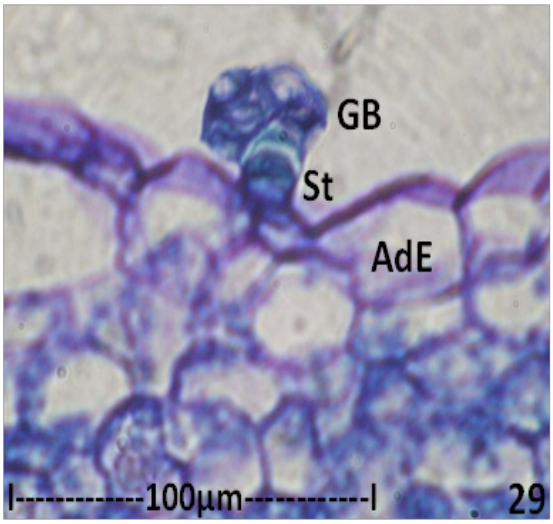
Solereder[37] remarks that the epidermal cells of the leaf are larger and have thick straight lateral (anticlinal) walls in Begoniaceae. The size of the epidermal cells is said to be employed for the recognition of the sections of the genera or in the diagnosis of individual species within the same species. In *Begonia dipetala,* the epidermal cells, particularly the adaxial cells are quite thick; the anticlinal walls are always thick and straight. The stomata are either cyclocytic with encircling subsidiary cells, or anisocytic with three unequal subsidiaries. These features of the epidermal tissues are of taxonomic values.

Petiole with discrete bundles forming a ring and presence of medullary bundles with or without sclerenchyma elements have been said to be of diagnostic values for *Begonia*. In *Begonia dipetala,* there is neither medullary bundle nor sclerenchyma elements. The petiole is wavy in sectional view; there is a thin layer of chlorenchymatous cells inner to the epidermis. Isolated small collateral bundles are organized in a ring (Fig.20&21). These features are specific for *Begonia dipetala*.

The internode of *Begonia* has been studied by some investigators (Lee, 1974). According to the available literature, the general features of interest are the cortical bundles and medullary bundles. In some of the species of both cortical and medullary bundles have been reported, in others either cortical bundles alone or only medullary bundles occur Lee,[35]; Solereders,[37].

Begonia dipetala lacks both cortical bundles and medullary bundles. The young stem has **eustelic** type of vascular system comprising several discrete primary collateral vascular bundles arranged in a ring. As the stem grows in thickness, medullary rays give rise to interfasicular cambial strips which produce initial secondary xylem and phloem. The epidermal layer is unistratose. In the old stem, the epidermal layer and a subepidermal layer get compressed into thick dark surface on the stem. The superficial cortical cells form a thin, less prominent periderm layers (Fig.22&24). Wide circular pith canal develops in the old stem due to lysigenous process of the pith cells.

Begonia dipetala is a rhizomatous species having horizontal, succulent rhizome. The **rhizome** consists of a wide hollow central homogeneous parenchymatous cortex and a continuous vascular cylinder. The cylinder comprises several radially stretched collateral vascular bundles which are interlinked by interfasicular secondary xylem-phloem elements (Fig.28). The vessel elements are poorly differentiated both in the fasicular and interfasicular portions. The **root** has thick darkly stained outer zone of crushed epidermis; subsequently an incipient periderm is formed from the outer cortical cells. The secondary xylem cylinder is dense and solid comprising sparsely distributed, thin walled, narrow angular vessels (Fig.26&27). The rhizome and root do not possess any specific or unique features and they do not throw much light on taxonomic identity of *B.dipetala*.



GB – Glandular Body, St – Stalk, AdE – Adaxial Epidermis

Figure 29. A peltate glandular trichome on the adaxial side of the lamina

Figure 30. Nonglandular shaggy type of trichome

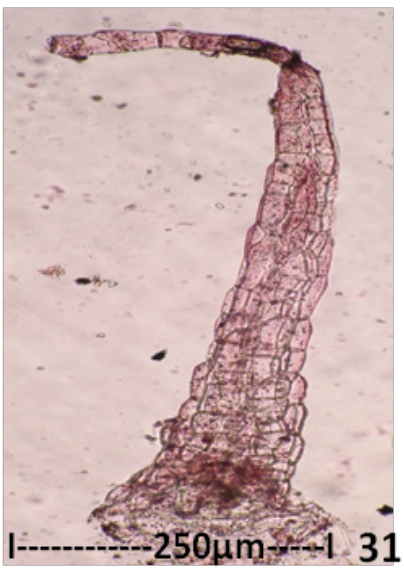


Figure 31. Nonglandular shaggy type of trichome

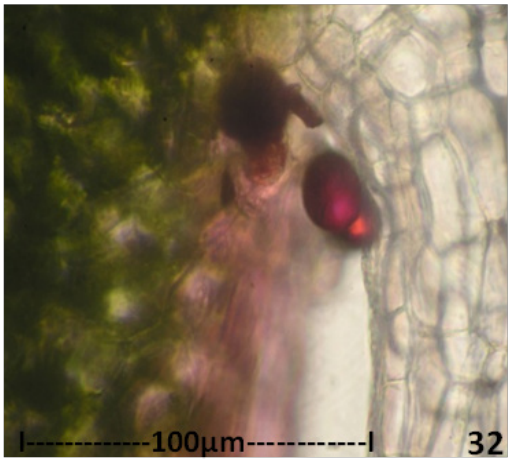
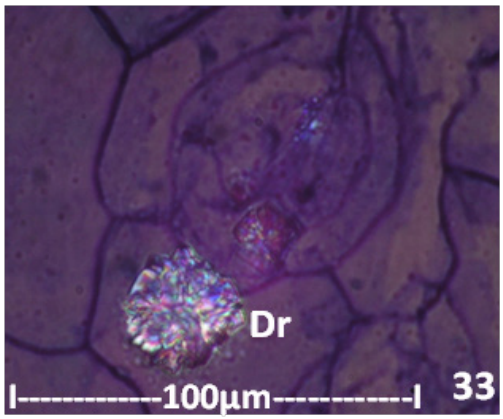
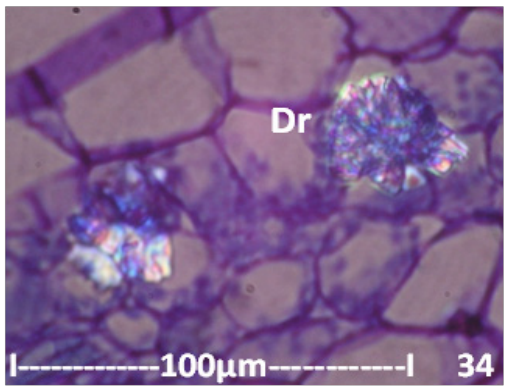


Figure 32. Deeply coloured sub-sessile spherical glandular trichome on the lamina



Dr – Druses

Figure 33. Calcium oxalate druses in the mesophyll tissue of the lamina

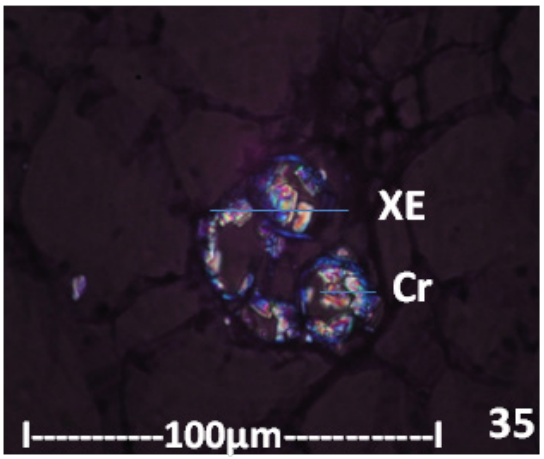


Dr – Druses

Figure 34. Calcium oxalate druses in the mesophyll tissue of the lamina

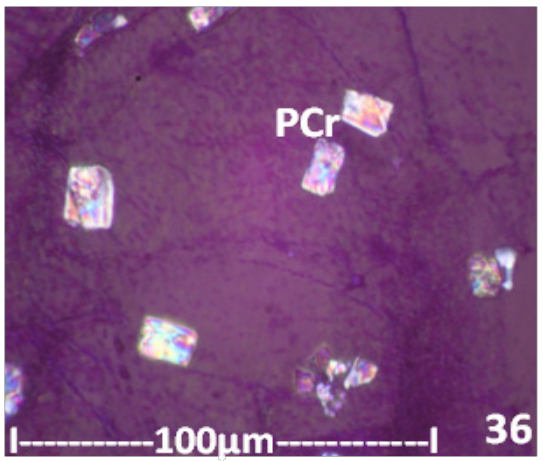
Ergastic substances of diversified chemical composition and morphological categories are persuasive in the tissues of all part of the plant. Calcium oxalate crystals are most widespread storage material in plant. Unfortunately, there is much that remains obscure and opinions differ concerning the role that calcium oxalate crystals play in the plant metabolism. Neutralisation of unwanted oxalic acid, reintroduction into metabolic cycle during the demand of calcium ions, mechanical protection of the plants against

plant feeding animals and mechanical rigidity to the tissues which bear the crystals are commonly prevailing views on the role of crystals in plant nutrition. Irrespective of the physiological activities of the crystals, their morphology coupled with their specific localization in the plant organs is much reliable in diagnostic procedure of plants. Among different morphological categories of calcium oxalate crystals **druses** and **prismatic** types are more prevalent in plants. In *B.dipetala* druses are found in the ground parenchyma of the cortex of rhizome, stem and petiole. It is unique that druses are seen in xylem elements occluding lumen of the cells (Fig.35). Occurrence of druses within the vessel elements was seen in some more plant species (personal observation). The process of accumulation of crystals within the xylem elements and reason for this process are problems for more detailed studies. Calcium oxalate druses are found in the mesophyll tissue of lamina (Fig. 33&34). Prismatic crystals of cuboidal type are seen in the stem, rhizome and petiole mixed with druses. Starch grains occur in the interfascicular derivatives of the stem and rhizome.



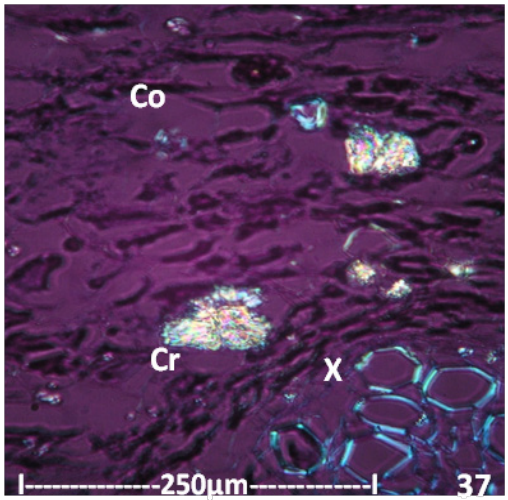
XE – Xylem Element

Figure 35. Crystal deposition in the lumen of xylem elements



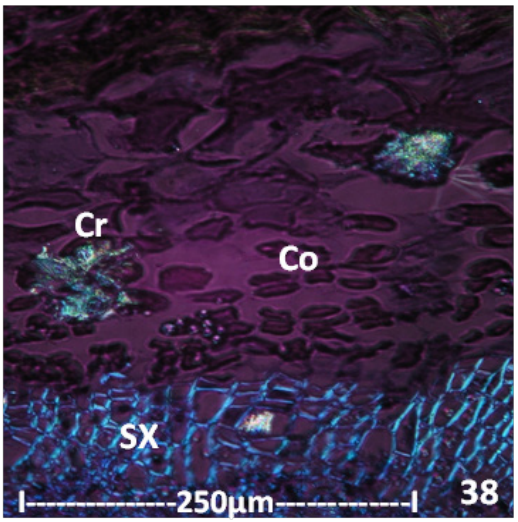
PCr – prismatic Crystals

Figure 36. Prismatic crystals in the cortical parenchyma of the stem



Co – Cortex, Cr – Crystals, X – Xylem

Figure 37. Druses in the cortex of the Rhizome



Cr – Crystals, Co – Cortex, SX – Secondary Xylem

Figure 38. Druses in the cortex of the Root

The general anatomical profile of *B.dipetala* stands aloof from the species studied by other investigators. Many structure reported by Solereder[37], Metcalfe and Chalk[36] and others are absent in *B.dipetala*. An overview of all features of *B.dipetala* and a holistic approach to the problem may help to resolve the taxonomic status of *B.dipetala*.

REFERENCES

[1] D.J. Mabberely, The Plant Book, 1st South Asian ed. Cambridge University Press. 2005. p 81.

[2] H. Santhapau and A.N. Henry, A dictionary of the flowering plants of India. 1972. Publ. Infor. Directorate CSIR, New Delhi.

[3] Mathan C.Nisha, Sevanan Rajeshkumar, Thangavel Selvaraj

and Modurpalayam S.Subramanian. A valued Indian medicinal plant – *Begonia malabarica* Lam.: Successful plant regeneration through various explants and field performance. *Maejo Int. J. Sci. Technol*. 3(2009) p 261 – 268.

[4] H.H. Johnstone, 1826. in Spotlight of Begonia dipetala by Mary Weinberg. *Begonian*. 1990. 57: 59 – 60.

[5] Thwaites, G.H.K., *Enumeratio Plantarum Zeylaniae*, 1859. On *Begonia dipetala* Graham. 128.

[6] A de Candolle, On *Begonia dipetala* Graham in A.P. de Candolle, Prodr. 15(1864): 392.

[7] E. Irmscher, In Pareys Blumengartnerei. 1960. On *Begonia dipetala* Graham. 78

[8] E.S. Santhosh Kumar, Seema G. Gopal and G.M. Nair , Re-appearance of Rheede's *Tsjeria-narinampuli* – Its identity after the discovery and a note its allied species. *Begonian*. 72 (2005) p135 – 140.

[9] H. Rose, and M. Hurd-Karrer, Differential staining of specialized cells in *Begonia* with indicators. *Plant Physiology* 2(1927) p 441 – 453.

[10] Watson., and M.J.Dallwitz, The families of flowering plants: descriptions, illustrations, identification, and information retrieval. 1992. Website http:// biodiversity. uno. edu/ delta/ [accessed 00 Month Year]

[11] Anisoara Stratu, Naela Costica and Oana Puschiu, Histo – Anatomical and physiological aspects of plant species with flower decoration. *Biologie vegetala*. 7 (2011) p 13 – 18.

[12] Hooker. 1830 cited in A study of stem anatomy of *Begonia* by Lee. 1974.

[13] F. Hildebrand, Anatomische Untersuchungen uber die Stamme Der Begoniaceen. 1859. Berlin.

[14] C. Fellerer, Anatomie und Systematik der Begoniaceen. 1892. (Thesis). Germany.

[15] G. Haberlandt, Physiological Plant Anatomy. 1914. Macmillan, London

[16] H. Hailler, Uber die verwandtschaftverhaltnisse bei Engler's rosalen, parietalen myrtifolen und in anderen ordnungen der dikotylen. 1903. Hamburg.

[17] V.Vouk, Uber eignartige Pneumathoden an dem Stamme von *Begonia vitifolia* Schott. Berr. Deutsch. Bot. Ges. 30 (1912)p 257 – 262.

[18] K.A. Fotsch, Die Begonian. Works Press Administ. 1939. o.p. 465-03-3-630.

[19] K.S. Boghdan, Pubescence in *Begonia*. Northeastern University Thesis. 1967 (Unpublished).

[20] K.S. Boghdan, and F.A. Barkley, *Begonia* trichomes. *The Begonian* 36 (1969) p130 – 132.

[21] F.A. Barkley, and B. Hozid, Leaf Anatomy of *Begonia*. *The Begonian* 38 (1971) p135 – 142.

[22] K.S. Boghdan, The foliar development in three species of *Begonia*. Northeastern University Dissertation. 1973 (Unpublished).

[23] S. Ganesan, N. Suresh, and L. Kesaven, Ethnomedicinal survey of lower Palani Hills of Tamil Nadu. *Indian Journal of Traditional Knowledge* 3(2004) p 299 – 304.

[24] Rheede Tot Drakenstein, H.A. van 1689. *Hortus Indicus Malabaricus*. Vol.9. Amsterdam.

[25] C.B. Clarke, Begoniaceae: In Hooker, J.D. (Ed.) *Flora of British India*. L. Reev & Co. London. 2(1879) 650 – 656.

[26] J.S. Gamble, Begoniaceae: *Flora of Presidency of Madras*. 1919. p. 544 – 547, Aldanrd & Sons, Limited.

[27] S. Rajbhandary, M. Hughes, T. Phutthai, D.C. Thomas, and K.K. Shrestha, Asian *Begonia*: out of Africa via the Himalayas? *Garde Bulletin Singapore* 63(2011) p277 – 286.

[28] T. McLellan, Correlated evolution of leaf shape and trichomes in *Begonia dregei* (Begoniaceae). *American Journal of Botany*. 92(2005) p1616 - 1623.

[29] E.S. Santhosh Kumar, *Begonia dipetala* Graham. *var hydrophila* (C.B.Clarke) Santhosh & Seema a little known endemic species of the Western Ghats of India. *Begonian* 74 (2007) p 223 – 225.

[30] Ku Shin – Ming, Kono Yoshiko and Liu Yan. *Begonia pengii* (sect. Coelocentrum, Begoniaceae), a new species from limestone areas in Guangxi, China. *Botanical Studies* 49 (2008) p167 – 176.

[31] T. Shaju, P.K. Shaji, M.P. Geethakumary, and E.S. Santhosh Kumar, *Begonia tenera* Dryander: A new report for India. *Begonian* 7 (2010) p129 – 131.

[32] E.S. Santhosh Kumar, and P.E. Roy, *Begonia aliciae* C.E.C. Fischer a little known critically endangered species of Southern western Ghats, Kerala, India. *The Begonian* 79 (2012) p 16 – 17.

[33] E.S. Santhosh Kumar, M. Abdul Jabbar, and M. Saleem, Rediscovery of *Begonia aliciae* C.E.C. Fischer (Begoniaceae) from the Western Ghats of Kerala. *Rheedea* 12(2002) p 185 – 188.

[34] C.E.C. Fischer, New or little known plants from Southern India. X. *Begonia aliaciae* C.E.C. Fisch. Kew Bull. 5(1939) p 247.

[35] You Sung Lee. A study of stem anatomy in *Begonia* L. *Phytologia* 27 (1974) p 464 – 489.

[36] C.R. Metcalfe, and L. Chalk, Anatomy of the Dicotyledons. Vol.I 1950. p. 691 – 695. Oxford at the Clarendon Press, London.

[37] H. Solereder, Systematic Anatomy of the Dicotyledons. (Transl. L.A. Boodle and F.E. Fritsch, revised by D.H. Scott.) Vol- I 1908. p398 - 406. Clarendron Press. Oxford.