



Research Article

Anatomical studies on medicinal and endangered CAM plants of Milkweed family from Thar Desert, Rajasthan, India.

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Abstract: The present investigation was carried out to screen anatomical features of *Sarcostemma viminale* (L.) R.Br., *Ceropegia bulbosa* Roxb. var. *bulbosa* and var. *lusbii* (Grah.), belonging to family Asclepiadaceae. Plant specimens were collected from semi-arid region of Thar Desert in Rajasthan. These plants are medicinally important and endangered, have been traditionally used as an antimicrobial, antifungal, anticancerous and antioxidant. *Sarcostemma viminale* (L.) R.Br. is used to cure diarrhoea, oedema and tuberculosis. *Ceropegia* species is used to cure deafness. Tubers are used in the treatment of kidney stone, urinary tracts diseases and eaten by ladies to enhance fertility and viability. The microscopical illustrations revealed interesting features i.e. presence of stone cells, rosette crystals, oil globules, phloem fibers, intraxylary phloem, pericycle patches, large medullary rays, annular vessels, more amount of palisade, mesophyll and multi-cellular trichomes etc. These features supported the habitat condition of Thar Desert and its Crassulacean Acid Metabolism (CAM) nature. This study provides referential pharmaco-botanical information for identification, authentication, standardization and detection of adaptation strategies and to develop a protocol to conserve them for further use.

Key words: *Sarcostemma viminale* (L.) R.Br., *Ceropegia bulbosa* Roxb. var. *bulbosa* and var. *lusbii* (Grah.), Asclepiadaceae, CAM, Intraxylary phloem, Rosette crystals.

Introduction

Medicinal plants are a source of great economic value all over the world (Maobe *et al.*, 2013). Medicinal plants become one of the major objects of interest as these are gifts of nature to cure limitless number of diseases in human beings (Busha and Ganga, 2003). Medicinal plants are the richest bio-resources of folk medicines, traditional medicine, food supplements, nutraceuticals, pharmaceuticals and chemical entities for synthetic drugs (Ncube *et al.*, 2008). The quality of medicinal plant may be affected by the number of environmental factors such as climate, altitude, rainfall and other conditions which affect the growth of plants (Rajak *et al.*, 2015). Rajasthan is identified as the “Desert state of India”, it spread across the western part of India. In this region vegetation is sparse as the plants with xerophytic adaptation can only survive. Xerophytic plants are characteristics of desert and semi desert regions; they grow naturally in dry climates and adapted to live in low humidity, can tolerate extreme temperature and drought for long period of time. CAM plants efficiently survive in typical and harsh conditions of Desert, they are extremely good at conserving water. CAM is a mechanism that enables plants to survive in draught areas. Both the genus under study has been added to the list of Indian endangered plants (Nayar and Sastry, 1987). Plants using CAM also shows reduced leaves with thick cuticle and sunken stomata, they stored malic acid in the vacuoles during the night and uses it during

the day (Raven *et al.*, 2005). Plant anatomy deals with the structure, contents and development of cells and tissues, it has primary importance for all aspects of research in plant sciences such as morphogenesis, taxonomy, physiology, ecology, evolution, genetics and reproduction etc. (Fahn A, 1990).

The plants of family Asclepiadaceae remains distributed from south east Asia, India, Madagascar, Tropical Arabia, Canary Island, and Africa except Mediterranean region, New Guinea and north Australia (Bruyns, 2003). Asclepiadaceae, now ranked as subfamily of Apocynaceae, is a large group of many plant species with wide therapeutical properties. This view was also upheld by the Angiosperm Phylogeny Group (APG 2003, 2009). It is comprised of approximate 130 genera and 2000 species, tropical as well as subtropical, mostly shrubs, often twining or perennial herbs (Indhumathi and Kalvimoorthi, 2010).

The roots, stem and latex of *Sarcostemma* are employed in treating many disease like mouth ulcer, jaundice, sour, throat, ulcer (Khan, 2002). The genus *Ceropegia* as a whole is under threat, owing to either destructive collection or habitat degradation. *Ceropegia bulbosa* is one of the widely distributed species but still threatened (Yadav and Kamble, 2008). Several interesting morphological similarities and differences in both plants under study have

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been observed right from habitat to seed formation (Arora and Meena, 2016). In present study an effort is being made to describe detailed anatomy of both endangered plants.

Materials and Methods

Plant material of *Sarcostemma viminalis* (L.) R.Br. was collected from Jodhpur and Udaipur district and *Ceropegia* species was collected from Bhilwara, Chittorgarh, Jaipur, Udaipur and Karolli district of Rajasthan. "The Flora of Indian Desert" (Bhandari, 1978) was consulted for specimens identification. The specimens were finally authenticated by BSI (Botanical Survey of India), Jodhpur (Rajasthan). Various plant parts were excised from healthy plants and fixed in FAA (formalin-5ml, acetic acid-5ml, 70% ethyl alcohol-90ml). Hand cut transverse section were studied is safranin and were observed in Quasmo 66502 binocular pathological research

microscope for finest resolution. Photomicrographs of different magnification (4x, 10x, 40x, 100x) were taken using a Nikon 750 digital camera.

Result and Discussion

The study revealed valuable differences and similarities in anatomy of two representative genera of family Asclepiadaceae.

(1) Botanical name- *Sarcostemma viminalis* (L.) R.Br.

Syn. *Euphorbia viminalis* L.

Accepted Scientific Name is *Cynanchum viminalis* (L.) L.

Local name - Caustic vine, khir-khimp, somlata, Melktou, Rapunzel

(2) Botanical Name: *Ceropegia bulbosa* Roxb. var. *bulbosa*

Local name: Art khadula, Barbatta, Khaparkundis

Botanical Name: *Ceropegia bulbosa* Roxb. var. *lusbii* (Grah.) Hook.f.

Local name: Hedulo, Solari,



Fig. 1. T.S. of Root (10X)

Cambium cells (40X)

Rosette Crystals (40X)



Fig. 2. T.S. of Root (4X)

T.S. of Root (10X)

Vascular bundles & Starch grains (40X)



Fig.3. T.S. of Root (4X)

T.S. of Root (10X)

Vascular bundles (40X)

Root

Sarcostemma viminalis (L.) R.Br.

Bark on the exterior gets ruptured and followed by cork that is 3-4 layered (squarish), Primary cortex get ruptured, pericycle forms cork cambium (2 layered), it cuts of cork on outer and secondary cortex (phelloderm) on inner side. Secondary cortex is made up of parenchyma, collenchymas and scleranchymatous ring that posses stone cells too. Stelar region contains cambium it made up of 6-7 layers. It forms secondary xylem on inner side and secondary phloem on outer. Secondary phloem

contains phloem fiber. Secondary xylem contains long medulary rays showing alternating light and dark regions. Conjunctive tissue is in enormous amount. Primary xylem is present in the inner core of stele with few parenchymatous pith cells (Fig.1).

Ceropegia bulbosa Roxb. var. *bulbosa*

Transverse section of root revealed single layered epidermis with extended root hairs on the complete periphery. Cortex is widest part, composed of vaerulated parenchyma. Inner most part is surrounded by single layer pericycle and

endodermis. Xylem is triarch and exarch. Phloem alternates xylem. Pith is small and parenchymatous with starch grains (Fig.2).

Ceropegia bulbosa Roxb. var. *lushii* showed almost similarly except difference lies in stellars region only where xylem is tetrarch (Fig. 3).

Stem

Sarcostemma viminale (L.) R.Br.

Transverse section of stem revealed presence of cork at exterior most. It is followed by periderm (2-3 layered). Cork cambium forms cork on outer side and cortex on inner side. Cortex is made up of chlorenchyma, collenchyma and parenchyma. Chlorenchyma is outer most tissue and is present in least amount, while parenchyma is inner most part of cortex and is present in maximum amount without any airspace. Cortex is followed by single layered endodermis. Pericycle is present next to endodermis in form of brittle patches. These patches are made up of thick walled cells and are interrupted by presence of elongated phloem fibers. Next to pericycle primary phloem fibers are present in a ring. This region is specific as it separates secondary phloem from pericycle. Vascular cambium is present as less stained 2 celled thick area. Secondary xylem is present next to vascular cambium, highly conspicuous; possess large vessels

and resin ducts. The vascular bundles are conjoint, collateral and open. Secondary xylem shows clearly defined secondary medullary rays. Primary xylem is pushed towards centre. Protoxylem faces centre. On exterior most of pith intraxylary phloem is present in uneven amount, making conical patches. Pith is present in conspicuous amount, having parenchyma with small airspace and large amount of rosette crystals (Fig.1).

Ceropegia bulbosa Roxb. var. *bulbosa*

Transverse section of stem showed thick cuticle, wavy outline, cortex broad made up of chlorenchyma and collenchymatous, pigments present in cell wall only. Endodermis single layered, pericycle in form of sclerenchymatous patches. Cambium ring separates vascular tissue in a crossform. Amount of vascular tissue in opposite sides is equal. Two poles contain more amount as compared to other poles of a cross. Vascular bundles are conjoint, collateral, open and endarch. Intraxylary phloem is present towards pith. Pith is more in amount with starch grains (Fig.2).

Ceropegia bulbosa Roxb. var. *lushii* showed almost similarly except amount of vascular tissue, that is xylem is more in amount as compared to xylem in *Ceropegia bulbosa* Roxb. var. *bulbosa* (Fig.3).

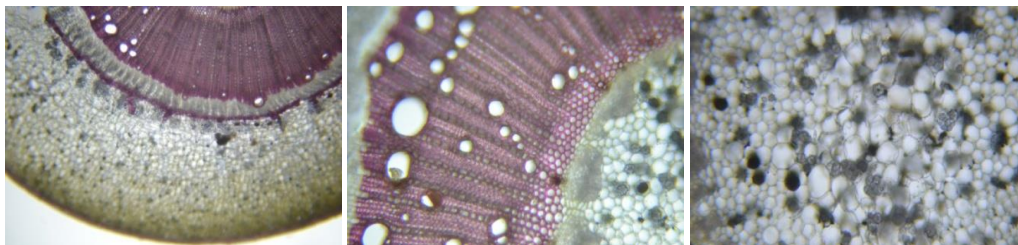


Fig.1. T.S. of Stem (4X)

Vascular bundles arrangement (10X) Pith and Rosette crystals (10X)

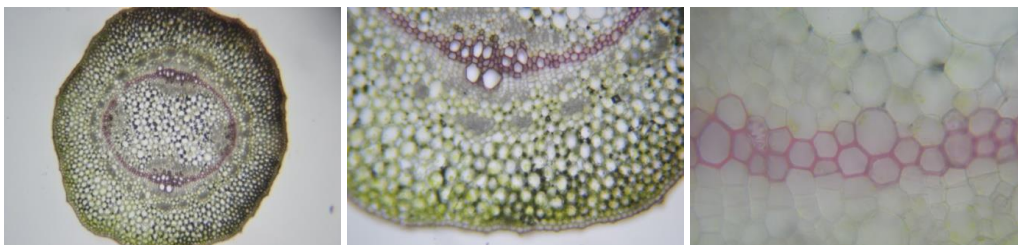


Fig.2. T.S. of Stem (4X)

Vascular arrangement (10X)

Vascular arrangement (40X)

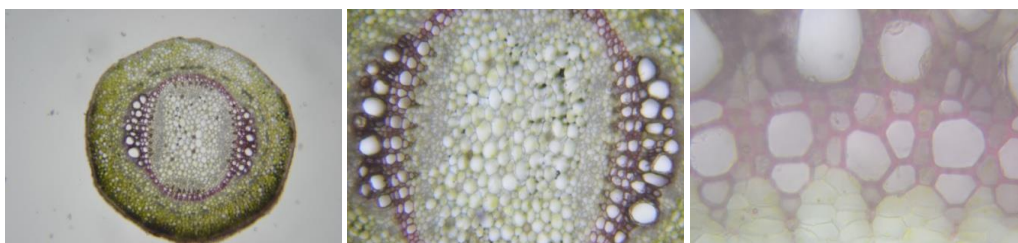


Fig.3. T.S. of Stem (4X)

Vascular arrangement (10X)

Vascular arrangement (40X)

Leaf

Sarcostemma viminalis (L.) R.Br.

Transverse section of leaf is boat-shaped. It is isobilateral. Epidermal cells isodiametric, size almost similar on both sides, covered with thick cuticle, stomata rare, present on lower epidermis only, they are paracytic type. Leaf trichomes diffused on radius, with septate cells. No tissue differentiation is seen. Cortical tissue is chlorenchyma with some air spaces. Some cells donot possess the green pigment. Tannin cells and oxalate crystals remain diffused in cortical cells. Vascular bundle are closer toward upper epidermis. Xylem element alternates with phloem. Metaxylem is present towards upper epidermis. Midrib region possess more xylary and phloem elements as compared to wings (Fig.1).

Ceropegia bulbosa Roxb. var. *bulbosa*

Transverse section of leaf shows upper and lower epidermis clearly defined, cuticle is present.

Margin shows some trichomes (septate). On lower epidermis anisocytic and paracytic stomata are present. Midrib region possess tissue differentiation. Upper side has palisade tissue while lower side possess collenchymatous tissue. In between both the tissue vascular bundle is present. Protoxylem faces upper epidermis. Single layered cambium is present between xylem and phloem. Phloem is present towards upper epidermis. Phloem fibers are also present between xylary elements. On lateral sides of midrib there is no tissue differentiation, all the tissue present is palisade. Annular vessels are present in between palisade (Fig.2).

Ceropegia bulbosa Roxb. var. *lushii* showed almost similar characters. Anisocytic stomata are present on lower epidermis. Pallisade is less as compared to *Ceropegia bulbosa* Roxb. var. *bulbosa*. More tissue differentiation on lateral side of midrib is shown (Fig.3).

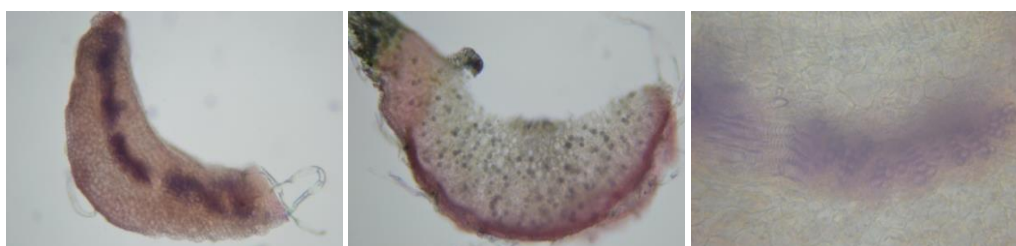


Fig. 1. T.S. of Leaf (10X)

Rosette crystals (10X)

Vascular bundle(40X)

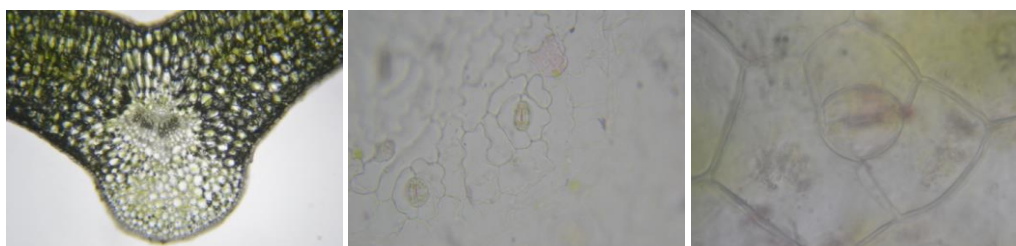


Fig. 2. T.S. of Leaf (4X)

Paracytic stomata (40X)

Anisocytic stomata(40X)

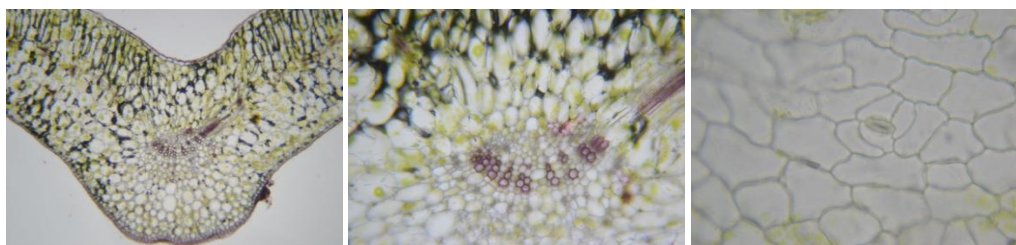


Fig. 3. T.S. of Leaf (4X)

Vascular bundle (10X)

Anisocytic stomata (40X)

Flower

Sarcostemma viminalis (L.) R.Br.

Flower is simple, umbel, creamish-white, tetra-pentamerous, actinomorphic, glandular, gynostegium present over inner coronary flap (modified petal), staminal column with translators, pollinia large and curved. Ovary superior, bicarpellary and apocarpous. Stigma disc shaped, marginal placentation with many ovules/locules (Fig.1).

Ceropegia bulbosa Roxb. var. *bulbosa*

Flower is umbel or cymose, pentamerous, white with purplish coronary cage. Corona is associated with pollination and nector production, gynostegium present, ovary superior, apocarpous, bicarpellary and multiovular (Fig.2).

Ceropegia bulbosa* Roxb. var. *lushii

Flower almost similar to *Ceropegia bulbosa* Roxb. var. *bulbosa*. Color is less purplish at upper part of corollary cage (Fig.2).

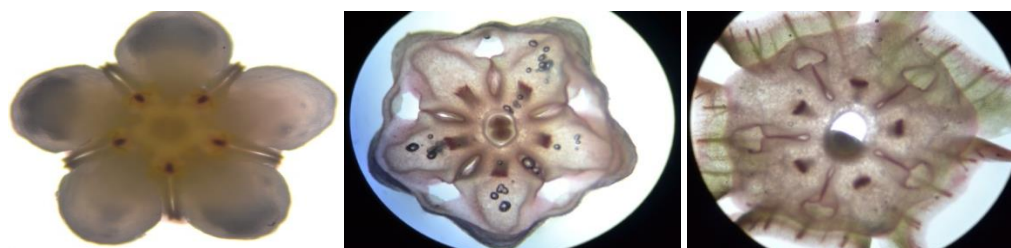


Fig. 1. T.S. of flower (4X)

T.S. of flower (10X)

T.S. of flower (40X)

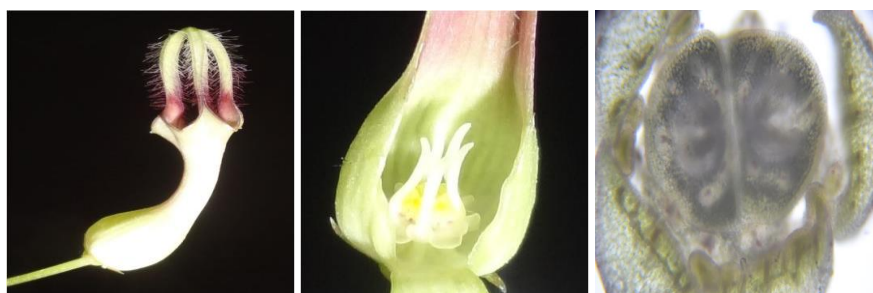


Fig.2. Flower (10X)

Open flower (20X)

T.S. of Ovary (40X)

Conclusion

Plant anatomy is a very important tool in study of plant systematic. Our objective was to develop a system of classifying plants in a way that all the similarities and dissimilarities are set out in sequential order. Microscopical studies have provided valuable details for identification and authentication. Plant morphology and anatomy may help to understand basic aspects as new approaches need to correct a past draw back. Anatomical description may serve as a conceptual framework to link with other approaches, thus this approach may be seen as an integrated and continuous process that goes through reference points. The study might contribute to enhance interest of researchers in developing a protocol towards taxonomic and anatomical resolutions. All the plants under study are categorized as endangered and threatened so they require utmost attention and care towards conservation.

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