

Botanical standardization of some Solanaceae members

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Abstract

Background/Objectives: Some of the anatomical characters are liable to become modification in response to environmental change; however, many characters are more fixed genetically and such characters are to be employed for taxonomic studies. For distinguishing two species, differences in the anatomical structures usually provide much reliable grounds for identification.

Methods/Statistical Analysis: For anatomical studies the customary techniques of sectioning and staining will be followed. It is aimed to collect data of anatomical characters of easily available plant parts such as leaf, petiole, stem and root. It is hoped that these studies will enable anybody to identify the plants concerned in the sterile or fragmentary conditions.

Findings: Anatomical structure is most likely to provide evidence concerning the interrelationships to families or in establishing the real affinities of genera of uncertain taxonomic status.

Application/Improvements: Microscopical studies are also very essential tool to establish the botanical identity of commercial samples of medicinal value and may also aid in checking adulteration or substitution. Apart from anatomical studies, palynological investigation is also to be attempted. This will give additional information both for diagnosis of the plants and for confirmation of their taxonomic position.

Keywords: *Capsicum frutescens* L., *Cestrum diurnum* L., *Physalis minima* L., Trichomes, Palynology.

1. Introduction

Several members of Solanaceae are known as vegetables such as *Capsicum*, *Physalis*, *Solanum tuberosum*. Apart from afore said popular members of Solanaceae, there are several other weed taxa of this order. Metcalfe and Chalk (1950) in their monograph of systematic anatomy of dicotyledons have enumerated the anatomical features of various vegetative organs of the members of Solanaceae [1]. Calcium oxalate crystals occur in stem, petiole and leaf in different forms including crystal-sand whereas, *cestrum* has said to lack the crystals. Stomatal types are said to be mostly anomocytic and anisocytic and diacytic types also occur in certain taxa. Certain characters of the veins are of confirmatory value in the identification of species and genera. This petiole has shallow or deep arc or U-shaped vascular strand intraxylary phloem in the form of continuous or separate strands accompanied by sclerenchyma cells is universally present at the periphery of the pith. On the basis of the fact that plants exhibit a high degree of physiological, morphological and anatomical plasticity to changes in spectral quality, they aimed to compare anatomical features of leaf and stem of capsicum grown under LEDS with different spectral regimes to evaluate the use of LEDs for plant growth in future space-based plant research chambers or bioregenerative life support systems. The results of their study showed that in plants grown under the broad spectrum MH lamp have greatest cross-sectional area of the stems, thickness of secondary xylem, number of intraxylary strands, leaf thickness, number of chloroplasts and thickness of strands, leaf thickness, number of chloroplasts and thickness of palisade spongy mesophyll tissues. This fact prompted us to take taxonomical and anatomical studies of some common weeds of Solanaceae which are of high medicinal values. The present study combines both systematic anatomy and pharmacognosy of a few selected taxa.

2. Materials and Methods

Three species belonging to three different genera of Solanaceae were selected for the present study. Table 1 provides the details of taxa studied and other aspects of the species. To begin with, taxonomic studies of all three taxa were undertaken. Plant specimens were collected from different localities within Chennai and its

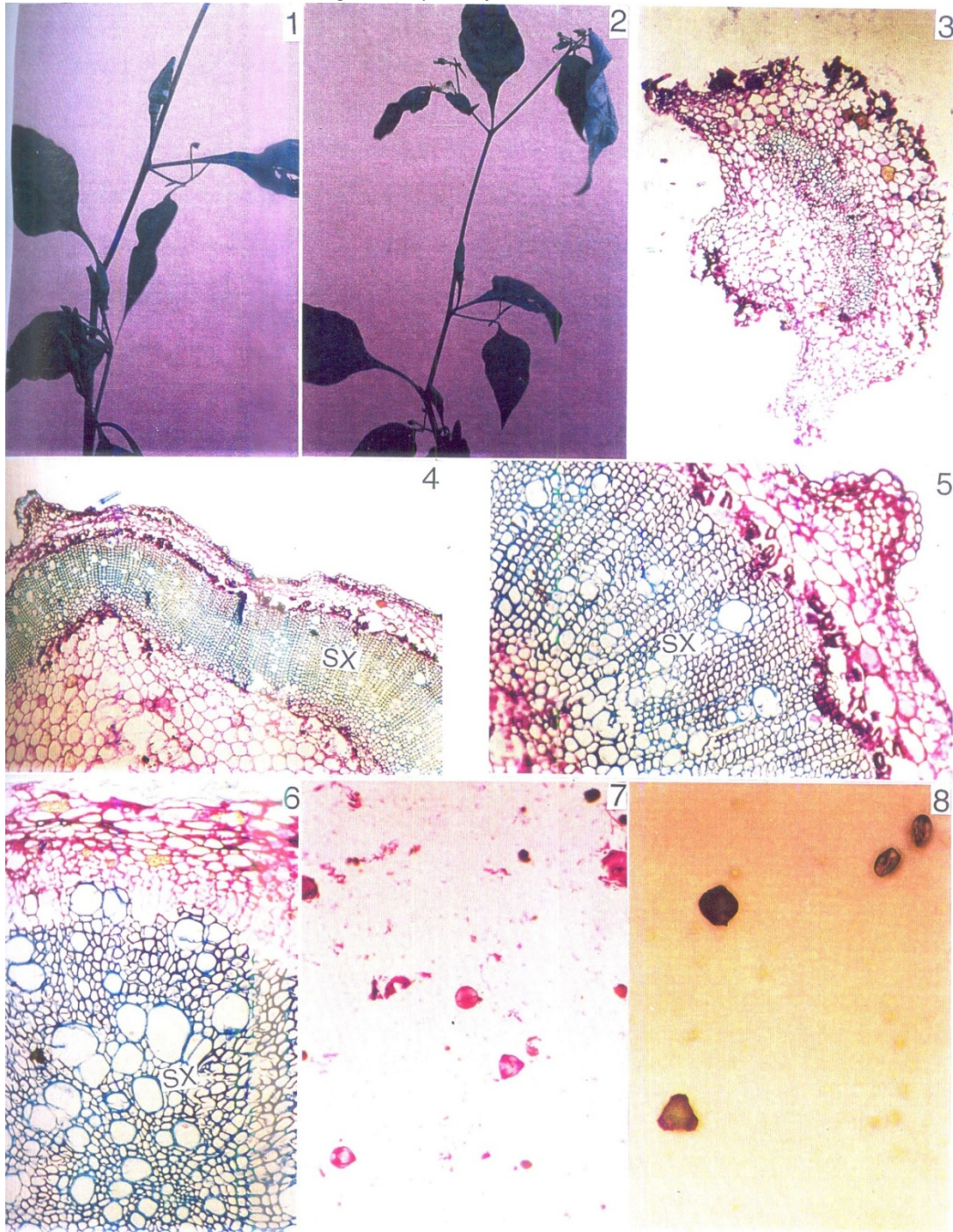
neighbourhood. Data pertaining to both vegetative and floral characters were obtained from fresh specimens. The specimens were also compared with the herbarium materials preserved in the Presidency College, Chennai. Taxonomic descriptions were followed as per the method of standard books [2] [3]. The materials were collected from the natural habitats wherever possible. Small portions of properly trimmed leaf, petiole, inter node (mostly thin) and root (tap root or lateral root) were cut with the help of sharp blade directly in the field and fixed in FAA (Formalin, Acetic Acid and Alcohol). For epidermal peeling of upper and lower sides of the leaf, pulling with forceps was found to be satisfactory. The epidermal peelings so removed from the leaf were immediately soaked in a stain on the slide, rinsed after sometimes and mounted in a drop of glycerine for microscopic observations. Partial maceration by employing Jeffrey's fluid was also followed wherever handpulling did not work out. For leaf, petiole and stem paraffin embedded rotary microtome method of sectioning was followed. The materials fixed in FAA were washed in water and dehydrated, cleared wax infiltrated by passing through graded series of ethyl alcohol and xylene [4]. Leaf and petiole sections were cut at 8-10 μm thickness, stem sections cut at 10-12 μm thickness [5-7]. Microtome sections were stained with tannic acid ferric chloride and safranin [8] or Toluidine blue [9]. Epidermal peelings were stained with either Toluidine blue or safranin. Pollen grains were collected from unopened flowers and observed directly after staining with crystal violet. The pollen grains were also acetalysed and studied. Measurements of stomata were based on 25 readings and range with mean values was given. Other readings such as stomatal frequency pollen diameter etc were also computed on 25 readings. Photomicrographs were taken with NIKON-Lab Phot-II Microscope. Identification of plants was done with the help of different Floras guides [2, 3, 10-13].

Name of the plants	Month & Year of collection	Place of collection	Period of flowering
<i>Capsicum frutescens</i> L.	November 2017	Pattabiram	Sep. to December
<i>Cestrum diurnum</i> L.	August 2017	Gandhi Nagar, Avadi	August to December
<i>Physalis minima</i> L.	August 2017	West Tambaram	August to November

3. Observations

***Capsicum frutescens* L.** (Figure 1) – A soft, stemmed shrub up to 2 m. high. **Stems:** somewhat angular and grooved, \pm pubescent; **Leaves:** simple, alternate, elliptic-ovate, 5-10 x 4-6 cm, thin-charactaceous, lateral nerves 6 pairs, puberulous along nerves, base obliquely rounded to acute, margin entire, apex gradually acuminate; petiole 3 cm long; **Inflorescence:** Axillary cyme; **Flowers:** Bisexual, actinomorphic, pentamerous, hypogynous, 2 at each node, sometimes 1, pedicel to 1.5 cm; **Calyx:** Sepals 5, gamosepalous, copular, 4 mm long, triangular, acute, little enlarged in fruit; **Corolla:** Petals 5, gamopetalous, triangular ovate, wheel shaped, 6 mm long, acute; **Androecium:** Stamens 5, epipetalous, alternipetalous, anthers oblong, 2.5 mm long; **Gynoecium:** Bicarpellary, syncarpous, superior ovary, 1.5 mm, style 4 mm long, stigma capitate; **Fruit:** Berry, elongate, cylindrical; **Seeds:** Compressed, circular. **Anatomy** (Figure 1) **Leaf:** upper epidermis stomatiferous, stomatal type anomocytic; stomatal frequency 15-25 (21) / mm^2 ; stomatal size 24-34 (31) μm x 18 x 24 (21) μm . Epidermal cells lobed radial walls sinuous. **Lower epidermis** stomatiferous, stomatal type anomocytic; stomatal frequency 30 – 45 (37) / mm^2 ; stomatal size 30-36 (30) μm x 20-22 (20) μm . Epidermal cells lobed, radial walls highly sinuous, thin. **Petiole:** bowl-shaped in CS view with prominent lateral wings and stout conical adaxial projection. Surface even and glabrous or sparsely hairy. Ground tissue differentiated in to outer narrow collenchymas, rest of the tissue being parenchymatous. Vascular strand consists of uniseriate radial lines of xylem elements forming a shallow arc with outer and inner phloem and crystal bearing cells common. **Stem:** Diameter of the stem 3.2 mm; CS view circular with longitudinal blunt ribs. Epidermis unistratose, cortex narrows with outer chlorenchyma layer and inner parenchyma zone. Secondary phloem narrow. Cortical fibres thin layered and continuous. Secondary xylem solid hollow cylinder of fibres and vessels. Fibres fairly thick walled, arranged in regular radial files. Vessels less in frequency, solitary, circular, thin walled up to 50 μm in diameter. Pith wide, homogenous and parenchymatous. Pith phloem occurs in small nests. Pith fibres occur in small nests. **Roots:** Lateral root of 3 mm diameter was studied. There is a thin periderm followed by a narrow zone of cortex. Secondary phloem elements occur in narrow band of radially arranged files. Secondary xylem thick, dense and solid cylinder of fibres and vessels. Fibres wide lumened and thin walled. Vessels numerous, circular, solitary or in multiples; thin walled, diameter up to 100 μm .

Figure 1. *Capsicum frutescens* L.



1 & 2: Fruiting and Flowering shoots; 3: Petiole T.S. (Winged); 4 & 5: Stem T.S. (Ridged); 6: Root T.S. (Dense xylem); 7 & 8: Pollen grains (Sx-Secondary Xylem)

***Cestrum diurnum* L.** (Figure 2) – An ornamental shrub, 2 m. tall; **Leaves:** simple, entire, oblanceolate oblong, 8-15 x 3 – 5 cm., base rounded to acute, apex acute, petiole to 1 cm; **Inflorescence:** Axillary Panicles; **Flowers:** Bisexual, pentamerous, complete, actinomorphic, hypogynous; **Calyx:** Sepals 5, gamopetalous, tube like, 1.5 cm long, reflexed at the tip, obtuse; **Androecium:** **Stamens** 5, filaments basally adnate to the tube, 2 mm long, anthers oblong, 1 mm long; **Gynoecium:** Bicarpellary, syncarpous, bilocular, superior ovary, ovules many attached to the axile placentation, ovary globose, 1.5 mm across, stigma capitate; **Fruit:** A globose berry, black or white.

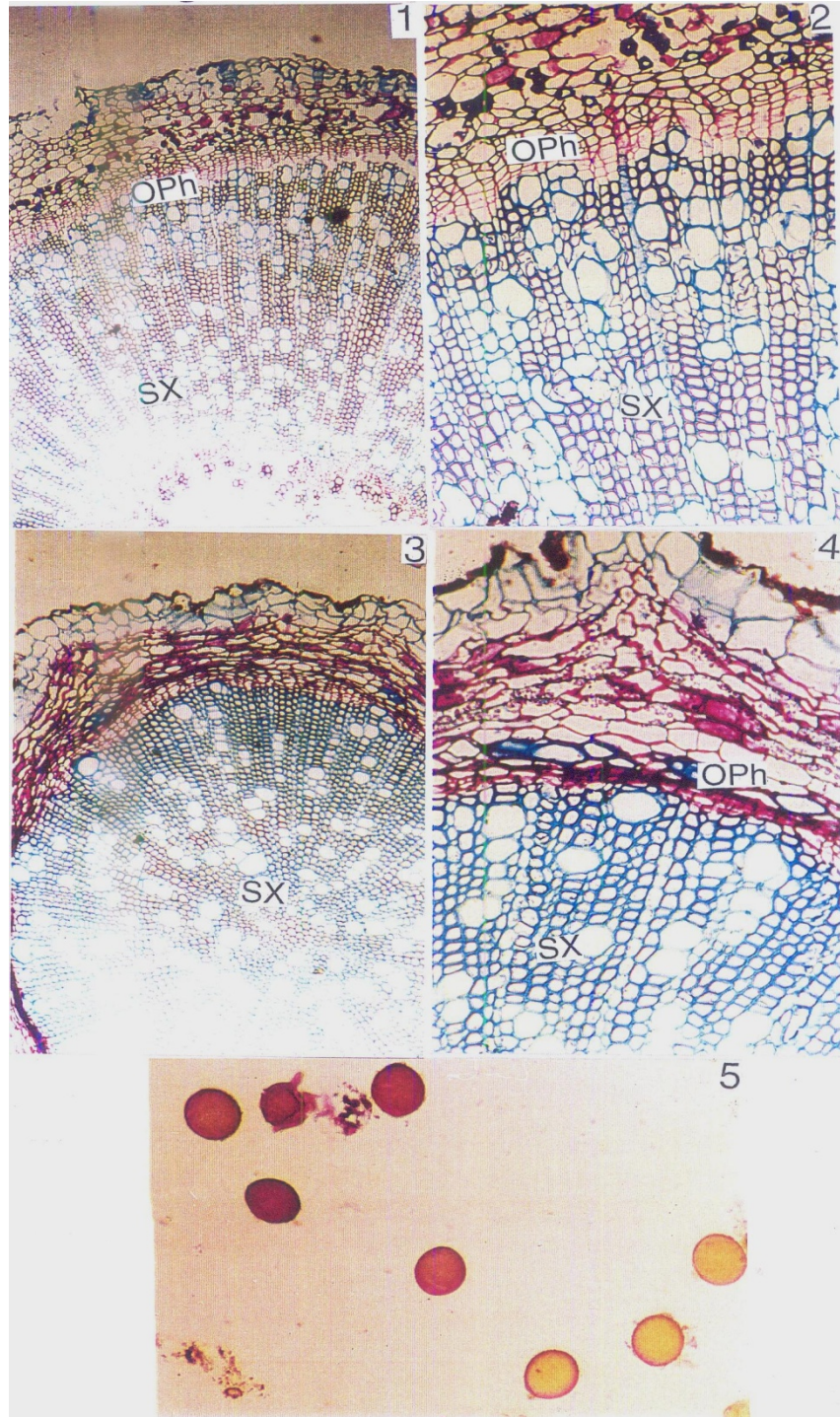
Figure 2. *Cestrum diurnum* L.

1 - 3: Flowering shoots; 4: Petiolar vasculature; 5: Abaxial stomatiferous epidermis;
6: Adaxial nonstomatiferous epidermis

Anatomy (Figure 3) – **Leaf:** upper epidermis non stomatiferous, cells lobed, radial walls wavy and fairly thick. Lower epidermis-stomatiferous, stomatal type mostly anomocytic, stomatal frequency 15-25 (23); stomatal size 36-50 (44) μm x 24-30 (26) μm . Epidermal cells lobed and irregular, radial walls sinuous and thin. **Petiole:** 2 mm in diameter, cross-sectional shape-plano-convex with flat adaxial side, the central part on the adaxial being raised slightly. Surface even glabrous; epidermis unistratose, epidermal cells, cubical or barrel shaped; cuticle thin. Ground tissue homogeneous, parenchymatous, thin walled, less compact, inclusions not evident. Vascular system consists of a shallow crescent shaped rows of xylem elements with small nests of phloem elements both on the outer and inner sides of the xylem arc, isolated, solitary or groups of 2-3 sclerenchyma elements are seen towards the concavity of the vascular arc very small accessory strands, are on either side of adaxial part are

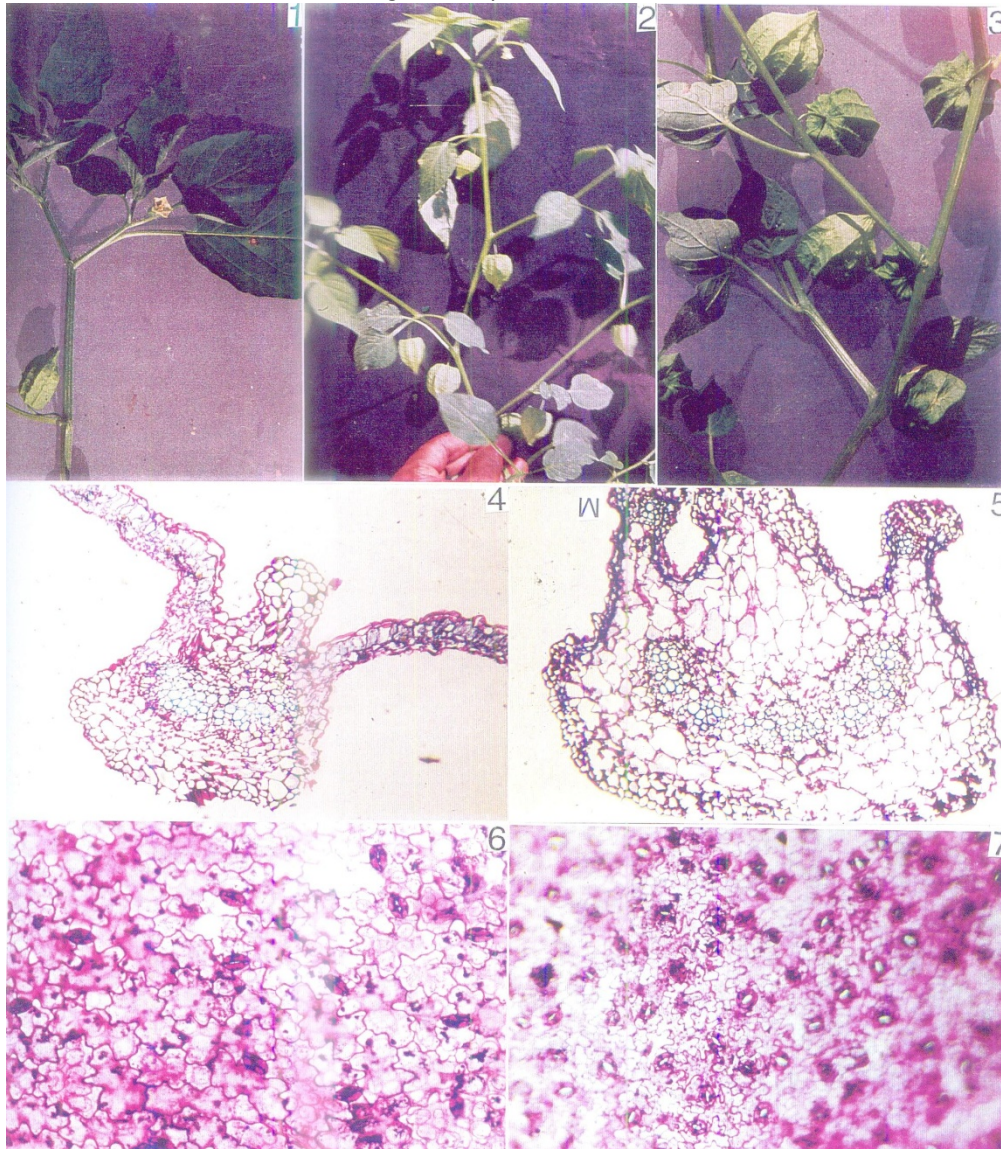
seen. **Stem:** Diameter of the specimen examined 0.5 cm; circular and even in CS outline. Specimen exhibits well developed vascular tissues of secondary origin. Epidermis intact at certain places, but more frequently replaced by a narrow zone of 4 layered periderm and shallow fissures. Cortex narrow, 4-6 layers of parenchyma cells, compact and thin walled, delimited internally by a thin continuous layer of fibres. Secondary phloem consists of radial files of tangentially oblong sieve elements with lateral companion cells, phloem parenchyma and rays. Secondary xylem is broad and dense; vessels occur in radial rows; xylem rays thin and straight. Vessel elements angular, thick walled, about 30 µm in diameter, ground tissue libriform fibres. Medullary phloem occurs in small nests along the outer boundary of the pith; small groups of pith fibres are seen intermixed with the medullary phloem nests. Pith narrow lent district, parenchymatous, thin walled.

Figure 3. *Cestrum diurnum* L.



1 & 2-Stem T.S; 3 & 4-Root T.S; 5-Pollen grains (OPh: Outer Phloem; SX-Secondary Xylem)

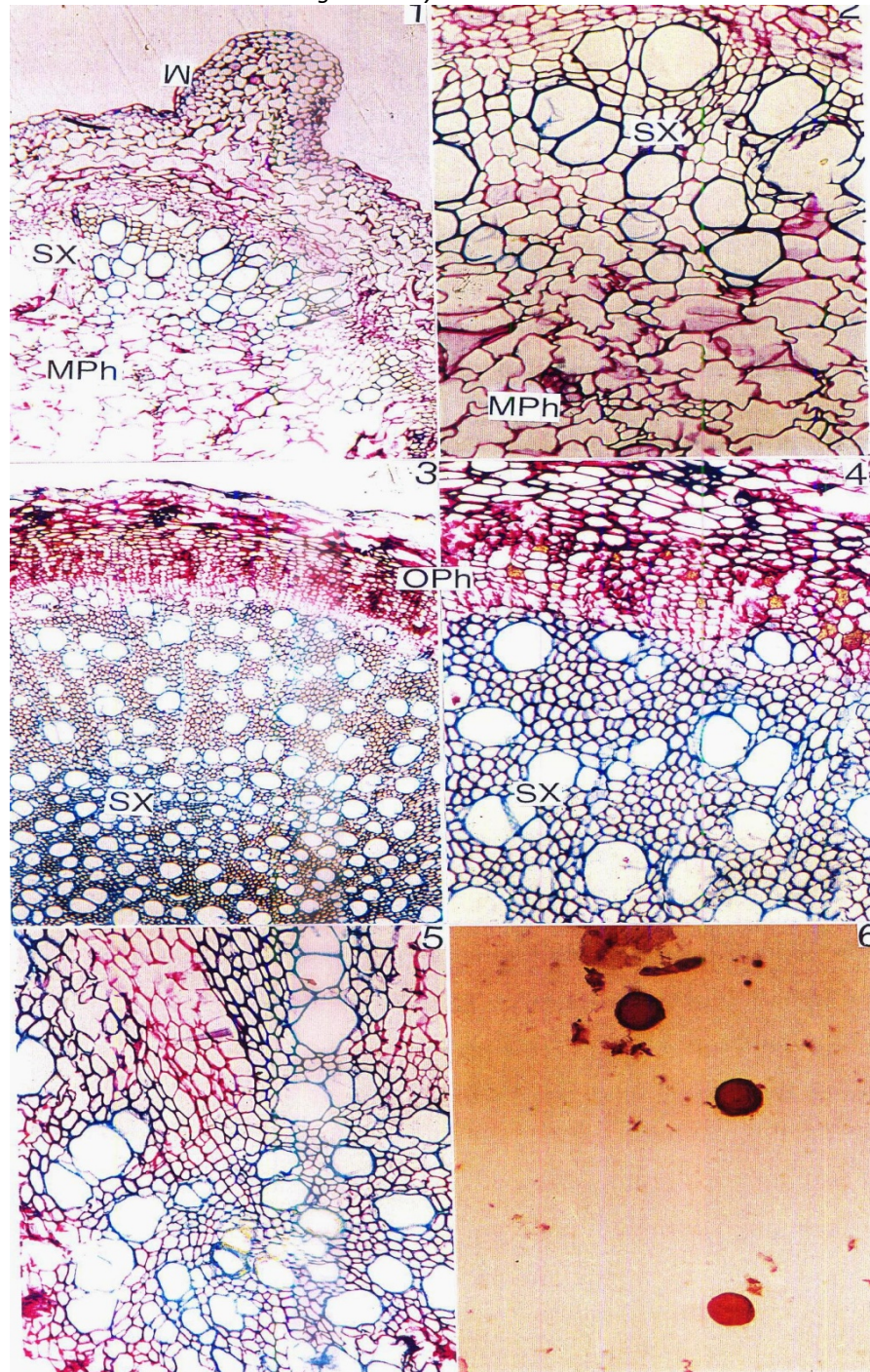
Root: Diameter of the root studied 2 mm; lateral root; mature root with thick secondary xylem cylinder; Epidermis replaced by 2-4 layers of periderm of thin walled suberised cells; Periderm followed by a narrow zone of 4 or 5 layers of cortex with tangentially oblong parenchyma cells. Starch grains are fairly abundant in the cortical cells. The inner boundary of the cortical cells is marked by small, isolated groups of fibres. Secondary phloem zone is narrow with about 4 layers of cells. Secondary xylem cylinder is wide and prominent, dense with angular, thick walled vessels. Occurring in radial multiples or solitary and thick walled libriform fibres bearing starch grains. Pith less evident, diameter of vessel elements 40-80 μm . ***Physalis minima* L.** (Figure 4)- A herbaceous pubescent annual, 15-30 cm high; **Leaves:** simple, alternate, 6 cm long, ovate, base obliquely rounded to acute, margin coarsely toothed, apex acute; petiole 5 cm long; **Flower:** solitary, axillary, clear yellow, 1 cm long, bisexual, actinomorphic complete, pentamerous, hypogynous; **Calyx:** sepals 5, gamosepalous, valvate, triangular, 3.5 mm, triose pubescent without, basally connate, acute; **Corolla:** Petals 5, gamopetalous, valvate, triangular ovate, 4 mm, prominently nerved, connate from the middle, acute; **Androecium:** stamens 5, unequal, epipetalous, filaments 2-4 mm anthers oblong, 2.5 mm; **Gynoecium:** Bicarpellary, bilocular, syncarpous, superior ovary, 1.5 mm, style 4 mm, ovules many on axile placentation, stigma capitate; **Fruit:** Berry 0.7 mm across, accrescent calyx 1.5 cm long.

Figure 4. *Physalis minima* L.

1-3: Flowering and fruiting shoots; 4: Leaf mid-rib T.S.; 5: Petiole T.S.; 6: Adaxial stomata; 7: Abaxial stomata

Anatomy (Figure 5) – **Leaf:** lower epidermis stomatiferous, stomatal frequency 48 / mm²; stomatal type anomocytic; size of the guard cells 25 x 30 μm; abnormal stomatal types, such as stomata with one guard cells, narrow and thin stomata, are quite frequent; epidermal cells with lobed and wavy tangential walls; cells are also much lobed and irregular in shape. Epidermal trichomes absent. Upper epidermis stomatiferous, stomatal frequency 5-3.5/mm²; stomatal type anomocytic; size of the guard cells 32 x 35 μm; abnormal stomatal types less common; epidermal cells lobed with wavy tangential walls; spherical chloroplasts, one too many per cell, are frequent in the epidermal cells; trichomes sparse, restricted to veins; 2-4 celled, uniseriated, unbranched, pointed, non-glandular. **Petiole:** Dorsiventrally differentiated into abaxial hemispherical part and adaxial three-ridged part. Horizontally 2 mm and vertically 1.8 mm thick. The adaxial ridges are quite prominent; two lateral ridges are hemispherical and the middle one is conical / pyramidal in CS view; trichomes are aggregated at the summit of the ridges.

Figure 5. *Physalis minima* L.



1 & 2: Stem T.S.; 3 - 5: Root T.S.; 6: Pollen grains (MPh – Medullary Phloem; OPh – Outer Phloem; SX – Secondary Xylem; W - Wing)

Epidermis: unistratose, cells cubical, cuticle thin; subepidermal zone collenchymatous, 2-3 layered, angular type; collenchymas zone followed by 3-4 layers of wider, thin walled, compact parenchyma cells. Collenchyma zone is interrupted by small patches of chlorenchyma, especially along the ferrous of the adaxial side. Vascular system consists of an arc of three bicollateral strands; the median strand is smaller than the lateral ones; the hemispherical lateral ridges also have small riles traces, one in each strand. Xylem elements small, thick walled, angular and occur in distinct short radial short rows; phloem elements occur both on the centre and inner sides of the vascular strands; elements are in small groups. **Stem:** Diameter of the stem-0.5 cm; cross sectional outline wavy and three-winged; wings stout and rounded; surface glabrous. Central core of the stem occupied by a wide lysigenous cavity surrounded by persistent of outer pith cells. The thickness of stem portion outer to the pith canal is about 1 mm. **Epidermis:** Unistratose, cells narrowly tubular, thin walled; epidermis is followed by four layers of collenchyma which extends and occupies the entire wings of the stem. Collenchyma portion is followed by a narrow zone compressed cortical parenchyma. Vascular system consists of thin cylinder of xylem and phloem produced by limited activity of the vascular cambium. The xylem tissue is massive especially beneath the wings consisting of wide, thin walled, angular vessel elements. Diameter of the vessel elements 80 μm . Medullary phloem occurs in small nests along the periphery of the pith cavity. The crystal sand is not evident. Root material studied, lateral root, 3 mm thick. Periderm narrow and thin; cortex collapsed due to secondary growth. Cortical fibres are found as small groups at isolated places. Secondary phloem narrow and continuous around the xylem cylinder. Phloem elements are arranged in radial rows. Xylem forms the thick solid core of the root. The central part of the secondary is denser with thick walled fibres and narrow vessels. The outer secondary xylem has thinner fibres and wider vessels. Vessels are mostly solitary, circular or oval, thin walled upto 60 μm diameters.

4. Discussion

Three unrelated species belonging to three different genera of Solanaceae were selected for the present study [14]. An attempt will be made to prepare an anatomical and palynological data obtained during the present investigations. Simplified taxonomic details will also be provided with the help of characters of both vegetative and floral organs. In the present study certain interesting aspects have come into light which will be discussed in connection with the before said view points of the three species studied, *Cestrum* as merits due consideration, because it's certain unique features. Diameter of the pollen of *Capsicum frutescens* 38 μm , tricolpate, exine smooth, triangular in shape in equatorial view, circular in polar view, colpa broadly elliptical in surface view. Characteristic features of *Cestrum diurnum* pollen 38 μm , monocolpate, colpatriad, exine smooth, spherical in shape both in polar and equatorial views. Whereas diameter of *Physalis minima* pollen 20 μm in equatorial plane, 26 μm in polar plane. Monocolpate, triradite, exine thin and smooth, shape nearby triangular in equatorial view, elliptical in polar view. Inamdar and Murthy (1981) investigated the diverse forms of vein endings in *Capsicum* species and *Cestrum* species [15]. They distinguished free vein endings, isolated vein endings and isolated tracheids. The vein endings are multiseriate in *Cestrum* species. Tracheids at the veins endings were reported to vary in size, shape and orientation. Isolated vein-endings and isolated tracheids were noticed in *Cestrum* species. They stressed the taxonomic significance of these structures. It was found in the present study that the leaves of *Cestrum* are hypostomatic; the epidermal cells of the leaf also have straight anticlinal walls and polyhedral epidermal cells. These features are at variance with those observed in other species studied. The placement of *Cestrum* in a distinct series Cestreae by Wettstein is justified by these anatomical features. Barring *Cestrum diurnum*, all other species studied at present have sinuous anticlinal walls of the epidermal cells. It is also indicative of the environmental niche which these taxa choose to develop. Occurrence of stomata on both surfaces of the leaves (amphistomatic condition) seems to be predominant in the taxa studied with the only exception of *Cestrum*, where the leaves are hypostomatic. The preponderance of amphistomatic leaves is again an indication of the phylogenetic affinities of the taxa. The functional significance of amphistomatic condition is not definitely known. In the present study, the petiole was found to be planoconvex shape with lateral wings was observed in *Capsicum* and *Physalis*. The ground tissue is invariably differentiated into outer collenchyma zone in all the taxa investigated excepting *Cestrum*. The vascular system

may be a shallow arc or deep bowl shaped with discrete or coalesced vascular strands. Accessory strands are found in all species studied.

The variation met with the cross sectional shape and vascular system offer useful diagnostic features of the taxa [16]. The cross sectional outline of young stem differs significantly in the species studied. The cross sectional shape is even in *Cestrum* and angular in *Physalis minima*. These cross sectional configurations are easy access for identification of fragmentary stem materials.

5. Conclusion

The present study of three different genera belonging to Solanaceae members attempts with special reference to taxonomic, anatomical and palynological studies. These microscopical studies reveal the best clue to identify three unrelated plants even in the fragmentary samples. Each species is having specific character both morphological and anatomical features. To cite a few among the many, corolla sub rotate, stamens exerted, ripe fruit red in *Capsicum*; whereas, corolla salver form, stamens included, ripe fruit black in *Cestrum*. The plant *Physalis* pertain calyx persistent and accrescent, petiole with axial ridge and lateral wings. The results have enabled us to identify accurately on the basis of taxonomy, anatomy and palynology of the taxa of the species studied. On the basis of data retrieved from the present investigation, it is advocated that the three species may be treated as independent taxa.

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