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COMPARATIVE PHARMACOGNOSTICAL STUDIES OF ORIGINAL TAXA WITH SUBSTITUENT USED IN AYURVEDIC DRUG KAKANASA

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ABSTRACT

Kakanasa an ayurvedic drug prescribed for immune resistance by ayurvedic medicaments. This drug acts as immune modulator and it is described as a controversial drug in many literatures. we attempted for the causes of controversy and investigated pharmacognostical studies of original taxa *Asclepias curassavica* L. used in *Kakanasa* with its adulterant taxa, *Pentatropis capensis* (L. f.) Bullock. To address this issue, data related to the prevailing botanicals was gathered from herbalists, specimen collectors and herbal vendors. Results of comparative pharmacognostical studies and anatomical characterization revealed that *Kakanasa* is adulterated in local herbal markets by different herbal drug manufacturing units and herbal vendors without the originality. We hereby recommend that investigations in prevailing authenticated botanicals are necessary to work out the contribution of the raw drug trade.

INTRODUCTION

A fair idea of the prevailing authenticated botanicals is necessary to work out the contribution of the raw drug trade. To address this issue, data related to the prevailing botanicals was gathered from two sources i.e. (i) the botanicals quoted in the 'mandis' and (ii) the procurement prices paid by different herbal manufacturing units. Survey and analysis of data also brought out price trends in respect of some entities along with prominent factors that influence the prices of botanicals.

Quality and purity of the raw drugs does seem to have an impact on their prices. A number of these botanicals are traded in markets at different levels of grading and their different grades command different prices. In our investigation we have cited an example *Asclepias currasavica* L.(*Apocynaceae*) used in the preparation of ayurvedic drug 'kakanasa' is adulterd with *Pentatropis capensis* (L. f.) Bullock (*Apocynaceae*), *Trichosanthes cucumerina* L (Cucurbitaceae), *Dicliptera paniculata* (*Acanthaceae*) and *Martyniaannua* L.(*Martyniaceae*). From morphologic point of view some of these taxa are similar and one can be used as a substitute for the other by the drug dealers.In this paper we have undertaken the comparative pharmacognostical investigation for *Asclepias currasavica* (Figure A) with its adulterant taxa *Pentatropis capensis* (Figure B).

Pharmacognostical Studies

The crude drugs can be identified mainly on the basis of their morphology, macroscopic and microscopic characters. Macroscopic characters are also known as organoleptic characters. It refers to the evaluation of crude drugs by colour, odour, taste, size, shape and special features like texture and fracture etc. The microscopic characters reveal the anatomy of root, stem, bark, leaf, petiole, flower, fruit and trichomes etc. Microscopical examination and characterization of phytodrug were very essential in the pharmacognostic studies. Botanical identity of the phytodrug is an important prerequisite for understanding the analysis of medicinal properties of any plant. If the plant identity of the drug is incorrect, the entire work on the plant becomes invalid. Hence, more care is taken that botanical identity of a crude drug is the threshold in the process of biological investigation. The research should be equipped with all possible diagnostic parameters of the plant on which the researcher plan to work. Investigations in prevailing authenticated botanicals are necessary to work out the contribution of the raw drug trade. Results of comparative pharmacognostical studies and anatomical characterization reveal original and adulterant taxa.

Research Article

MATERIALS AND METHODS

Physicochemical Studies

The whole plant coarse powder (Leaf, root and stem in 1:1:1 ratio) of two selected taxa were used for physicochemical studies as per ethnic healers formulations.

Microtomy

Healthy whole plant (root, stem, petiole, leaf) were cut and removed from the plant and fixed in the field immediately in FAA (Formalin-5ml + Acetic acid – 5ml + 70% ethylalcohol – 90 ml). After 24 hours of fixing, the different organs were dehydrated with graded series of tertiary-butyl alcohol as per the procedures (Sass, 1940). After dehydration, infiltrations of the organs were carried out with gradual addition of paraffin wax (Melting point 58-60°C) until tertiary-butyl alcohol solution attained super saturation. The plant organs were cast into paraffin blocks.



Figure 1: *Asclepias curassavica*



Figure 2: *Pentatropis capensis*

The paraffin embedded organs were sectioned with the help of rotary microtome. The thickness of the sections maintained to be between 10-12 μm . The sections were stained with toluidine blue as per the method of O'Brien *et al.*, 1964. Wherever necessary sections were also stained with saffranin and fast-green and potassium iodide for starch to stain blue colour, 1% solution of ferric chloride give light blue or black colour to the tannins (Ashokan, 2006). Paraffin embedded lamina were used for paradermal sections. From these sections, the epidermal layers as well as vein-islets and vein-terminations were studied. The alternative method was clearing the leaf fragments by immersing the material in warm alcohol (to remove chlorophyll) followed by treating with 5-10% sodium hydroxide. Finally, the materials were rendered transparent due to loss of cell contents. The cleaned materials were washed thoroughly, stained with saffranin for further studies. Microscopic descriptions of all necessary cells and tissues are supplemented with photomicrographs and measurements are given. Photographs of different magnifications were taken with Nikon Lab Photo-2 microscopic unit. For normal histological observations, sections were photographed under bright field light. For the study of crystals, starch grains and lignified cells, polarized light was employed. Since these structures have bi-refrangent property, under polarized light they appear bright against dark background. Magnifications of the Figures are indicated by the scale-bars. Descriptive terms of the anatomical features were used as per the terminology found in standard anatomical books of Easu (2007) and Fahn, (1990).

Research Article

Powder Microscopy

Powder microscopic studies were carried out by examining the powder of the plant samples as well as by macerating the plant samples using Jeffery's maceration fluid (Johansen, 1940; Sass, 1940). The separated vessel members (vessel elements) and fibres were used to study the lateral wall characteristics and dimensional variations. Physicochemical parameters like ash values (total ash, water soluble ash, acid soluble ash and alkalinity of water soluble ash), extractive values (ethanol soluble, water soluble, hexane soluble and chloroform soluble extracts) and solubility values (ethanol, water and methanol) were determined according to the standard procedures recommended as per WHO guidelines (Anonymous, 2009&1996) and Kokate,(2008). Fluorescence analysis of the whole plant powder drugs was carried out according to the methods followed by Chase and Pratt (1949), Kokoski *et al.*, (1958).

1. Anatomical characterization of *Asclepias currasavica*

1.1 Leaf Midrib (Figure 1.1)

The midrib is broadly convex and semicircular on the abaxial side and shallow wide concavity on the adaxial side. The epidermal layer is thin and the cells are small, thick walled and squarish in shape. The ground tissue is homogeneous and parenchymatous; the cells are circular to angular and compact. The vascular strand is broadly are shaped and wide and thick. It is bicollateral. The vascular are is 600 μm thick and 1 mm wide. The vascular strand is 500 μm thick wide and 160 μm thick. It includes short, narrow and compact row of xylem elements. The xylem elements are narrow, angular and thick walled. Phloem elements are in small isolated islands distributed in a line on the adaxial and abaxial portions of the xylem (Figure 1.1).

1.2 Leaf margin (Figure 1.2)

The marginal part of the lamina is blunt, semicircular and 200 μm thick. The epidermal cells of the marginal portion are smaller thick walled and darkly stained. The subepidermal tissue of the margin includes compact mass of parenchyma cells.

1.3 Lamina

The middle part of the lamina is 250 μm thick. It consists of fairly wide spindle shaped adaxial epidermis and this, rectangular cells on the abaxial epidermis. The abaxial epidermis is stomatiferous. The mesophyll tissue includes less distinct palisade layer of cylindrical cells. The spongy mesophyll includes these vertical filaments of small spherical cells and wide air-chambers.

1.4 Epidermal cells and Stomatal type (Figure 2.1, 2)

The epidermal tissue was studied by paradermal sections and in surface view. The epidermal cells are fairly thick walled and highly wavy and the cells appear amoeboid. The stomata are anomocytic type and they do not possess distinct subsidiary cells. The guard cells are narrowly elliptical with narrow slit like stomatal pore. The guard cells are 20 x 12 μm in size. Calcium oxalate crystals of druses are sparsely distributed in the mesophyll cells. The druses are small and diffuse in distribution (Figure 2.2). They are 10 μm in diameter.

1.5 Venation pattern (Figure 3.1, 2)

The veins are thick and prominently visible; they are straight. The vein-islet's are wide with distinct vein-boundaries. The outline of the islets varies from hexagonal, polygonal to squarish shape. The vein-terminations are well expressed. They are simple, unbranched and curved. Some of the vein-terminations are branched once or twice and more common, the terminations are branched repeatedly giving rise to dendroid outline of the terminations (Figure 3.2).

1.6 Petiole (Figure 4.1, 2)

The petiole is 700 μm thick and 800 μm wide. It is circular in sectional view, with steep, v-shaped groove on the adaxial side and semicircular abaxial side (Figure 4.1). The epidermal cells are thin and the epidermal cells are small, squarish with dark cells contents. The ground tissue is homogeneous and it is uniformly parenchymatous, angular and thin walled and compact dispersed in the ground tissue are small thick walled darkly stained latifers (Figure 4.2).

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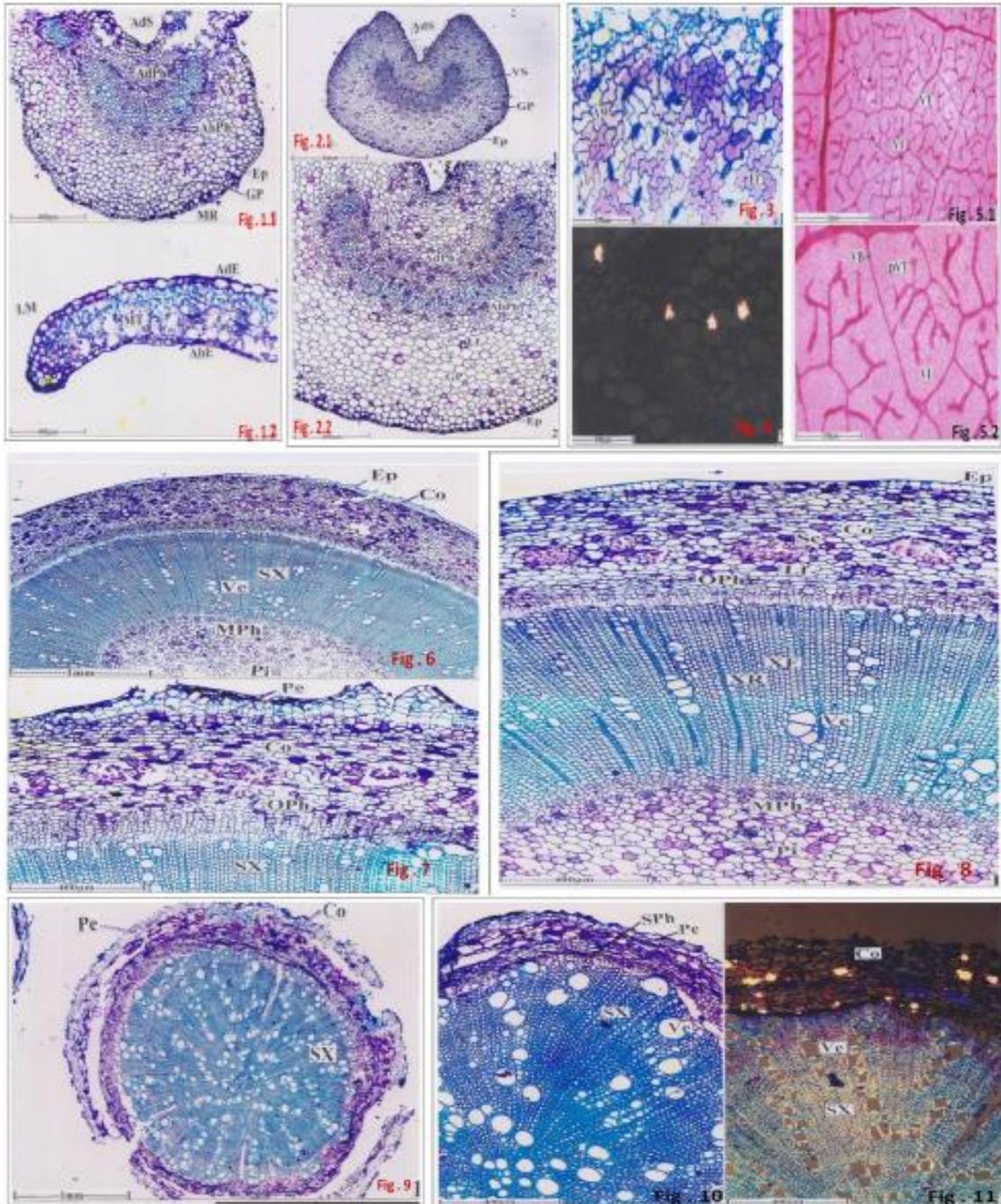


Figure 1: Anatomical characterization of *Asclepias currasavica*. Figure1.1:T.S of Midrib; Figure 1.2:T.S of Leaf margin; **Figure2.1, 2.2:** T.S of Petiole (entire view & section enlarged); **Figure 3 :** Paradermal view of epidermal cells and stomatal type. **Figure 4:**Calcium oxalate crystals in the mesophyll tissue as seen under polarized light.; **Figure 5.1** venation pattern, **5.2:** vein islet and vein terminations enlarged. **Figure 6:** TS of thin stem **Figure 7:** T.S. of thin stem enlarged; **Figure 8:** T.S. of stem showing cortex, outer & inner phloem and sec. xylem. **Figure 9:** TS of root entire view; **Figure 10:** T.S of root a sector enlarged; **Figure 11:** Crystals in the cortex.

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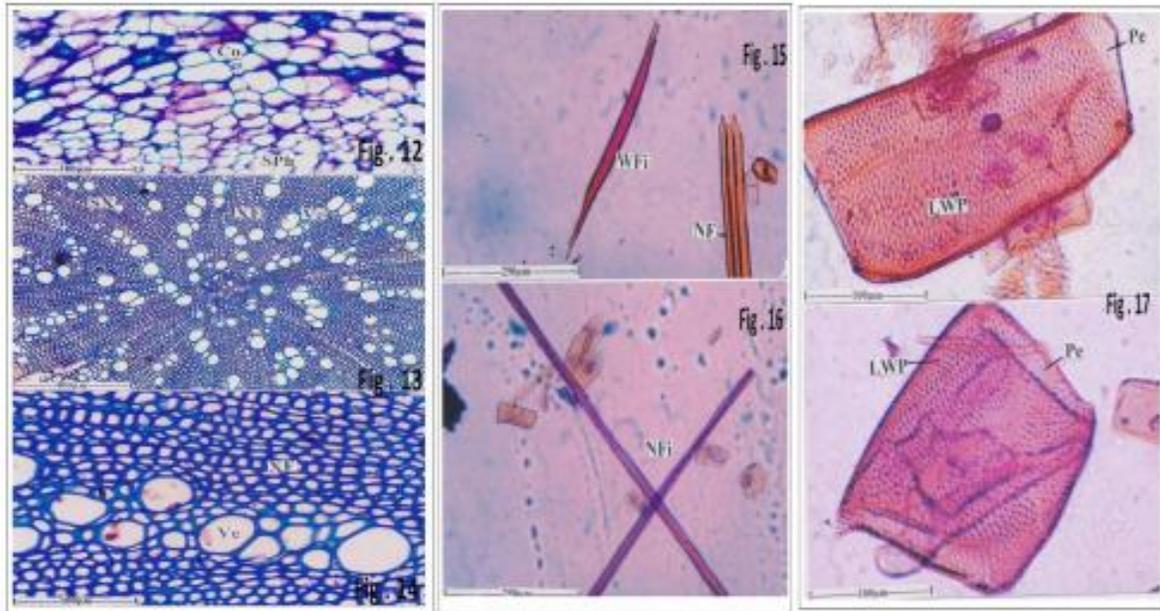


Figure 12 T.S of root phloem zone. **Figure 13** Sec. xylem along middle point. **Figure 14:** sec. xylem showing vessels and fibres. **Figure 15 & 16:** wide fibres and Narrow fibres; **Figure 17:** Vessel elements with wide perforation and dense lateral wall pits.

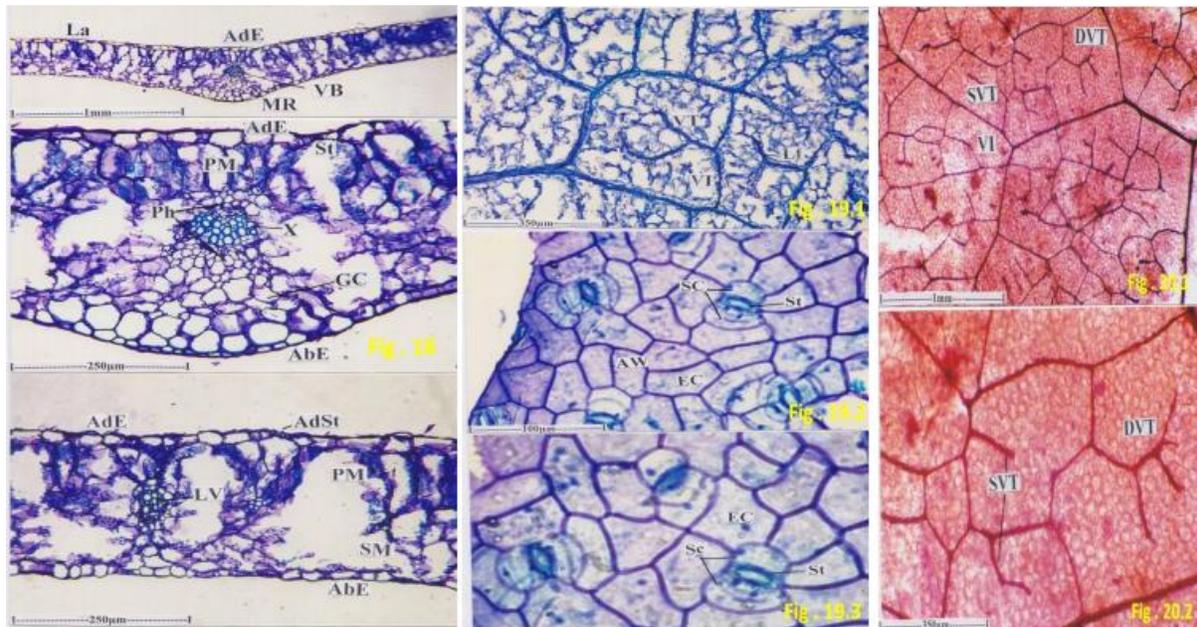


Figure18-20.2: Anatomical Sections of *Pentatropis capensis* **Figure 18:** T. S of leaf through midrib, T. S of leaf midrib enlarged, T.S of lateral veins of the lamina; **Figure 19.1:** Paradermal section of the venation; **Figure 19.2:** Paradermal section of the lamina showing stomatal type and epidermal cells; **Figure 19.3:** Paradermal section of the lamina showing stomatal type and epidermal cells; **Figure 20.1:** Lamina cleared to show the venation pattern, vein islets and vein termination; **Figure 20.2:** Lamina cleared to show the venation pattern, vein islets and vein termination

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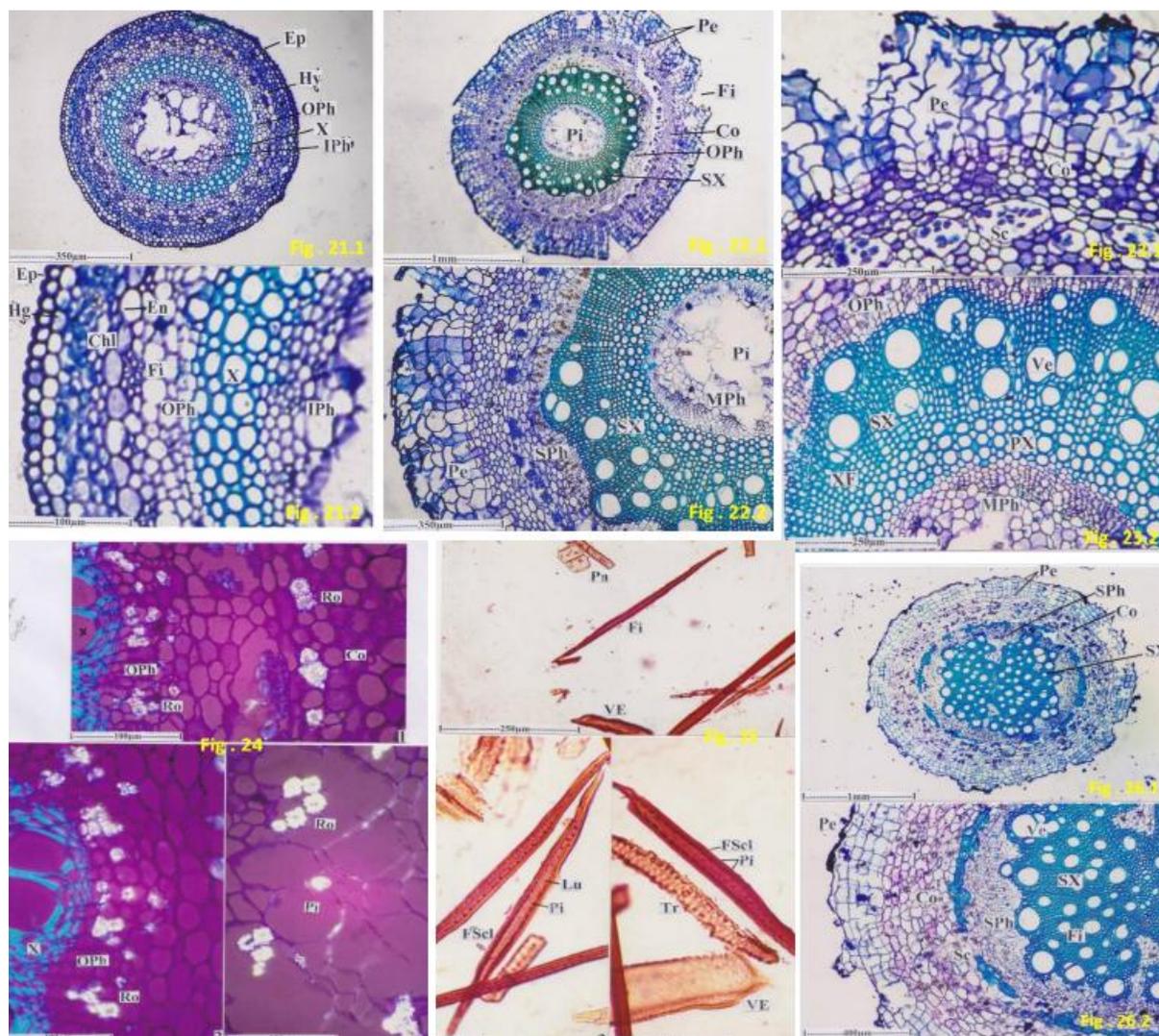


Figure 21.1-26.3: Anatomical Sections of *Pentatropis capensis* **Figure 21.1:** T. S. of petiole-entire view; **Figure 21.2:** T. S. of petiole-A sector enlarged; **Figure 22.1:** T. S of stem- entire view; **Figure 22.2:**T. S of stem- A sector, enlarged; **Figure 23.1:** T. S of stem showing periderm and vascular cylinder; **Figure 23.2:** Vascular cylinder enlarged; **Figure 24:** Rosette crystals in the cortex of the stem, Rosette crystals in the phloem of the stem, Rosette crystals in the pith of the stem; **Figure 25 :**T. S. of root- entire view, T.S of root- A sector enlarged; **Figure 26.1:** Fibres in the powder; **Figure 26.2:** Fibre sclerids; **Figure 26.3:** Fibre- tracheid, vessel element and a tracheid.

The vascular strand is wide, thick and bowl shaped. It consists with parenchymatous gaps in between the xylem rows. Phloem occurs in small discrete masses in row along the adaxial and abaxial parts of the xylem strands (Figure 2.2).

2. Stem

2.1 Thin Stem (Figure 5.1; 6)

The thin stem has thin, continuous intact epidermis, which exhibits origin of periderm at isolated region of the epidermis. The cortex is 400 µm thick. The cortical tissue includes thin parenchymatous compact cells with discrete large masses cortical fibres which are less lignified (Figure 6).

Research Article

The xylem cylinder is hollow and dense. It is 700 μm thick. It includes major portion of secondary xylem and narrow zone primary xylem found along outer periphery of the pith. Secondary xylem includes narrow straight xylem rays, radial multiples of thin walled wide angular vessels and xylem fibres. Xylem fibres are thin walled with wide cell lumen (Figure 6). Phloem occurs in continuous cylinder along the outer part of the xylem cylinder and in small circular nests in the inner border of the xylem cylinder.

2.2 Thick stem (Figure 5.2; 7.1, 2)

The thick stem has well developed periderm comprising 4-6 layers of phloem cells. The epidermis is intact with narrow fissures at frequent intervals (Figure 5.2). The cortex is 350 μm thick. It includes compact parenchyma cells and discrete masses of xylem fibres. The outer phloem is wide and consists of radial rows of small compact cells (Figure 5.2). Inner Phloem (medullary phloem) is in circular units, the cells being small and compact (Figure 7.2). The secondary xylem is thick with long, thin, parallel lines of vessels, where both narrow and wide vessels are intermixed. The vessels lines are separated from each other by wide gaps of xylem fibres (Figure 7.1, 2). The fibres are thin walled with lumen.

Root (Figure 8.1, 2, 3; 9.1, 2, 3)

The root is 2.6 mm thick; shows well developed periderm and secondary phloem and secondary xylem (Figure 8.1).

The periderm is thin and superficial and consists of three or four layers of narrow, thin walled cells. The cortex includes three to five layers of parenchyma cells including a single layer of cortical fibres (Figure 8.2). Secondary phloem consists of outer zone with collapsed phloem elements and inner zone of intact non-collapsed phloem elements (Figure 9.1).

Secondary xylem is thick, dense cylinder measuring 1.9 mm in diameter. It comprises vessels and fibres. Growth rings are absent. The vessels occur in long radial multiples or radial chains with wide gaps in between the vessel rows. The vessels occur in long radial multiples or radial chains with wide gaps in between the vessel rows. The vessels are wide or narrow, circular or angular and thin walled; they are 30-70 μm wide. The xylem fibres are thick walled and lignified (Figure 9.2, 3).

Crystals

Calcium oxalate crystals are distributed in the cortex. The crystals are druses (Figure 8.3).

Powder Microscopic Observations

Powder or macerated preparation of the plant shows mostly fibres and vessel elements. The fibres are two types:

(i) Narrow fibres (Figure 10.1, 2): The narrow fibres are long, thick walled with narrow lumen. Some of the narrow fibres have prominent simple pits on their walls (Figure 10.1). They are 400-500 μm long and 12 μm wide.

(ii) Wide fibres (Figure 10.1; 11.1, 2): The wide fibres are short and spindle shaped. The walls are thin and the lumen is wide (Figure 11.2). No pits are seen on the walls cell inclusions also absent. The wide fibres are 350 μm long and 20-30 μm wide.

Vessel elements (Figure 12.1, 2)

The vessel elements characteristically short wide and barrel shaped. They have distinct circular bordered pits which are multiseriate and dense. The perforation is simple, wide and horizontal. The vessel elements are 190-260 μm long.

Anatomical characterization of *Pentatropis capensis*

Leaf

The leaf consists of a less prominent plano convex midrib which gradually slopes into the lamina (Figure 1.1). The midrib is 400 μm thick and 500 μm wide. It consists of fairly prominent adaxial epidermis, measuring 20 μm thick cylindrical cells. The abaxial epidermis is less prominent and the cells are 10 μm thick. Small bicollateral vascular bundle placed at the central part of the midrib. The bundle consists of 5-7 vertical, short compact xylem elements which are angular and thick walled. A small cluster of phloem elements occurs at the adaxial end and wider mass of phloem occurs at the abaxial end

Research Article

of the xylem strand (Figure 1.2). Palisade cells are trans current along the adaxial and of the vascular strand. The lateral veins are also small, circular and median in position (Figure 1.3). They have circular bundle sheath parenchyma cells and small group of xylem elements with adaxial and abaxial phloem.

Lamina (Figure 2.1, 2)

The lamina is uniform in thickness with smooth and even surfaces. It is 250 μm thick. The lamina is bifacial and amphistomatic (having stomata on both adaxial and abaxial sides). The adaxial epidermal layer is thick with cylindrical cells. The abaxial epidermis is thin, the cells being narrowly rectangular. The mesophyll tissue consists of adaxial, single layer of thin palisade cells and abaxial, loosely arranged spongy parenchyma cells.

Leaf margin is semicircular and bluntly conical measuring 250 μm in thickness. The epidermal cells along the marginal part are squarish in shape and are thick walled with prominent cuticle. There is subepidermal layer or fairly large compact parenchyma cells (Figure 2.2).

Stomata

Stomata were studied in surface view of the paradermal sections of the lamina (Figure 3.1, 2). The epidermal cells are polyhedral with thick and straight anticlinal walls. Stomata occur both on the adaxial and abaxial sides. The type and density of the stomata are similar on both sides. But, the abaxial epidermal cells are slightly larger than those on the adaxial side. The stomata are paracytic type. A stomata has one parallel subsidiary cell on either side. Some stomata have two subsidiaries on one side and one on the opposite side. The guard cells are elliptic and are 10 x 20 μm in size.

Venation pattern of the lamina (Figure 4.1, 2)

The veins are thin comprising uniseriate vascular strands. The veins are straight and form fairly wide polygonal veins-islets (Figure 4.1). The vein boundaries are distinct and well defined. The vein-termination is present in almost all islets. They are either simple (unbranched) or branched once or twice. The terminations are thick and straight or curved. No terminal tracheids or sclereids are seen on the vein endings.

Petiole (Figure 5.1, 2)

The petiole is circular in sectional view and measures 650 μm in diameter. It consists of epidermis which includes circular thick walled cells with smooth prominent cuticle. There is a subepidermal layer of wider, hyaline, barrel shaped cells. Inner to the subepidermal layer of hypodermis, there is a wide zone of chlorenchyma cells. Following the chlorenchyma zone, there is a distinct layer of endodermis comprising wide and thick layer of cells. There are small clusters of fibres situated all around the inner zone of the endodermis (Figure 5.1).

The vascular cylinder is circular closed and hollow. It consists of short, parallel lines of xylem elements with phloem elements situated both on the outer and inner portions of the xylem cylinder. Pith cells are disintegrated forming pith canal (Figure 5.2).

Stem (Figure 6.1, 2; 7.1, 2)

Stem measuring 1.6 mm thick was studied. It is circular with uniformly thick, continuous layers of periderm (Figure 6.1). The periderm cells are radially oblong or squarish, thin walled and suberised. There is steep V-shaped fissures at frequent intervals. Epidermal cells remain intact at certain places of the surface. The periderm is 200 μm thick (Figure 6.2; 7.1).

Inner to the periderm is a distinct zone of parenchymatous cortex measuring 100 μm thick. The cells are small and compact.

The inner boundary of the cortex demarcated by a line of discrete masses of fibres (Figure 6.2). Vascular cylinder is a hollow cylinder of wavy outline with wide central hollow pith. The xylem cylinder consists of regular radial parallel lines of primary xylem and dense outer secondary xylem comprising wide vessels and fibres. The vessels are salivary and thin walled. The fibres are thick walled and lignified (Figure 7.2). The vessels are 30-60 μm wide.

Research Article

Phloem occurs both on the outer and inner zones of the xylem (Figure 7.2). The outer phloem is wavy continuous layer abutting the xylem cylinder (Figure. 6.2). The inner pith phloem or medullary phloem is in small isolated groups distributed all along the inner boundary of the xylem (Figure. 7.2).

Crystal distribution (Figure 8.1, 2, 3; 9.1, 2)

Calcium oxalate crystals are abundant in the stem and root. The crystals are mostly rosette type. The crystal in circular body with dark central dot or organic material and outer calcium oxalate crystal (Figure 8.1, 2, 3). In the stem throsettes are found in cortical cells as well as in the phloem cells (Figure 8.1, 2). They are also located in the outer pith cells (Figure 8.3). In the root, the rosettes are abundant in the secondary phloem and cortical parenchyma cells (Figure 9.1, 2). The crystals are up to 20 μm in diameter.

Root (Figure 10.1, 2)

The root is circular in sectional view, it is 2 mm thick. The root consists of a wide periderm which is uniform encircling the root. The periderm is 6 layered and 50 μm thick. The periderm cells are thin walled and suberised.

The cortical zone is wide and parenchymatous. It is 60 μm thick. The cortical cells are thin walled and compact. A long the inner boundary of the cortex is a thin cylinder of Sclerenchyma cells (Figure 10.2).

The vascular cylinder consists of a wide secondary phloem which encloses central solid and dense secondary xylem (Figure 10.2). The xylem cylinder is uneven in outline. It includes diffusely scattered solitary vessels and thick walled lignified fibres. The vessels narrow in the centre and become wider towards the periphery. The outer vessels are 80 μm wide; the inner vessels are 20 μm wide.

Powder Microscopy

The root sample was powdered or macerated to study the ingredient elements found in the powder. The following elements were observed

Libriiform fibres (Figure 11.1)

Long, narrow fibres with tapering ends are frequently seen in the powder. They have thick lignified walls and narrow lumen. They are 200-300 μm long and 15 μm thick.

Fibre-Sclereids (Figure 11.2, 11.3)

The fibres sclereids are abundant in the powder. They are long, narrow and fibre like in appearance. But unlike the fibres, the fibre-sclereids have dense simple pits which are wide and canal-like. The cell lumen is also wide and the cells appears with thick walls and the pits are canal like and wide (Figure 11.2). The fibre sclereids are 300 μm long and 20 μm thick.

Vessel elements (Figure 12.1)

Narrow, cylindrical as well as wide barrel shaped vessel elements are common in the powder. They have narrowed and circular bordered pits are seen in several vessels (Figure 12.1). The end wall perforation is simple, wide and slightly oblique and is upto 200 μm long.

Tracheids (Figure 12.2)

Cells which are long and wide with multiseriate circular and wide bordered pits and similar to the pits on the vessels. But the tracheids donot possess end wall perforations. The tracheids are less frequent in the powder and are 250 μm long and 20 μm wide.

Parenchyma cells (Figure 11.1)

Narrow rectangular or wide squarish cells with thin walls and wide lumen are common in the powder. These are parenchyma cells. Some of the cells have dense, circular simple pits. Cell inclusions are seen in the cells.

Table 1: Powder characteristics of the drug

Name of the plant	Colour	Appearance	Odour	Taste
<i>Asclepias curassavica</i>	Green	Fine powder	No Characteristic	Light Bitter
<i>Pentatropis capensis</i>	Light Brown	Fine powder	No Characteristic	Bitter

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Table 2: Powder analysis of the drug

Treatment	<i>Asclepias curassavica</i>	<i>Pentatropis capensis</i>
Powder treated with water	Non sticky	Non sticky
Powder shaken with water	Foam like froth	Foam like froth
Powder treated with 5% aqueous NaOH	Greenish froth	Brownish Froth
Powder treated with 60% aqueous sulphuric acid	Blacky	Greeny
Powder pressed between filter paper for 24 hours	No oil stains	No oil stains

Table3: Ash values of the drug

Name of the plant	Total ash (% w/w)	Water soluble ash (% w/w)	Acid soluble ash (% w/w)	Alkalinity of water soluble ash (ml)
<i>Asclepias curassavica</i>	9.02	35.65	59.01	0.5
<i>Pentatropis capensis</i>	5.33	19.34	53.04	0.3

Table 4: Extractive values of the drug

Name of the plant	Ethanol soluble extract (% w/w)	Water soluble extract (%w/w)	Hexane soluble extract (% w/w)	Chloroform extract (% w/w)
<i>Asclepias curassavica</i>	66.89	25.11	7.50	63.01
<i>Pentatropis capensis</i>	56.33	16.89	4.95	50.34

Table5: Solubility values of the drug

Name of the plant	Ethanol (% w/w)	Water (aqueous)(% w/w)	Methanol (% w/w)
<i>Asclepias curassavica</i>	56.44	28.33	68.34
<i>Pentatropis capensis</i>	60.34	25.65	70.33

DISCUSSION

Many a times botanicals produced in, or obtained from, some specific region are perceived to be of better quality and, therefore, command better price. Another factor influencing the price seems to be the presence of physical impurities or adulterants in the traded botanicals. There is a definite season for harvesting of appropriately mature plant based raw material from the wild as well as from cultivated sources. Production of raw drugs, whether obtained from wild or from cultivation, was reported to fluctuate from year to year directly impacting their prices. One of the factors for this fluctuation was cited to be climate, especially precipitation, as bulk of the medicinal plants is obtained from rainfed areas. Another factor for this fluctuation in production seems to be the regulations in place for harvesting from the forests under which different forest areas are opened for harvesting once in a harvesting cycle of 3-4 years. Moreover, harvesting of some botanicals is sometimes temporarily suspended by the state forest departments to recoup wild populations of such botanicals. Microscopical examination and characterization of phytodrug were very essential in the pharmacognostic studies. Botanical identity of the phytodrug is important prerequisite for understanding the analysis of medicinal properties of any plant. If the plant identity of the drug is incorrect, the entire work on the plant becomes invalid. Hence the more

Research Article

care is taken that botanical identity of a crude drug is the threshold in the process of biological investigation.

Table6: Fluorescence analysis of the drug powder

Experiments	Visible/Day light			UV light		
	254 nm	365 nm	365 nm	254 nm	365 nm	365 nm
Drug powder	<i>Asclepias curassavica</i>			<i>Pentatropis capensis</i>		
DP+1 N NaOH(aq.)	Green	Brown	Black	Brown	Black	Black
DP + 1 NNaOH(alc.)	Dark green	Black	Black	Green	Black	Black
DP + 1 N HCl	Green	Black	Black	Dark green/Brown	Black	Black
DP + 50% H ₂ SO ₄	Green	Black	Black	Pale Brown/green	Green	Green
DP + 50% HNO ₃	Brown	Black	Black	Brown/green	Dark green	Dark green
DP + Picric acid	Brown	Black	Black	Green/brown	Dark brown	Dark brown
DP + Ferric chloride	Green	Green	Green	Brown/green	Brownish black	Brownish black
DP + HNO ₃ + NH ₃	Green	Green	Black	Brown/green	Green	Dark green

The research should be equipped with all possible diagnostic parameters of the plant on which the researcher plan to work.

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