



MICROMORPHOLOGICAL AND MICROMETRIC EVALUATION OF SECURINEGA LEUCOPYRUS (WILLD)MUELL. LEAF AND STEM- UNEXPLORED DRUG

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ABSTRACT

Securinegaleucopyrus (Willd.)Muell, belonging to Euphorbiaceae family. Commonly known in Sri Lanka by “Katupila” and in Gujarat by “Humri” is a plant used by the indigenous people for the treatment of wounds and cancer in Sri Lanka. Extracts of leaves had exhibited in vitro broad spectrum antimicrobial activities. Till date there is no pharmacognostical scientific work has been done on its leaf and stem. For the first time, T.S of stem, petiole, T.S of leaf through midrib, Surface study and micrometry was carried out. Results show Rosette crystals and starch grains in petiole, 2-3 layer of palisade parenchyma, spiral vessels in leaf, stomatal index i.e. 26 in lower epidermis. Powder microscopy of shade dried powder shows prismatic crystals and oil globules.

Key words: Securinegaleucopyrus, leaf, stomata, micrometry, stem.

INTRODUCTION

Securinegaleucopyrus (Willd.)Muell has long been used by the tribes of Sri Lanka and in India. Common in scrub jungles, limited to India, Srilanka and Burma. Armed shrubs, 1-2 mts tall, branchlets ending in thorns. Leaves elliptic, obtuse or emarginate, Flowers 1-2mm across greenish yellow in axillary fascicles. Fruits 3-6mm across, globose, white, 3 valved, seeds trigonous, brown ^[1].

This is a common weed found all over Sri Lanka although it's a desert climatic plant. Application of *Securinegaleucopyrus* (Katupila /Humari) is a commonly used plant in the management of acute and chronic types of wounds in Sri Lankan folklore medicine. Extracts of leaves had exhibited in vitro broad spectrum antimicrobial activities^[2]. Still it has been practiced and the fresh leaf always used in the form of paste for the treatments as a single or with combination with other herbs. The leaves are also used internally for the treatment of cancer and its long been believed to cure cancer with reported cases still existing. In spite of its high medicinal value, *Securinegaleucopyrus* (Willd.)Muell pharmacognostical characters of its leaf and stem, is not reported yet. Hence, in this research paper an attempt has been made to establish the genuinity of the plant through pharmacognostical characters of leaf and stem which includes macroscopic, microscopic, micrometric characters including histochemical analysis.

MATERIAL AND METHODS

Fresh leaves and stem were collected from the natural habitat of Raka Katia forest area Jamnagar. Pharmacognostical evaluation of fresh drug was carried out by taking free hand sections ^[3]. Both upper and lower epidermis was used to surface study through hand peeling method. Micrometric readings of both surface i.e. stomatal length, stomatal index etc. were scientifically studied and mean value taken in to consideration^[4]. Histochemical test were done by using both thick sections to observe the presence of tannins, lignin, starch etc^[5]. Powder microscopy of shade dried both powders were carried out. Photomicrographs were taken using Carl zeisstrinocular microscope attached with camera.

RESULTS AND DISCUSSION

Plant collected and identified through various flora and with help of pharmacognosist^[6]. The leaves were separated from the stem, leaves and stem washed with running fresh water and few pieces stored in solution of AAF (Alcohol: Acetic acid: Formalin) in the ratio of (90:5:5) ^[7] to utilize them for microscopic studies. Remaining fresh leaves and stem pieces were shade dried, pulverized and sieved through 80 mesh and preserved in an airtight glass container for future studies Phm No. 6068/13. *S. leucopyrus* is large straggling shrub, branchlets arrested, stiff thorn like. Leaves simple, alternate, moderate sized, distichious,

thin stipulate, stipules lanceolate. Leaves 3-5 cm long and 1-1.5 cm broad, coriaceous oblong to elliptic in shape. Apex obtuse, venation reticulate 6-8 veins and many veinlets. Upper dark green lower parrot green in colour Inflorescence axillary fascicles. Flowers unisexual, female fewer than male. Fruit globose white^[8]. (Fig 1)



Fig.1. Natural Habit of Plant

Microscopic:

Transverse section of petiole:

The transverse section of the petiole shows outer single layered compactly arranged barrel shaped epidermal cells covered with cuticle followed cortex. Cortex widely distributed, made up of parenchyma cells, large number of rosette and prismatic crystals of calcium oxalate, oil globules are also present. A ring of pericyclic fibres covers the vascular bundles. Vascular bundles consists of one large vascular in the centre. Each bundle is conjoint, collateral, surrounded by a parenchymatous bundle sheath. Vascular bundles radially arranged metaxylem towards periphery, protoxylem towards centre. Xylem consist xylem parenchyma and its fibres. Phloem present below the xylem with some sieve elements. (Fig 2).



Fig. 2. T.S. of Petiole

Transverse section of leaf through mid rib:

Detailed T.S. shows upper and lower epidermis covered with thick cuticle. Lower epidermis made up of somewhat papillae like parenchyma cells. Lamina shows 2-3 layers of palisade underneath the upper epidermis and 3-4 of rows of spongy parenchyma traversed with obliquely cut vascular bundles and rosette crystals of calcium oxalate. Section passing through the midrib shows collenchymatous tissue is located underneath both the epidermis. Vascular bundles consists of one large vascular in the centre. Each bundle is conjoint, collateral, surrounded by a parenchymatous bundle sheath. Vascular bundles radially arranged metaxylem towards periphery, protoxylem towards centre. Xylem consist xylem parenchyma and its fibres. Phloem present below the xylem with some sieve elements. (Fig 3).

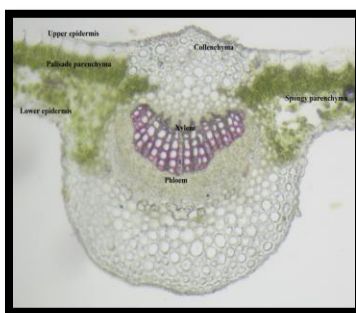


Fig. 3.T.S. Through Midrib

Surface preparation:

Thin upper and lower surface prepared by simple peeling method the lower epidermis shows the numerous stomata, where as upper epidermis devote of stomata, stomata mainly of anamocytic, epidermal cells, prismatic and rosette crystals of calcium oxalate and large quantities of oil globules distributed all over the surface. (Fig 4 A, B, C).

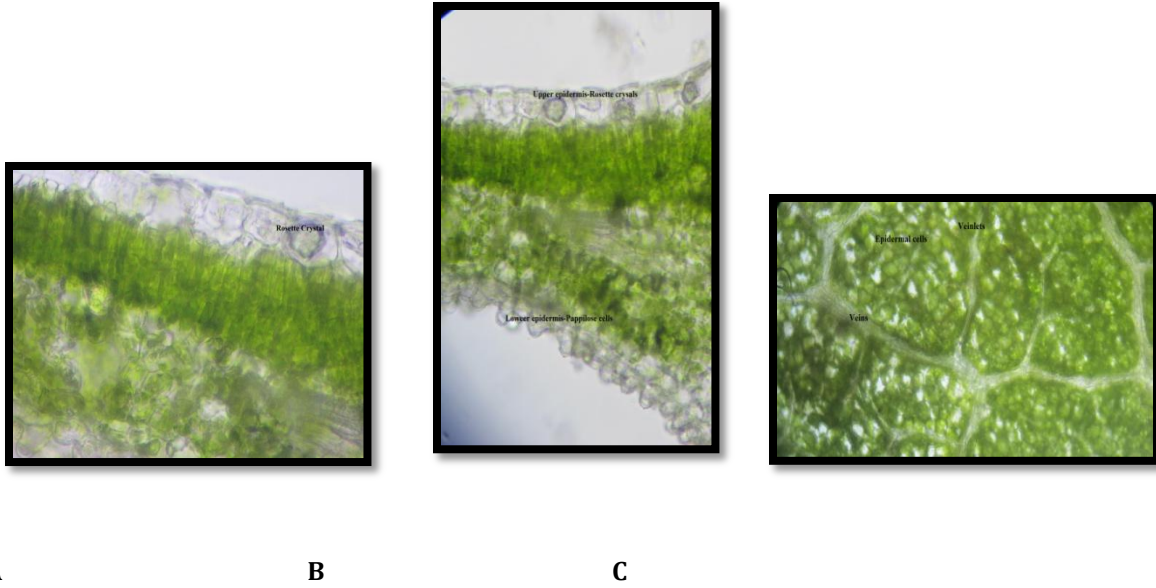


Fig.4. A. Rosette crystal upper epidermis. B. Lower epidermis(Papillate cells), C. Epidermal cells, Veins and veinlets.

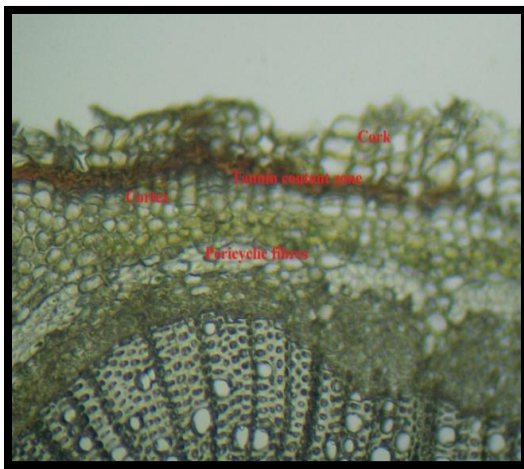


Fig. 5. T.S. of Stem Unstained

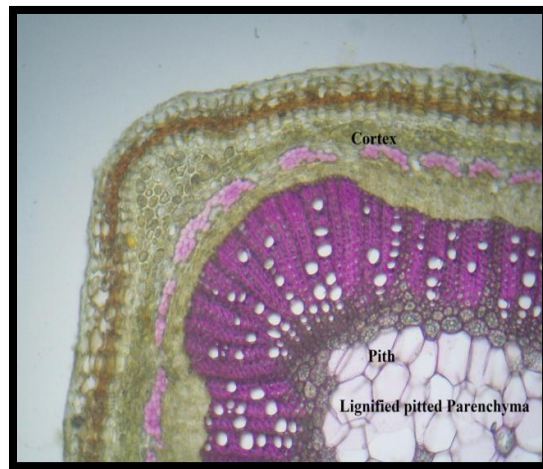


Fig. 6. T.S. of Stem Stained

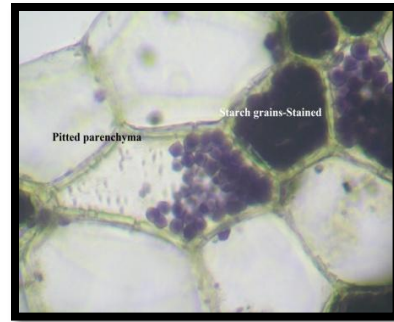
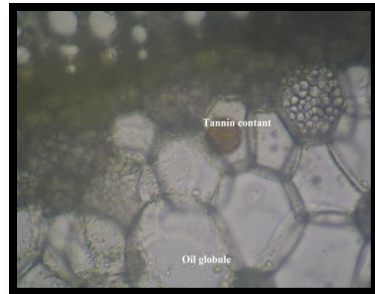
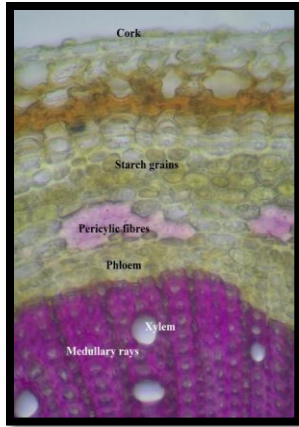


Fig. 7. T.S. of Stem Stained Fig. 8. T.S. of Stem Stained Fig. 9. T.S. of Stem Stained

Powder microphotographs:

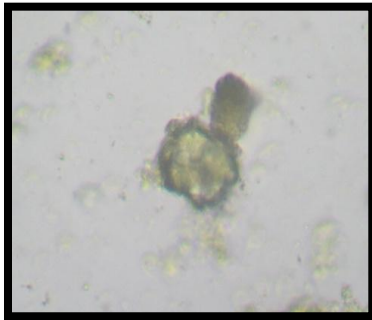


Fig. 10. Rosette crystal



Fig. 11. Schleroid

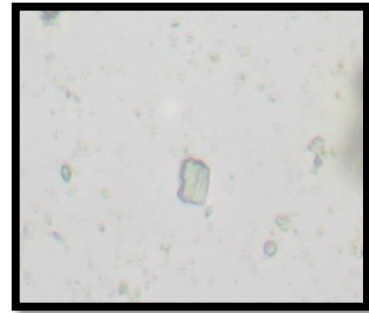


Fig. 12. Prismatic crystal



Fig. 13. Fibre

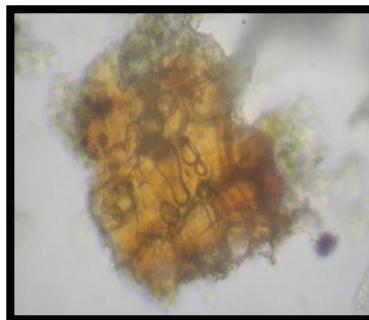


Fig. 14. Tannin content

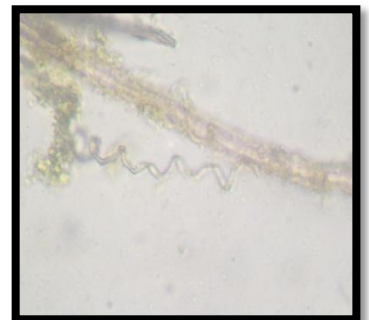


Fig. 15. Spiral vessel



Fig. 16. Oil globule- leaf

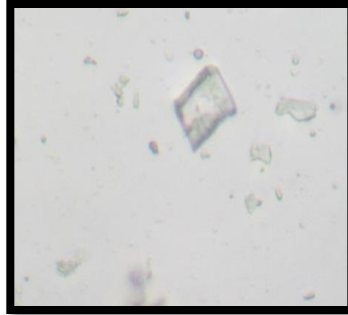


Fig. 17. Prism of stem



Fig. 18. Lignified fibres of stem

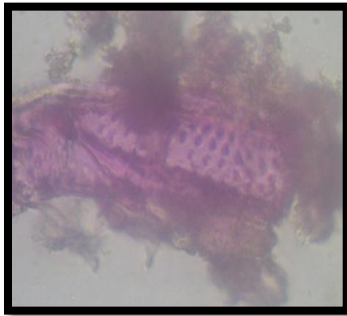


Fig. 19. Pitted vessel of stem

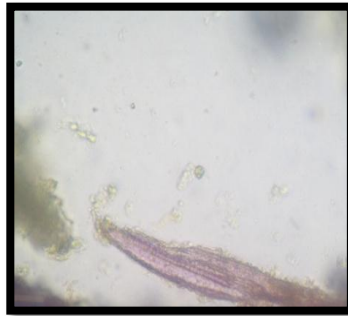


Fig.20.Fibre with lumen- stem

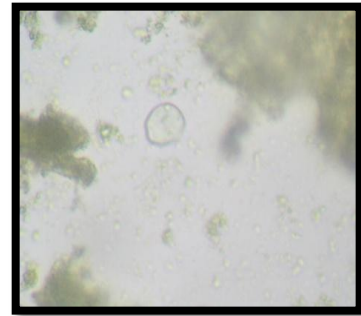


Fig. 21. Oil globule- stem

Surface study Microphotographs of Leaf:

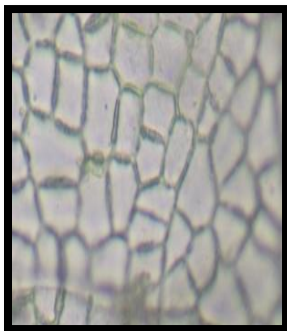


Fig.22. Upper epidermis

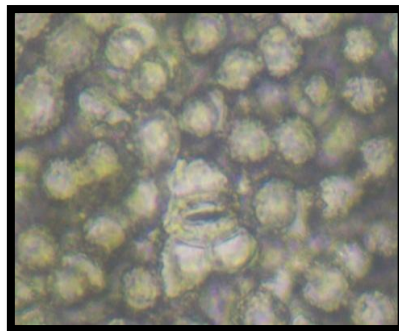


Fig.23.Lower epidermis with stomata

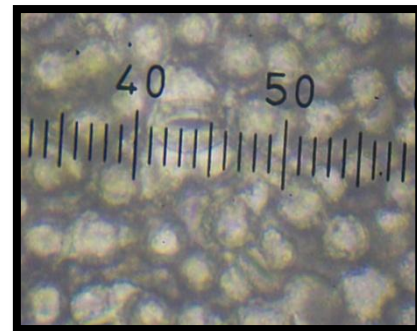


Fig. 24. Micrometric studies

Powder microscopy:

Organoleptic characters shows leaf powder is dark green in colour, astringent in taste, aromatic in odour and coarse in touch. Diagnostic powder microscopic characters were oil globules, prismatic and rosette crystals of calcium oxalate, anamocytic stomata. Epidermal cells, fragments spiral vessels, lignified fibres. (Fig 10-16).

Micrometric analysis:

Stomatal length, breadth, stomatal index and other constituents were scientifically measured (3 successive readings) the mean value taken into consideration and results depleted in table no. 1. (Fig 22-24).

Sr. No.	Characters	Lower epidermis	Upper epidermis
1	Stomata length	11.50 μm	-----
2	Stomata breadth	8.5 μm	-----
3	Stomata radius	75 μm^2	-----
4	Stomatal Index	26	-----
5	Pallisade ratio	—	2-3
7	Vein islets no.	16-18	---
7	Epidermal cells	195 μm^2	205 μm^2
8	Prismatic crystal	35 μm^2	32 μm^2
9	Rosette crystal	42 μm^2	42 μm^2

Table 1: Micrometric analysis of leaf

Transverse section of Stem:

The diagrammatic section is somewhat quadrangular in shape, shows outer epidermis followed by cortex along with the pericyclic fibre zone, radially arranged vascular bundles and centrally located large parenchymatous pith. (Fig 5-9).

Outer layer composed of barrel shaped 3-5 rows of cork cells, inner cells filled with tannin content. Cortex made up of compactly arranged parenchymatous cells, some of the parenchymatous cells are filled with prismatic crystal of calcium oxalate, simple starch grains with hilem and oil globules. Endodermis single layered somewhat elongated with thin walled parenchymatous cells forming ring like structure. Pericyclic fibers situated above the xylem forming an arc like structure, and are lignified. Medullary rays uniserrate, separates the vascular tissues, somewhat longitudinally arranged barrel shaped cells filled with some oil globules and starch grain. In vascular bundles metaxylem towards periphery and protoxylem towards pith, xylem consists of xylem parenchyma and fibres, phloem situated above xylem forming cap like structure with

few elements and fibre. Pith covers nearly half portion of the section, made up of thick walled, compactly arranged parenchyma cells with some prismatic crystals and oil globules. Tail region of the vascular bundle consist of thick walled lignified pitted parenchyma cells, loaded with starch grains. (Fig 5-9).

Powder Microscopy of Stem:

Stem powder light yellow in colour, astringent in taste with characteristic odour. The diagnostic characters of the stem powder shows Tannin content, cork in surface view, Prismatic crystal of calcium oxalate, lignified fibres, pitted vessel, fibre with lumen and oil globule (Fig. 17-21).

Histochemical tests:

Various Histochemical tests were conducted on sections of the leaf and stem. The results are depleted in table no. 2.

Sr. No.	Reagents	Observation	Characteristics	Leaf	Stem
1.	Phloroglucinol+ Conc. HCl	Red	Lignified cells	++	++
2.	Iodine	Blue (Petiole)	Starch	--	++
3.	Phloroglucinol+ Conc. HCl	Dissolved	Calcium oxalate crystals	++	++
4.	FeCl ₃ solution	Dark blue to black	Tannin cells	++	++
5.	Sudan III	Red	Oil globules	++	++

Table 2: Histochemical tests for leaf and Stem

CONCLUSION

Pharmacognostical work reveals that petiole shows large number of prismatic crystals. Leaf T.S

showed 2-3 layers of palisade parenchyma cells, prismatic and rosette crystals. Surface study shows anamocytic stomata. The values extracted from micrometry were scientific and constant. Stem section and its powder shows the presence of calcium oxalate crystals, large amount of tannin and oil helpful in the treatment of cuts and wounds may be considered for further research works.

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