REVIEW ARTICLE

PHARMACOGNOSTICAL ANALYSIS OF *CLITORIA TERNATEA* LEAVES:

A COMPREHENSIVE REVIEW

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Abstract:

Clitoria ternatea L., commonly known as butterfly pea or aparajita, is a member of the Fabaceae family and holds significant importance in Ayurvedic and Southeast Asian traditional medicine. The present study focuses on the pharmacognostical evaluation of its leaves to establish diagnostic features for quality control and standardization. Macroscopic examination revealed compound leaves with ovate to orbicular leaflets and narrowly triangular stipules. Microscopic analysis of transverse sections stained with Safranin and Astra blue displayed key anatomical features such as paracytic stomata, trichomes, vascular bundles, and calcium oxalate crystals. Quantitative microscopy provided essential parameters like stomatal number, stomatal index, vein islet and termination numbers, and palisade ratio. Powder microscopy further confirmed the presence of diagnostic elements including starch grains and trichomes. These pharmacognostical findings provide a reliable basis for authentication, identification, and quality assessment of *C. ternatea* leaves in herbal formulations, ensuring safety and efficacy in traditional and modern medicinal applications **Keyword:** Clitoria ternatea, Butterfly Pea, Leaf Microscopy, Powder Microscopy, Macroscopic Evaluation, Quantitative Microscopy, Anatomical Study.

Introduction:

The plant species *Clitoria ternatea* L, which is indigenous to the Indonesian island of Ternate, is a member of the Fabaceae family and is sometimes referred to as Asian pigeonwings, bluebellvine, blue pea, butterfly pea, ordofan pea, or Darwin pea. It is usually referred to as aparajita in Indian Ayurveda. In India, it is occasionally mistaken for Convolvulus prostratus (Convolvulus pluricaulis). Different plant parts have long been used in Ayurvedic and indigenous Southeast Asian medicine to treat neurological disorders, anxiety, depression, inflammation, fever, snake and scorpion bites, digestive and respiratory conditions, diarrhea, wounds, and constipation. They are also used to support

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memory and cognition (as a medhya rasayana). Numerous pharmacological actions, including antioxidant, anti-inflammatory, analgesic, antidiabetic, antibacterial, neuroprotective, hepatoprotective, antipyretic, insecticidal, and even cytotoxic effects against specific cancer cell lines, have been proven by recent studies. [1][2][3]

Taxonomical Classification [4]

Kingdom: Plantae

Phylum: Magnoliophyta Class: Magnoliopsida Subclass: Rosidae

Order: Fables
Family: Fabaceae
Genus: Clitoria
Species: ternatea
Distribution [5, 6]

A plant species that grows in the pantropics. Though widely naturalized and grown throughout the world's tropics and subtropics, it is thought to have originated in tropical Asia, specifically in India and Southeast Asia. Because of its resilience to climate change, quick growth, capacity to fix nitrogen, and use as a fodder, decorative, and medicinal plant, it has spread well. The plant is suitable for both natural and managed ecosystems since it grows well in tropical and subtropical regions with moderate rainfall and can withstand poor, sandy, or degraded soils.

Vernacular Names [7]

Tamil: Sanghu pushpam

English: Butterfly pea, Asian pigeon-wings, blue pea, cordofan pea, Darwin pea

Malayalam: Sangu pushpam

Telugu: Sankha pushpam

Sanskrit: Aparajita, Vishnukranta, Yonipushpa

Hindi: Aparajita

Kannada: Sattaga; Shankha pushpa

Botanical Description [8]

Leaves

- Type: Imparipinnately compound, usually having a terminal leaflet in addition to two to three (sometimes even four) pairs of leaflets.
- **Leaflets:** Elliptic to ovate orbicular, typically 1.5–5 cm long and 0.3–3 cm wide, with a cuneate or rounded base and a sharp, rounded, or notched (emarginate) apex; surfaces are occasionally glabrous or sparsely appressed pubescent.
- **Stipules & Petioles:** Stipules are narrowly triangular (2–5 mm long); stipels filiform up to 2 mm; petiole length is around 1.5–3 cm; and petiolules are 1-2 mm.

Pharmacognostical Studies

Methodology

- 1. **Macroscopy** [9]: External feature of the test sample was documented using Nikon D-5600 Digital camera.
- 2. **Microscopy** ^[10]: The Sample was preserved in fixative FAA for more than 48 h. The preserved specimens were cut into thin transverse sections using a sharp blade and the sections were stained with 0.8% Safranin and 0.5% Astra blue. Transverse sections were photographed using an Axiolab5 trinocular microscope attached with a Zeiss Axiocam208 colour digital camera under bright field light. Magnifications were indicated by a scale bar.
- 3. Quantitative Microscopy [10]: Leaf fragments of about 5 x 5 mm in size were placed in a test-tube containing about 5 ml of saturated aqueous solution of chloral hydrate and heated in a water bath for 10-15 minutes. Then it was kept on a slide and mounted it in glycerine. Examine under a microscope with a 4x objective and a 10x eyepiece, equipped with a camera lucida. Leaf parameters like Stomatal number, Stomatal index, Vein islet number, Vein termination number, and Palisade ratio are determined.
- 4. Powder Microscopy [11]: A pinch of the powdered sample was mounted on a microscopic slide with a drop of 50% glycerol after clearing with a saturated solution of chloral hydrate. The sample was treated with iodine solution to confirm the presence of starch grains. Characters were observed using a Nikon ECLIPSE E200 trinocular microscope attached with a Zeiss ERc5s digital camera under bright field light. Photomicrographs of diagnostic characters were captured and documented.

Results:

1. Macroscopy

Fresh leaves are green, pinnately compound with 5 to 7 leaflets, imparipinnate, leaflets are smooth, thin, delicate, elliptic to ovate shaped, margin entire, tip emarginate, base is broadly cuneate, measuring 2.5 to 4 cm long and 2 to 3 cm wide; petiole up to 3 cm long; with characteristic odor and bitter taste (Figure. 1).

2. Microscopy

Petiole

TS of petiole is cordate in outline; outer layer is single layered epidermis covered by cuticle and bears few simple covering trichomes; cortex is formed of 2 to 3 layers of collenchyma cells followed by 4 to 5 layers of chlorenchyma cells; 8 to 9 conjoint, collateral and closed vascular bundles are arranged on ring in the inner cortex; vascular bundles are made up of normal vascular elements; each bundle is capped by 3 to 4 celled thick pericyclic fibers; phloem is found towards outside and xylem elements towards inner side; parenchymatous pith is present at the central portion; few prismatic crystals are distributed in the pericyclic region; two trace bundles can be seen, each one below the upper wings (Figure. 2).

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(Abbr: Chl - chlorenchyma; Col - collenchyma; Ct - cortex; E - epidermis; Pa - parenchyma; PCr - prismatic crystal; Per - pericycle; Ph - phloem; Pi - pith; TB - trace bundle; V - vessel)
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Figure 1: Macroscopy of Clitoria ternatea leaf

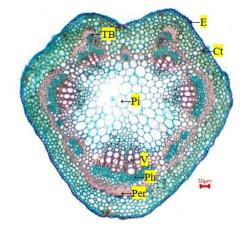


Figure 2: T.S. of Clitoria ternatea petiole

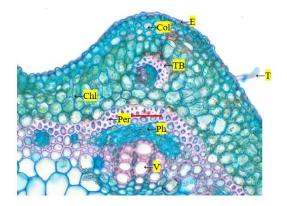


Figure 3: Enlarged upper region

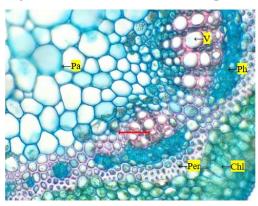


Figure 4: Lateral portion enlarged

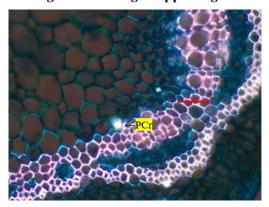


Figure 5: Under polarized field

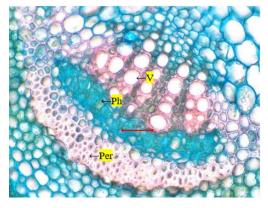


Figure 6: Vascular bundle enlarged

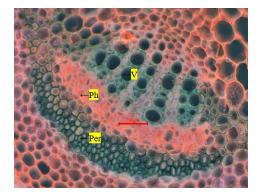


Figure 7: Vascular bundle enlarged

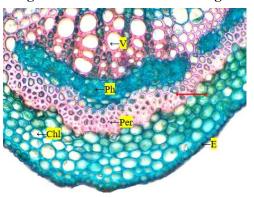


Figure 8: Enlarged lower portion

Leaf

TS of leaf shows slightly elevated upper and wavy lower midrib surface with lateral laminar extensions (Figure 9).

Midrib: TS of midrib shows single layered upper and lower epidermii covered by cuticle and bears few warty trichomes; 3 to 4 layers of collenchymatous hypodermal layer is present below the epidermii followed by parenchymatous ground tissue embedded with an arc shaped vascular bundle; 3 to 4 layer thick pericyclic fibers covers the lower portion of bundle followed by phloem tissue which surrounds the xylem elements; two small bundles can be seen just above this large bundle; few prismatic crystals are distributed in the pericyclic region (Figure 9).

Lamina: TS of lamina shows single layered upper and lower epidermii covered by cuticle and bears simple warty trichomes; mesophyll tissue is differentiated into upper compactly arranged columnar cells of palisade layer followed by 3 to 4 layers of spongy parenchymatous cells; veins can be seen traversing through the mesophyll tissue (Figure 9).

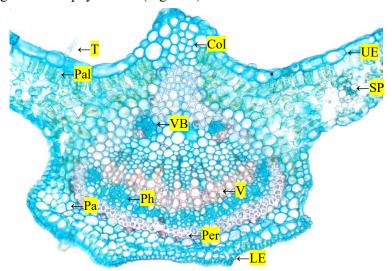


Figure 9. TS of Clitoria ternatea leaf passing through midrib

TS of leaf

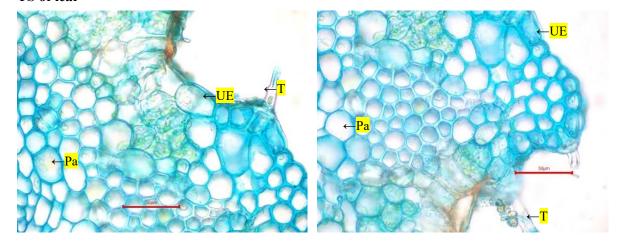


Figure 10, 11: Enlarged view of upper portion

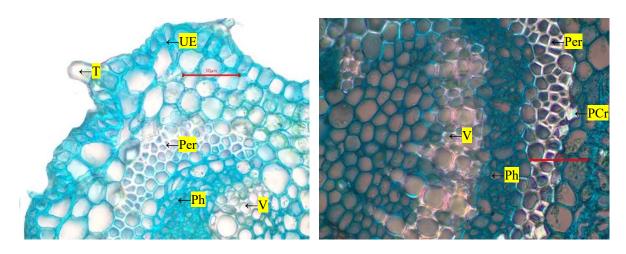


Figure 12: Lateral portion enlarged

Figure 13: Under polarized field

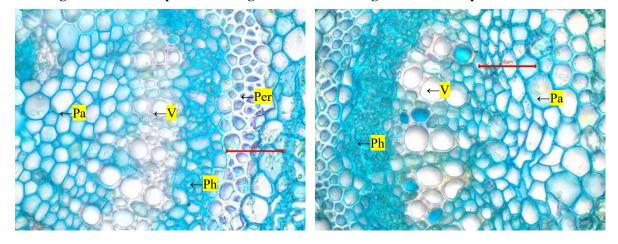


Figure 14: Vascular bundle enlarged

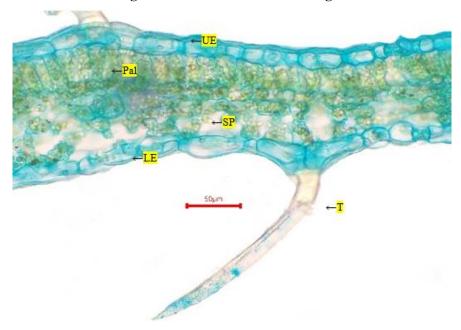


Figure 15: TS of lamina

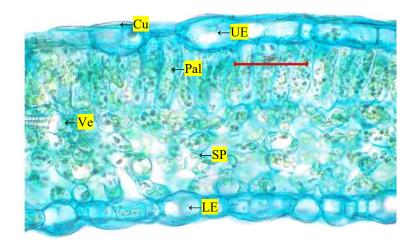


Figure 16: TS of lamina enlarged view

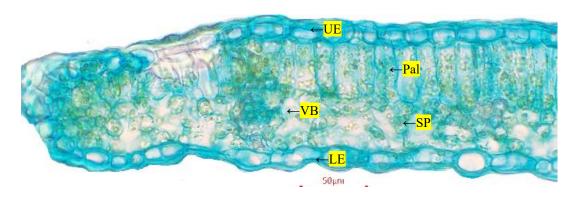


Figure 17: TS of leaf margin

Col - collenchyma; Cu - cuticle; LE - lower epidermis; Pa - parenchyma; Pal - palisade cells; PCr - prismatic crystal; Per - pericycle; Ph - phloem; SP - spongy parenchyma; T - trichome; UE - upper epidermis; V - vessel; VB - vascular bundle; Ve - vein

3. Quantitative microscopy

The quantitative parameters obtained during microscopic observation of epidermal peelings of leaf were recorded in Table 1. The leaf is amphistomatic with paracytic stomata on epidermis.

Table 1: Quantitative microscopy of of Clitoria ternatea leaf

Parameters	Upper epidermis (/mm²)	Lower epidermis (/mm²)
Epidermal number	700 - 800	900 - 1000
Stomatal number	200 - 220	300 - 320
Stomatal index	21.5 - 22.2	24.2 - 25
Palisade ratio	4 - 5	
Vein islets number	15 - 20	
Vein termination number	20 - 25	

Quantitative microscopy of of Clitoria ternatea leaf

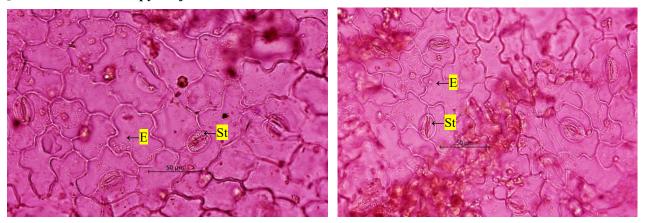


Figure 18. Upper epidermis

Figure 19. Lower epidermis

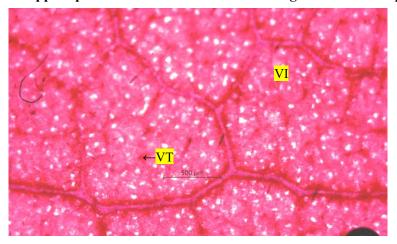


Figure 20. Vein Islets and Terminations

E - Epidermis; St - Stomata; VI - Vein Islet; VT - Vein Termination

4. Powder Microscopy

The powder is light green coloured with characteristic odour and bitter taste; shows the characters like fragments of simple trichomes from petiole, warty trichome from leaf, fragments of petiole epidermis, parenchyma cells, surface view of epidermis with paracytic stomata, palisade cells, crystal fibre, vessels with spiral and simple pitted thickenings, and prismatic crystals.

Powder microscopy of of Clitoria ternatea leaf

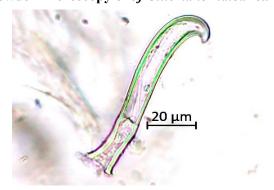


Figure 21: Covering trichome from petiole

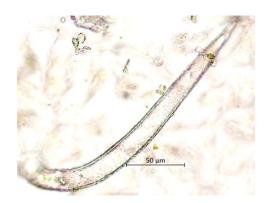


Figure 22: Warty trichome from leaf

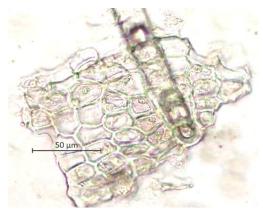


Figure 23: Petiole epidermis

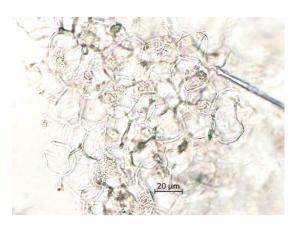
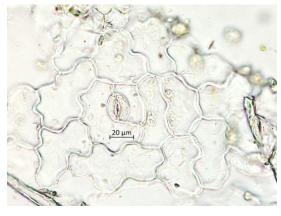


Figure 24: Parenchyma cells



20 µm

Figure 25, 26: Epidermis with paracytic stomata

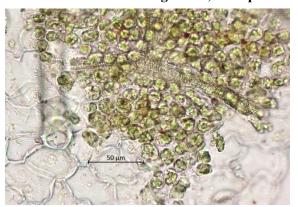


Figure 27: Palisade cells

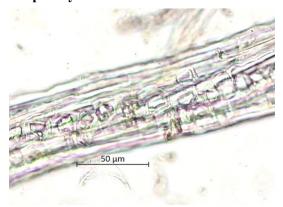


Figure 28: Crystal fiber

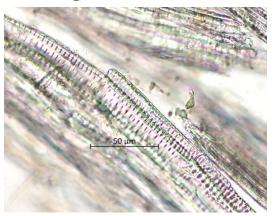


Figure 29: Pitted vessels

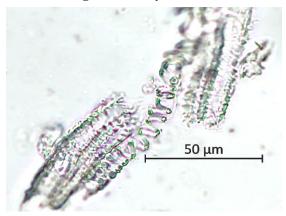
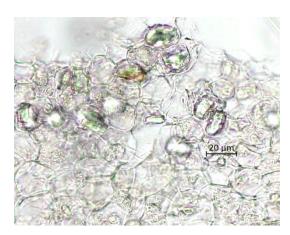


Figure 30: Spiral vessels



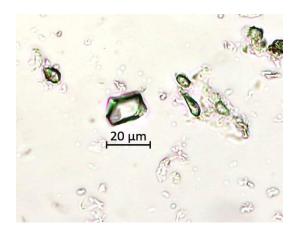


Figure 31, 32: Prismatic crystals

Discussion:

Macroscopy

The fresh leaves of *Clitoria ternatea* exhibit typical pinnately compound structure with 5 to 7 imparipinnate leaflets, characterized by smooth, thin, and delicate elliptic to ovate shapes. The entire margin, emarginate tip, and broadly cuneate base are consistent with botanical descriptions. Leaflet size and petiole length fall within expected ranges, confirming specimen maturity. The distinctive bitter taste and characteristic odor further aid in authenticating the plant material. These macroscopical features are vital for correct identification and quality assessment in pharmacognostic studies.

Microscopy

The transverse sections of petiole, midrib, and lamina of *Clitoria ternatea* reveal typical anatomical features such as a single-layered epidermis with trichomes and a well-developed cuticle. The petiole shows a cordate outline with collateral vascular bundles and pericyclic fibers, while the midrib exhibits a prominent arc-shaped vascular bundle with associated fibers. The lamina displays differentiated mesophyll with distinct palisade and spongy parenchyma layers. Presence of prismatic crystals in pericyclic regions supports species-specific identification. These microscopic traits are essential markers in pharmacognostical authentication of *Clitoria ternatea*.

Powder Microscopy

The light green powder of *Clitoria ternatea* exhibits characteristic bitter taste and odor, confirming its organoleptic identity. Microscopic examination reveals key diagnostic features such as simple and warty trichomes, fragments of petiole epidermis, parenchyma cells, and epidermis with paracytic stomata. The presence of palisade cells, crystal fibers, vessels with spiral and simple pitted thickenings, along with prismatic crystals, further supports the authenticity of the plant material. These powder characters are valuable for distinguishing *Clitoria ternatea* from adulterants in powdered form. Overall, powder microscopy is a vital tool in quality control and pharmacognostic evaluation.

Conclusion:

The pharmacognostical evaluation of the leaves of *Clitoria ternatea* has provided essential diagnostic features necessary for accurate identification and quality control of this medicinal plant. The macroscopic study revealed typical characteristics such as ovate to elliptical leaflets with smooth

margins and a green, slightly pubescent surface. Microscopic examination showed the presence of key anatomical features including paracytic stomata, unicellular covering trichomes, collateral vascular bundles, and calcium oxalate crystals—features that are valuable for distinguishing the species from possible adulterants. Overall, this study contributes to the standardization and pharmacognostical profiling of *Clitoria ternatea* leaves, providing a scientific basis for their use in herbal preparations and laying the groundwork for further phytochemical and pharmacological research.

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Declaration of Competing Interest: The authors declare that they have no competing interests.

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