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## Studies on histology, thin-layer chromatographic profile and physicochemical evaluation of stem bark of *Butea monosperma* (Lam.) Kuntze

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**Abstract**

*Butea monosperma* (Lam.) Kuntze (Family-Fabaceae), commonly known as 'Palāsa' (English - Flame of the forest), is widely used in Indian traditional systems of medicines and all the parts *vis.* Bark, leaves, flowers seeds and gum possess diversified medicinal properties for the treatment of various ailments. In the present study, histology, thin-layer chromatographic profile and physicochemical evaluation of stem bark of *Butea monosperma* has been carried out to establish the authenticity of the drug.

**Keywords:** *Butea monosperma*, bark, histology, thin-layer chromatography, ayurveda

**Introduction**

*Butea monosperma* (Lam.) Kuntze, is one of the most beautiful of all flowering trees, found throughout India and very conspicuous when in flowering. The bark is one of the important officinal parts of the plant and reported to be astringent, bitter, acrid, thermogenic, emollient, anthelmintic, aphrodisiac, appetizer, digestive, tonic and alterative. It is useful in vitiated conditions of *pitta* and *kapha*, anorexia, dyspepsia, hepatopathy, abdominal tumours, colic, intestinal worms, bleeding piles, ulcers, haemorrhages, amenorrhoea and dysmenorrhoea. The gummy exudation from the bark is an excellent astringent and is useful in diarrhoea, dysentery and haemorrhage. It is an important ingredient of traditional formulations like *Ayaskṛti*, *Cerīya arimēdas taila*, *Balā taila*, *Palāśa Kṣāra*, *Nyagrodhādi Kvātha Cūrṇa*, *Mahānārāyaṇa Taila* etc. [1-4]. Modern scientific studies of bark also reported anti-stress activity [5], anti-inflammatory activity [6], wound healing activity [7, 8], osteogenic and osteoprotective activity [9, 10]. The bark contains phytochemicals are kino-tannic acid, gallic acid, pyrocatechin, palasitrin, and major glycosides as butrin, butolic acid, cyanidin, allophanic acid, histidine, lupenone, lupeol, (-)- medicarpin, miroestrol, palasimide, shellolic acid, 3, 9-dimethoxypterocarpan, triterpenoid ester 3 $\alpha$ -hydroxyeuph-25-enyl heptacosanoate [11], tetratriacont-15-ene, heptacos-11-ene, 15-hydroxyethylheptadec-12-enoate, 10-hydroxy dodecyltridec-5enoate [12].

The pharmacognostic studies of aerial parts of the plant such as stem, leaves, flower and seed have been reported earlier [13-15]. The present study aims to provide a detailed macro and microscopic features, thin-layer chromatographic profile and physicochemical properties of stem bark of *Butea monosperma*.

**Materials and methods****Histological studies**

Fresh stem bark from authenticated mature tree (20 years old) was collected from Cholayil Medicinal Plant Conservation Park, near Uthukkottai, Thiruvallur district, Tamilnadu. The bark was cut in to small pieces and fixed immediately in FAA for 24h and embedded in paraffin wax after dehydration and infiltration. Sections were taken using rotary microtome to the thickness of 8-12  $\mu$ m, stained with toluidine blue and photographed [16].

**Thin-Layer Chromatographic (TLC) Analysis**

For the TLC analysis, stem bark was shade dried for a week and powdered. 2 g of powdered samples was extracted under reflux with methanol (3 times) in a water-bath, then concentrated and dried. The residue was re-dissolved in methanol and used for the TLC spotting. For stationary phase precoated Silica Gel F<sup>254</sup> (Merck) plate and for mobile phase Toluene: ethyl acetate (3:1) was used. After the plate development, dried and was dipped in 1% Vanillin H<sub>2</sub>SO<sub>4</sub> and heated at 105 °C for colour development and the spots R<sub>f</sub> value are recorded.

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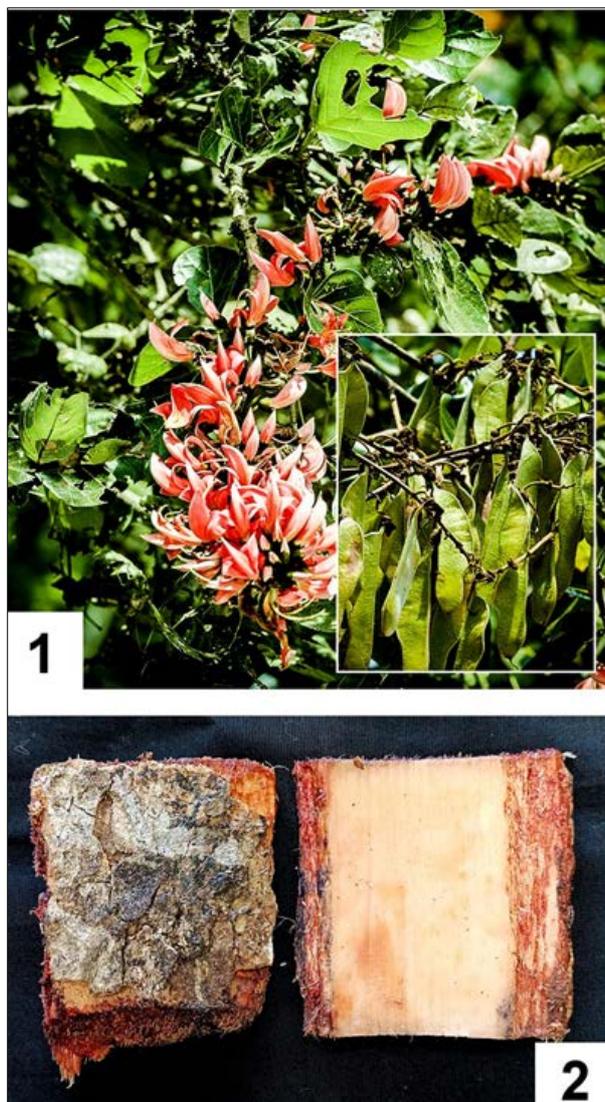
**Physicochemical evaluation**

For the analysis of water soluble extractives, Alcohol soluble extractives, Total ash and Acid insoluble ash methods adopted from the Ayurvedic Pharmacopoeia of India<sup>[17]</sup>.

**Observations**

**Macroscopic characters of stem bark** (Figure – 1: 2)

The mature stem bark is about 8 – 10 mm thickness, somewhat curved, external surface ash greyish colour, rough, cracked, irregular pieces of thin flakes due to exfoliation of rhytidome, peels of easily, exfoliated surface shows shallow. Internal surface is pale brown, rough, fibrous, cut surface orangish red colour due to exudation of gummy substance, astringent taste and no characteristic odour.



**Fig 1:** Plant habit & Macroscopic features of stem bark of *Butea monosperma*. 1. Plant in flowering & fruiting (Courtesy Mr. Pradeep Cholayil)  
2. Macroscopic features of stem bark

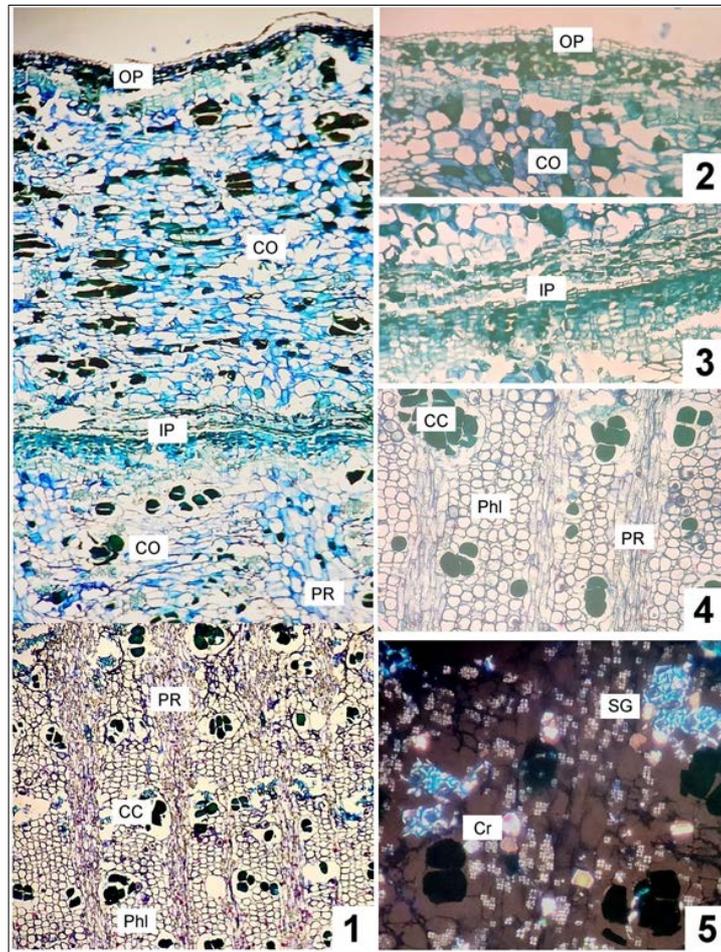
**Microscopic characters of stem-bark** (Figure-2:1-5; Figure-3:1-4)

Mature bark differentiated into periderm, secondary cortex and phloem. Periderm consist distinct alternating layers of phellem, phellogen and phelloderm both outer and inner region. Outer periderm shows 4 to 6 layers of tangentially elongated cork cells which are loosely arranged; along with outer secondary cortex it forms 2 to 3 mm thickness of rhytidome. Inner periderm distinctly marked by compressed zone consist of 6 to 8 layers rectangular cork cells and loosely arranged. Secondary cortex shows moderately thick walled parenchyma cells which are irregular in outline and compactly arranged (Figure – 2: 1&2). Dark brown colour tanniferous cells found abundantly, mostly in group, distributed both cortex and phloem regions (Figure – 1: 1&4). Small bundles

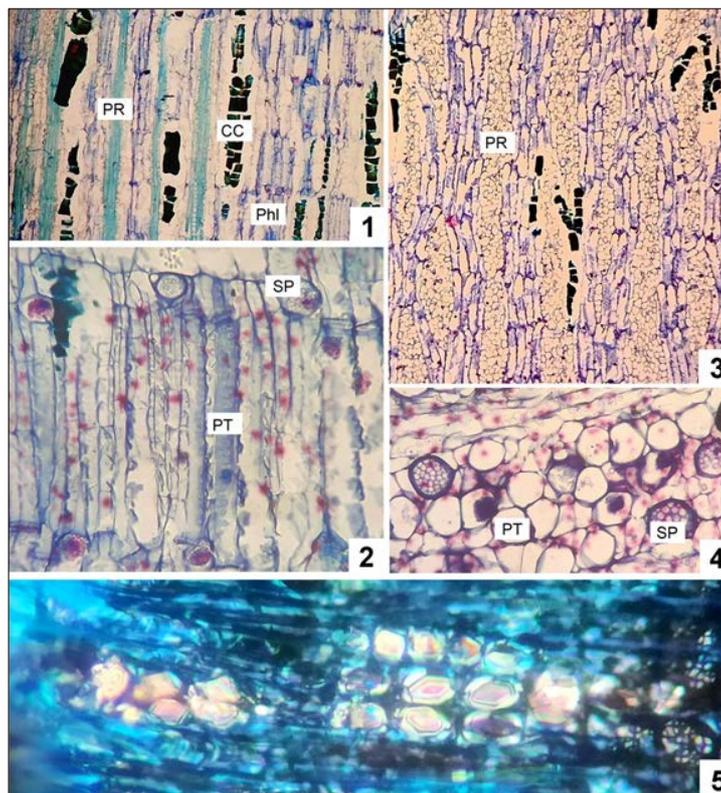
of phloem fibers found scattered. Prismatic type of calcium oxalate crystals are often distributed in both fibers and parenchyma cells. Starch grains are circular or oval in shape; simple or compound found in secondary cortex, phloem parenchyma and phloem rays (Figure – 2: 5). Phloem consist of sieve elements, phloem rays and tanniferous cells. Sieve tubes and companion cells are alternately arranged; sieve tubes mostly circular in outline; phloem rays multiseriate, homocellular, 4 to 8 cells wide. Sieve area and sieve plate distinctly visible (Figure – 3: 1-4).

**Thin Layer Chromatographic (TLC) analysis** (Table -1; Figure – 4)

The TLC profile and Rf – values of stem bark as follows



**Fig 2:** Anatomy of stem bark. 1 Cross section of bark, 2 & 3. Periderm enlarged, 4. Secondary phloem enlarged, 5. Secondary cortex enlarged (under polarized light)



**Fig 3:** Anatomy of stem bark, 1. Radial longitudinal section of bark, 2. Phloem radial longitudinal section enlarged, 3. Phloem tangential longitudinal section, 4. Phloem cross section enlarged, 5. Crystal fibres enlarged (under polarized light)



**Fig 4:** TLC fingerprint of stem bark, (CC – Cell content (Tanniniferous cell); CO – Cortex; Cr – Crystals; IP – Inner periderm; OP – Outer periderm; Phl – Phloem; PF – Phloem fibre; PR – Phloem rays; PT – Phloem tube; SG – Starch grains; SP – Sieve plate; SPhl – Secondary phloem)

**Table 1:** Rf – Values Visible light (After spray)

Rf – Values	Colour of the spot
0.08	Blue
0.16	Violet
0.25	Grey
0.31	Violet
0.35	Dark violet
0.51	Grey
0.65	Violet
0.70	Dark violet
0.81	Blue
0.86	Violet

### Physicochemical properties

**Table 2:** Physicochemical properties

Parameters	Values (%)
Water soluble extractive	14.15 ± 0.30
Alcohol soluble extractive	10.69 ± 0.65
Total ash	7.58 ± 0.03
Acid insoluble ash	1.72 ± 0.37

Values from triplicates (Mean ± SD)

### Discussion and conclusion

Tree bark is a potential crude drug and plays important role in various indigenous system of medicines. Structurally it is very complex and containing many primary and secondary metabolites. Phytochemicals present in the bark protects the plants from herbivorous animals and also useful for mankind in the preparation of many drugs for various ailments. The complex structure of the bark can be exploited for botanical identification to maintain the quality and purity of the drug [18].

The stem bark of *B. monosperma* is considered to be efficacious in the treatment of vaginal diseases, helminthic manifestation, haemorrhages etc. in Ayurveda and other

traditional system of medicines [1]. In the present study, we provides the detailed histological features and TLC fingerprint profile, physicochemical properties of the stem bark of *B. monosperma* to identify the adulterants from commercial drug.

### Diagnostic features of the bark

Easily separable rhytidome, distinct layers of inner outer periderm, orangish red colour gummy exudate, dark brown coloured group of tanniniferous cell, crystal fibres with prismatic type of calcium oxalate.

**TLC fingerprint:** The Rf- values and colour of the spot shown in Table – 1 and Figure - 4 can identify the stem bark. The physicochemical analysis shows strength and purity of the drug.

### Acknowledgement

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