Anatomical and Phytochemical study of plant parts of *Butea monosperma* (Lamk.) Taub.

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**ABSTRACT**

The plant kingdom is a treasure house of potential drugs and in the recent years there has been an increasing awareness about the importance of medicinal plants. Drugs from the plants are easily available, less expensive, safe and rarely have side effects. The plants which have been selected for medicinal use over thousands of years constitute for examining the current search for therapeutically effective new drugs such as anticancer drugs, antimicrobial drugs, antihepatotoxic compounds. *Butea monosperma* (Lamk.) Taub. is one such medicinal plant commonly known as palas (Flame of the forest) belonging to the family Fabaceae distributed throughout India. It is imperative that any crude drug for pharmacological use, needs to be subjected to scrutiny for botanical identity. The role of anatomical and phytochemical analysis is sought at this juncture to provide a set of diagnostic features of the plant which will help to a considerable extent to ascertain the botanical identification of the drugs. The present study has been carried out to determine the anatomical, phytochemical screening and HPTLC finger printing analysis of different parts of *Butea monosperma* (Lamk.) Taub. was carried out for crude drug identification and for standardization of the quality.

**Keywords:** *Butea monosperma*, anatomy, herbal medicine, morphological study, microscopical study, phytochemical study

**INTRODUCTION**

*Butea monosperma* (Lamk.) Taub. is a medicinal plant commonly known as palas (Flame of the forest) belonging to the family Fabaceae distributed throughout India, except arid parts (Sharma et al., 2000). Different species of *Butea* found in India are *Butea parviflora*, *Butea purpurea*, *Butea monosperma* and *Butea superb* (Almeida, 1998). It is said that this tree is a form of Agnidev, God of Fire. *Butea monosperma* (Lamk.) Taub. has various healing effects which are seen in treatment of many diseases. The bark is an appetiser, lessens inflammation, used in liver diseases.
disorders, fractures, topically in piles and purifies the blood. The leaf is an appetiser, astringent, anthelmintic, aphrodisiac, cures boils and piles. The flowers are astringent, diuretic and aphrodisiac and they are used to disperse swellings. The fruit and the seeds are bitter and oily and useful in piles, eye diseases and inflammation (Kirtikar and Basu, 1984). In the present work, the morphological, microscopical and phytochemical studies of different parts of *Butea monosperma* (Lamk.) Taub. have been carried out which will be useful for proper identification and authentication of crude drug.

**MATERIALS AND METHODS**

*Butea monosperma* (Lamk.) Taub. plant parts (young stem, old stem, bark, leaf, and petiole) were collected. Plant was identified and authenticated from Blatter Herbarium St. Xavier's College, Mumbai. The collected plant parts (leaf, bark and flowers) were dried under shade and powdered with mechanical grinder and stored in air tight containers and used for the study.

**Anatomical study**

Anatomical features were studied by taking transverse sections of young and old stem (T.L.S, R.L.S of stem), leaf and petiole of *Butea monosperma* (Lamk.) Taub. sections were stained by safranine and mounted in glycerine and observed under microscope at 10X (low magnification power) and 45X (high magnification power). Different wood elements were studied using maceration technique and by powder analysis.

**Phytochemical analysis**

The preliminary phytochemical analysis for plant parts (leaf, bark and flowers) of *Butea monosperma* (Lamk.) Taub. was carried out using standard methods (Sofowora, 1993; Trease and Evans, 1989 and Harborne, 1973). The quantitative analysis of secondary metabolites such as flavonoids, saponins and phenols of different parts were also carried using standard methods. HPTLC fingerprint confirmed the presence of flavonoids and saponins in leaf, bark and flowers of *Butea monosperma* (Lamk.) Taub. The separation of flavonoids was achieved using ethyl acetate-formic acid-glacial acetic acid-water (100:11:11:26) and for saponins using mobile phase chloroform-acetic acid-methanol-water (64:32:12:8) (Wagner and Bladt., 1986).

**RESULTS AND DISCUSSION**

**Anatomical study**

**Morphological study**

*Butea monosperma* (Lamk.) Taub. is a medium sized erect tree (12-15m) with crooked trunk and irregular branches (Figure 1.1). Bark is rough and ash coloured. Leaves are trifoliate (10-20cm long and broad), with...
long petiole and stipulate (Figure 1.2). Leaflets are coriaceous. All leaflets are obtuse, glabrous when old, finely silky and with reticulate venation. Flowers are large and produced in rigid racemes (15cm long) (Figure 1.3).

Three flowers together form the tumid nodes of the dark olive-green velvety rachis and pedicels are about twice as long as the calyx and densely brown-velvety. Bracts and bracteoles are small and deciduous. Calyx is long, dark olive-green, densely velvety on outside, clothed with silky hairs within teeth, the upper two are connate and the lower three are equal and deltoid. Corolla (3.5-5cm long) clothed outside with silky, silver hairs and is orange or salmon coloured with broad standard and semi-circular keel, beaked and veined. Pods are stalked, thickened at the sutures and reticulately veined.

**Microscopical study**

**T.S of old stem of Butea monosperma (Lamk.) Taub.**

Old stem of *Butea monosperma* (Lamk.) Taub.is wavy in outline. Old stem shows both extrastelar and intrastelar secondary growth. Extrastelar secondary growth gives rise to periderm made up of phellem, phellogen and phelloderm thus forming a bark. Intrastelar secondary growth shows development of wood (secondary xylem) which is of diffused porous type. In wood vessels are scattered and at some places they are arranged in long radial multiples. Number of vessels ranges from 1-4 in each radial cluster. Wood parenchyma is of paratracheal type where the parenchyma is in direct association with vessels. Secondary phloem shows presence of yellow gummy mass scattered here and there. Medullary rays are formed for radial conduction. Bundle caps of primary

**Figure 1.4: T.S. of old stem**  
**Figure 1.5: T.S. of old stem**  
A - Entire view, B- Secondary growth view  
**Keywords:** Ygm - Yellow gummy mass; Xv - Xylem vessels; Bc - Bundle cap; Mr - Medullary rays; T - Tracheid; P - Periderm

**Figure 1.6: T.S. of young stem**  
**Figure 1.7: T.I.S. of stem**  
**Keywords:** Ygm - Yellow gummy mass; Xv - Xylem vessels; Bc - Bundle cap; Mr - Medullary rays; T - Tracheid; P - Periderm
vascular bundle are pushed towards periphery. Parenchymatous pith is at the center with deposition of yellow gummy mass (Figure 1.4 and 1.5).

**T.S of young stem of Butea monosperma (Lamk.) Taub.**

Young stem of *Butea monosperma* (Lamk.) Taub. is slightly wavy in outline. Cortex is made up of two types of tissues, sclerenchyma forming outer cortex and parenchyma in inner cortex. Few innermost cells of inner cortex show chlorenchymatous tissue just above the bundle caps. Vascular bundles are arranged in ring with wavy outline. Each vascular bundle is conjoint, collateral and open with endarch xylem. Phloem region is broad with deposition of gummy mass. Stele siphonostele, as large parenchymatous pith is at the center which also shows deposition of gummy mass (Figure 1.6).

**T.L.S of stem**

T.L.S of stem of *Butea monosperma* (Lamk.) Taub. shows long uniseriate and multiseriate rays made up of parenchymatous cells (Figure 1.7).

**R.L.S of stem**

R.L.S of stem of *Butea monosperma* (Lamk.) Taub. shows xylem with tracheids and vessels. Radial wall of tracheids shows annular thickening. Vessels are with pitted and reticulate thickening. Other elements like xylem parenchyma and fibers are also found in traces (Figure 1.8)
Powder of old stem after maceration shows vessels with pits on their lateral walls, tracheids with annular thickening, vessels with reticulate thickening, yellow gummy mass and Ca-oxalate crystals (Figure 1.9).

**T.S of leaf**

Leaf of *Butea monosperma* (Lamk.) Taub. shows upper and lower epidermis with unicellular pointed hairs from their surface. Leaf lamina shows dorsiventral structure with palisade and spongy tissue. In midrib region, heterogeneous cortex was observed with sclerenchymatous and chlorenchymatous tissue. In the midrib vascular bundles are in ring with bundle caps on their outer side. Vascular bundles are conjoint and collateral. Center of midrib shows parenchyma with deposits of yellow gummy mass (Figure 1.10).

Powder of leaf shows presence of unicellular pointed hairs, vessels with wall thickening and yellow gummy mass (Figure 1.12 A-D).

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**Figure 1.10: T.S. of leaf**

**Figure 1.11: T.S of petiole**

**Figure 1.12: Powder characteristics of leaf of Butea monosperma* (Lamk.) Taub.**

**Keywords:** A – Vessel; B – Scalariform vessel; C – Yellow gummy mass; D – Unicellular hair
Table 1.1: Preliminary phytochemical analysis of *Butea monosperma* (Lamk.) Taub. leaf, bark and flowers.

<table>
<thead>
<tr>
<th>Phyto- Constituents</th>
<th>Method/Test</th>
<th>Plant parts</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>L</td>
</tr>
<tr>
<td>Triterpenes</td>
<td>Salkowski’s test</td>
<td>+</td>
</tr>
<tr>
<td>Phenol</td>
<td>Ferric Chloride test</td>
<td>+</td>
</tr>
<tr>
<td>Tannins</td>
<td>Ferric Chloride test</td>
<td>+</td>
</tr>
<tr>
<td>Proteins</td>
<td>Xanthoproteic test</td>
<td>+</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>Ammonium test</td>
<td>+</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>Mayer’s reagent and Wagner’s reagent test</td>
<td>-</td>
</tr>
<tr>
<td>Saponins</td>
<td>Froth Emulsion test</td>
<td>+</td>
</tr>
</tbody>
</table>

Keys: + = Present; - = Absent; L = Leaf; B = Bark; F = Flower

Table 1.2: Quantitative analysis of *Butea monosperma* (Lamk.) Taub. leaf, bark and flower.

<table>
<thead>
<tr>
<th>Method/Test</th>
<th>Plant parts collected from Murbad Location</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>L</td>
</tr>
<tr>
<td>Flavonoids</td>
<td></td>
<td>70.00±0.02</td>
</tr>
<tr>
<td>Alkaloids</td>
<td></td>
<td>0.00±0.25</td>
</tr>
<tr>
<td>Phenols</td>
<td></td>
<td>0.83±0.001</td>
</tr>
<tr>
<td>Saponins</td>
<td></td>
<td>29.00±0.001</td>
</tr>
</tbody>
</table>

Values are Mean±S.D. of three determinations

Keys: + = Present; - = Absent; L = Leaf; B = Bark; F = Flowers

T.S. of petiole

T.S of petiole of *Butea monosperma* (Lamk.) Taub. showed circular outline. Internally it is differentiated into epidermis, cortex and stele. Hairs are unicellular and pointed arising from surface of epidermis. Outer cortex is 1-3 layered and sclerenchymatous. While inner cortex is broad and parenchymatous. Stele is horse shoe or wedge shaped in outline. Vascular bundles are with sclerenchymatous cap on the outer side and are conjoint and collateral. Xylem vessels are arranged in radial clusters and are of endarch type. Stele is siphonostele (Figure 1.11).

Phytochemical analysis

**Preliminary Qualitative analysis**

Preliminary qualitative estimation of phytochemicals in powdered plant parts (leaf, bark and flowers) is summarized in (Table 1.1). Leaf, bark and flowers powder of *Butea monosperma* (Lamk.) Taub. showed presence of flavonoids, saponins, phenols and tannins. The powdered leaf and flowers of *Butea monosperma* (Lamk.) Taub. showed presence of protein and triterpenes while absent in the powdered bark of *Butea monosperma* (Lamk.) Taub. The powdered bark and flowers of *Butea monosperma* (Lamk.) Taub. showed presence of alkaloids and absent in leaf powder of *Butea monosperma* (Lamk.) Taub.

**Quantitative analysis**

Quantitative analysis was carried for flavonoids, alkaloids, phenols and saponins content in powdered plant parts (leaf, bark and flowers) of *Butea monosperma* (Lamk.) Taub. The flavonoids content in flowers samples collected was 130mg/g was higher as compared to that of leaf 70mg/g and bark was 50mg/g. Similarly the alkaloid content in flowers was 35mg/g and in the bark was 23mg/g. The saponin content in different plant parts showed very less difference. The saponin content in the flowers samples was 66mg/g, in leaf 29mg/g, and bark 38mg/g. The phenol content in bark 1.26mg/g of catechol, samples was higher than the flowers 0.91mg/g of catechol, and leaf 0.83mg/g of catechol in samples (Table 1.2).
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HPTLC analysis

HPTLC is a valuable tool for the investigation of herbal products with respect to different aspects of their quality (Yamunadevi *et al.*, 2012). HPTLC analysis was tested to obtain reproductive peaks for flavonoids and saponins. Flavonoids constitute one of the most characteristic classes of compounds in plants. They play important role in giving resistance to the plant species (Mishra *et al.*, 2012). The separation of flavonoids was achieved using ethyl acetate-formic acid-glacial acetic acid-water (100:11:11:26) as mobile phase. *Butea monosperma* (Lamk.) Taub. Leafshowed 9 bands. *Butea monosperma* (Lamk.) Taub. bark showed 4 bands. *Butea monosperma* (Lamk.) Taub. flowers showed 11 bands. Yellow coloured fluorescent zone at UV 366nm for flavonoids was observed in the chromatogram and confirmed the presence of flavonoids (Figure 2.1).

Saponins are known as non-volatile, surface active compounds that are widely distributed in nature. They

**Fig. 2.1:** HPTLC fingerprint of flavonoids in *Butea monosperma* (Lamk.) Taub. plant parts (leaf, bark and flowers)

**Fig. 2.2:** HPTLC fingerprint of saponins in *Butea monosperma* (Lamk.) Taub. plant parts (leaf, bark and flowers).
have a diverse range of medicinal properties (Yamunadevi et al., 2012). The methanolic extracts of leaf, bark and flowers of Butea monosperma (Lamk.) Taub. showed presence of different types saponins with different Rf values. The separation of saponins was achieved using chloroform-acetic acid-methanol-water (64:32:12:8) as mobile phase. Butea monosperma (Lamk.) Taub. leaf showed 12 bands. Butea monosperma (Lamk.) Taub. bark showed 9 bands. Butea monosperma (Lamk.) Taub. flowers showed 11 bands. Blue-Violet and Yellow-Brown coloured zones were observed from the chromatogram and showed presence of saponins in the sample (Figure 2.2).

CONCLUSION

Indian system such as Ayurveda and Siddha uses majority of crude drugs that are of plant origin. It is necessary to check the identity of the plant and ascertain its quality before use. A detailed pharmacognostic evaluation therefore is highly essential prerequisite (Dhale, 2011). According to World Health Organisation (WHO) the macroscopic and microscopic description of a medicinal plant is the first step towards establishing its identity and purity and should be carried out before any tests are undertaken (Anonymous, 2002). Thus the present study, the stem of Butea monosperma (Lamk.) Taub. Shows intrastelar and extrastelar secondary growth with prominent diffuse porous wood and periderm formation respectively. Deposition of yellow gummy mass in pith, phloem and periderm is very pronounced. Horse-shoe shaped or wedge shaped stele which is a unique feature found in petiole of Butea monosperma (Lamk.) Taub. These characters may be considered as anatomical features for identification of the plant. It has been observed that the plant parts (leaf, bark and flowers) of Butea monosperma (Lamk.) Taub. showed presence of flavonoids, saponins and phenols in preliminary and quantitative study. In quantitative study the amount of flavonoids content in flowers of Butea monosperma (Lamk.) Taub. was higher as compared to that of leaf and bark. The developed HPTLC fingerprint confirmed the presence of flavonoids and saponins in plant parts of Butea monosperma (Lamk.) Taub. The mobile phase was found to be suitable for the study of flavonoids and saponins separation. The developed phytochemical analysis will help the manufacturers to distinguish the adulterant and standardisation of herbal formulation.

In the present work, an attempt has been made to give an insight on the anatomical features and phytochemical analysis of various parts of the plant Butea monosperma (Lamk.) Taub. This will help in identifying the species and can be further used to determine the botanical identity of herbal medicine.

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