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INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH  
An official Publication of Human Journals

ISSN 2349-7203




Human Journals

Research Article


October 2017 Vol.:10, Issue:3

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## Pharmacognosy, Preliminary Phytochemical Screening and Chromatographic Study of Aerial Parts of *Butea monosperma*



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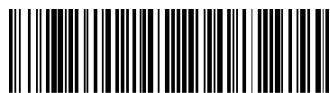


ISSN 2349-7203

**Mariyan R. Patel<sup>1</sup>**

<sup>1\*</sup> *Indukaka Ipcowala pharmacy college, New V.V.  
Nagar, Anand, Gujarat, India.*

**Submission:** 27 September 2017  
**Accepted:** 5 October 2017  
**Published:** 30 October 2017



HUMAN JOURNALS

[www.ijppr.humanjournals.com](http://www.ijppr.humanjournals.com)

**Keywords:** *Butea monosperma*, macroscopy, microscopy, phytoconstituents, Rf (retention factors).

### ABSTRACT

*Butea monosperma* (Fabaceae) is native to India and is found throughout the country. It is known as 'flame of the forest' because of its red colored flowers. This tree species has been of interest to researchers because it is a medicinal plant employed in the Indian indigenous system of medicine. Pharmacognostic standardization, physicochemical evaluation of the aerial parts of *Butea monosperma* was carried out to determine its macro- and microscopical characters and also some insoluble ash, alcohol- and water-soluble extractive values were determined for phytochemical evaluations. Preliminary phytochemical screening was also done to detect different phytoconstituents. The Proximate analysis of powder was also carried out in which extractive value, ash value and foreign matters were determined. Ethanol extract was carried by decoction method and aqueous extract was carried out by maceration method. Preliminary phytochemical screening of various successive extracts of aerial parts was done qualitatively which revealed the presence of sterols, saponins, tannins, alkaloids and carbohydrates.

## 1. INTRODUCTION

*Butea monosperma* which is popularly known as palas in Ayurveda and is distributed in Indian subcontinent areas and Southeast Asia. The flower is best remedy for skin and eye disease, also they are useful in diabetes, urinary disorders, dysentery and swelling. Leaves are used as memory enhancer. The leaves of the *Butea Monosperma* are used as tonic, aphrodisiac and memory enhancer and are also helpful in arresting bleeding or secretion <sup>[1-2]</sup> Literature revealed that pharmacognostic studies have not been reported for the aerial parts of this plant. Therefore the main aim of the present work is to study the macro, microscopic and some other pharmacognostic characters and physicochemical standards of aerial parts of *Butea monosperma* which could be used to discover this plant.

## 2. MATERIALS AND METHODS

### 2.1 Collection of Plant Material

Aerial parts of plant of *Butea monosperma* was collected from the botanical garden of G. J. Patel Institute of Ayurvedic Studies and Research (New V.V. Nagar). The collection was done in the month of February and is authenticated by the Taxonomist, Bioscience Dept., Sardar Patel University, Vallabh Vidyanagar, Gujarat, India.

### 2.2 Pharmacognostic evaluation

#### 2.2.1 Macroscopy

Macroscopical studies of aerial parts of plant were done by naked eye and shape, color, taste and odor were determined and reported.

#### 2.2.2 Microscopy

Pharmacognostical evaluation including histochemical study was carried out by taking free-hand sections according to Wallis and powder studies according to Evans. The section was stained with phloroglucinol and concentrated HCl solution and mounted in glycerin <sup>[3, 4]</sup>. Powder (Sieve mesh 60) of the dried aerial parts was used for the observation of powder microscopical character <sup>[5]</sup>. Photomicrographs were obtained by observing free-hand sections of drug under compound microscope. Results are shown in figure 1 to 6.

### 2.2.3 Physico-chemical evaluations

Physicochemical parameters of *Butea monosperma* plant parts powder were determined and reported as total ash, water-soluble ash, acid-insoluble ash, determination foreign matter, alcohol and water-soluble extractive values were determined to find out the amount of water and alcohol soluble components<sup>[6, 7]</sup>. The moisture content was also determined. Results are shown in Table No. 1.

## 2.3 PHYTOCHEMICAL ANALYSIS:

### 2.3.1 Preliminary phytochemical screening

The dried powdered plant material was successively extracted 70% v/v alcohol in a Soxhlet apparatus. Aqueous extracts were also prepared by using chloroform water I.P. by maceration process. The liquid extracts obtained were collected and the consistency, color, appearance of the dried extracts and their percentage yield were noted. The extracts obtained from powder by decoction and maceration extraction were subjected to qualitative examination for the phytoconstituents like alkaloids, glycosides, carbohydrates, phytosterols, fixed oils, saponins, phenolic compounds, tannins and flavonoids, proteins and amino acids by the reported methods<sup>[7]</sup>. Results are shown in table no. 2 & 3

### 2.3.2 Thin layer chromatography of extracts

The alcohol and water extracts were also subjected to thin layer chromatographic studies on silica gel – G as adsorbent and different detecting reagents using reported methods<sup>[9]</sup>. Results are shown in figure 7 – and table no.4, 5

## 3 RESULTS AND DISCUSSION

### 3.1 Macroscopy

Leaves trifoliate; petiole 7.5-20 cm long with small stipules; leaflets more or less leathery, lateral ones obliquely ovate, terminal one rhomboid-obovate, 12-27 x 10-26 cm, obtuse, rounded at apex, rounded to cuneate at base, with 7-8 pairs of lateral veins. Flowers in racemes, 5-40 cm long, near the top on usually leafless branchlets; calyx with campanulate tube and 4 short lobes; corolla 5-7 cm long, standard, wings and keel recurved, all about the same length, bright orange-red, very densely pubescent.



Figure 1: External morphology of *B. monosperma* aerial parts

### 3.2 Microscopy

#### 3.2.1 Transverse section of leaf

The T.S. of the leaves of *B. monosperma* having upper and lower epidermis covered with cuticle with uni-cellular covering trichomes which are slightly curved at the base and some are blunt. Presence of few glandular trichomes is also there in lower epidermis. Below the upper epidermis is single layer of palisade cells followed by 2-3 layered spongy parenchyma. Midrib region shows 2-5 layers of collenchymatous cells. Stomata are of anomocytic type, mostly on lower the surface. There is a fan shaped vascular bundle in the centre which is surrounded by 3-7 layered pericyclic fiber. Above xylem region, there is a presence of peri-medullary phloem in groups.

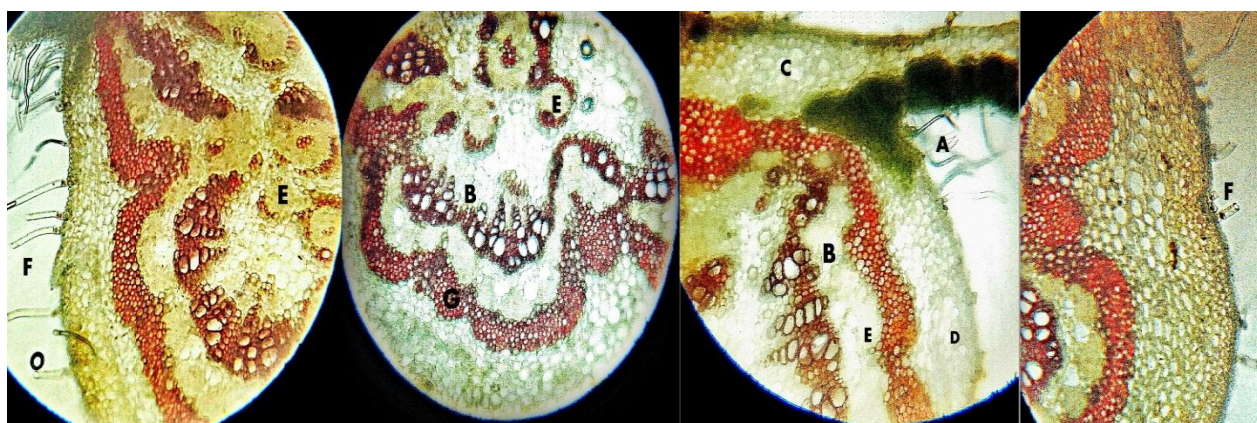


Figure 2: Microscopical view of *B. monosperma* leaf [A-Unicellular trichomes, B- xylem vessels, C- upper collenchyma, D- lower collenchyma, E- Peri-medullary phloem, F- blunt trichomes, G- pericyclic fibers]

### 3.2.2 Transverse section of petiole

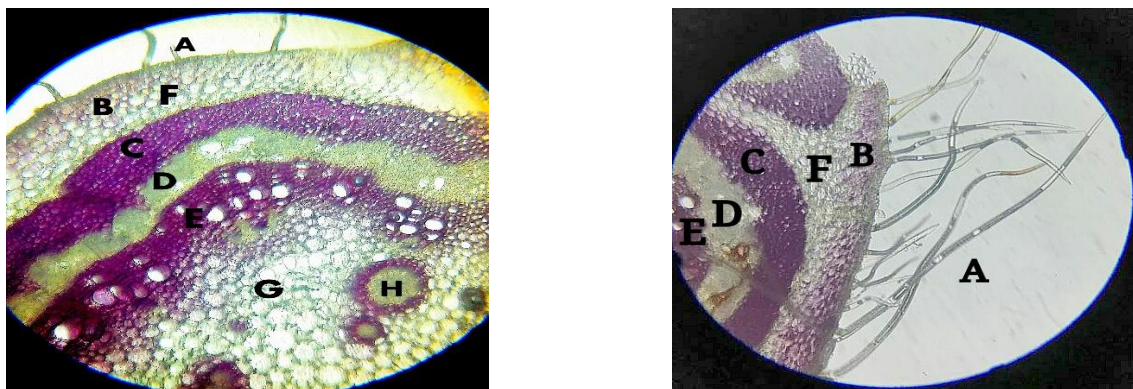


Figure 3: Microscopical view of *B. monosperma* petiole [A-Unicellular trichomes, B- lignified collenchyma, C- pericyclic fiber, D- Phloem, E- Xylem, F- Cortex, G- Pith, H- Peri-medullary phloem ]

### 3.2.3 Transverse section of stem

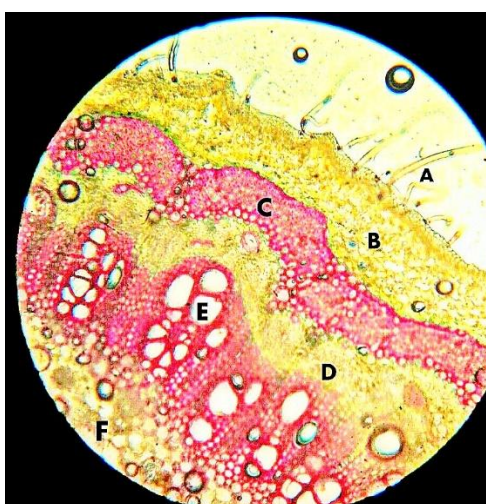


Figure 4: Microscopical view of *B. monosperma* stem [A-Unicellular trichomes, B- cortex, C- pericyclic fiber, D- Phloem, E- Xylem, F- Pith]

### 3.2.4 Powder microscopy of dried powder of aerial parts of *B. monosperma*

The powder of *B. monosperma* leaves and stems was greenish yellow, without characteristic odor and with slightly bitter taste. When powder was mounted with chloral hydrate, phloroglucinol and HCl the following elements were observed:



Figure 5: powder study of leaves of *Butea monosperma* [A: Stomata, B – Unicellular trichome, C: Orange Matter, D: Epidermal cells, E: Anomocytic stomata]

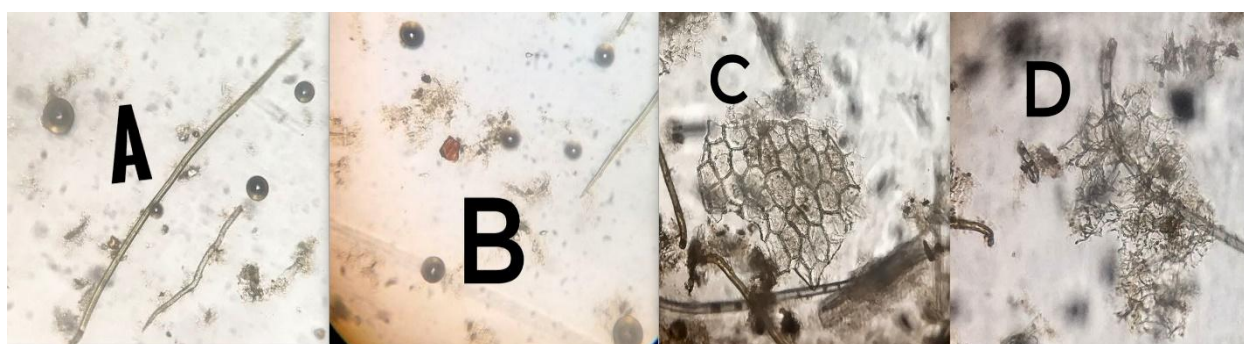


Figure 6: Powder study of flowers of *Butea monosperma* [A: Trichomes, B – tannin cells, C: Epidermal cells, D: Anomocytic stomata]

### 3.2.5 Physico-chemical evaluations

Table 1: Physico-chemical evaluations of aerial parts of *Butea monosperma*

Sr. no.	Parameter	Result [ Leaves]	Result [ Flowers ]
1.	Total ash	0.52	0.67
2.	Water-soluble ash	0.18	0.15
3.	water-insoluble ash	0.3	0.5
4.	Acid insoluble ash	1.85	0.3
5	Water-soluble extractive value	5.6 % w/w	12.8% w/w
6	Alcohol-soluble extractive value	10.6 % w/w	16.4 % w/w
7	Foreign matter	NIL	NIL

w/w - weight/weight

### 3.3 PHYTOCHEMICAL ANALYSIS:

#### 3.3.1 Preliminary profiles of Successive solvent extracts

The powder of *Butea monosperma* L. leaves and flowers were extracted with ethanol by decoction and aqueous extract was prepared by maceration. The results are described in Table no. 2

**Table 2: Preliminary profiles of solvent extracts**

Sr. No.	Part of plant	Extract	Color in daylight	Consistency	% w/w
1	Leaves	Ethanol	Dark green	Semi solid, Sticky	11.6 % w/w
2		Aqueous	Blackish brown	semi solid	7.5 % w/w
3	Flowers	Ethanol	Yellowish	Semi solid	17.4 % w/w
4		Aqueous	Yellowish brown	semi solid	13.8 % w/w

#### 3.3.2 Preliminary phytochemical screening:

All the above extracts were tested with various reagents and the results for the same are reported in table no.3. The various extracts showed the presence of sterols, saponins, tannins, alkaloids and carbohydrates.

**Table 3: Phytochemical Screening**

Sr. No.	Constituents	Leaves		Flowers	
		Ethanol	Water	Ethanol	Water
1.	Phytosterol	-	-	+	+
2.	Saponins	+	+	+	+
3.	Tannins and phenolics	+	+	+	+
4.	Carbohydrates	+	+	+	+
5.	Flavonoids	+	-	+	+
6.	Amino acids	-	-	-	-
7.	Alkaloids	+	-	+	+

3.3.3 TLC study of alcohol and water extract of plant

Table 4: Data of TLC study of flower extract

Sr. No.	Phytoconstituents	Mobile system	Flower extract under visible	
			Alcohol extract Rf value and color of spot	Water extract Rf value and color of spot
A	carbohydrates	Ethyl acetate: water (1:1)	Rf 0.46 (yellow) (under UV yellow) Rf 0.6 (light orange) (under UV blue ) Rf 0.64 (under UV pink ) Rf 0.7 (under UV blue) Rf 0.95 (under UV sky blue)	Rf 0.38 (light brown) (under UV yellow) Rf 0.55 ( light brown) (under UV black ) Rf 0.52 (under UV blue) Rf 0.7 (under UV blue)
B	Tannins	Ethyl acetate: acetic acid: formic acid: water (75:2:3:20)	Rf 0.47 (orange) (under UV blue) Rf 0.57 (yellow) (under UV black ) Rf 0.82 (light brown) (under UV blue )	Rf 0.32 (light brown) (under UV black) Rf 0.86 (light brown) (under UV black )
C	Flavonoids	Ethyl acetate: formic acid: glacial acetic acid: : water (100:11:11:26)	Rf 0.22 (yellow) Rf 0.64 (orange) Rf 0.78 (yellow)	Rf 0.91 (yellowish brown)



**Table 5: Data of TLC study of leaves extract**

Sr. No.	Phytoconstituents	Mobile system	Leaves extract under visible & UV light	
			Alcohol extract Rf value and color of spot	Water extract Rf value and color of spot
A	carbohydrates	Ethyl acetate: water (1:1)	Rf 0.65 ( yellow) (under UV pink) Rf 0.92 (green ) (under UV blood red)	-
B	Tannins	Ethyl acetate: acetic acid: formic acid: water ( 75:2:3:20)	Rf 0.38 (sky blue under UV) Rf 0.92 (Red under UV)	Rf 0.92 (sky blue under UV)
C	Flavonoids	Ethyl acetate: formic acid: glacial acetic acid: water (100:11:11:26)	Rf 0.85 (Green) Under UV Rf 0.85 ( Red) Rf 0.65 (Light green)	-

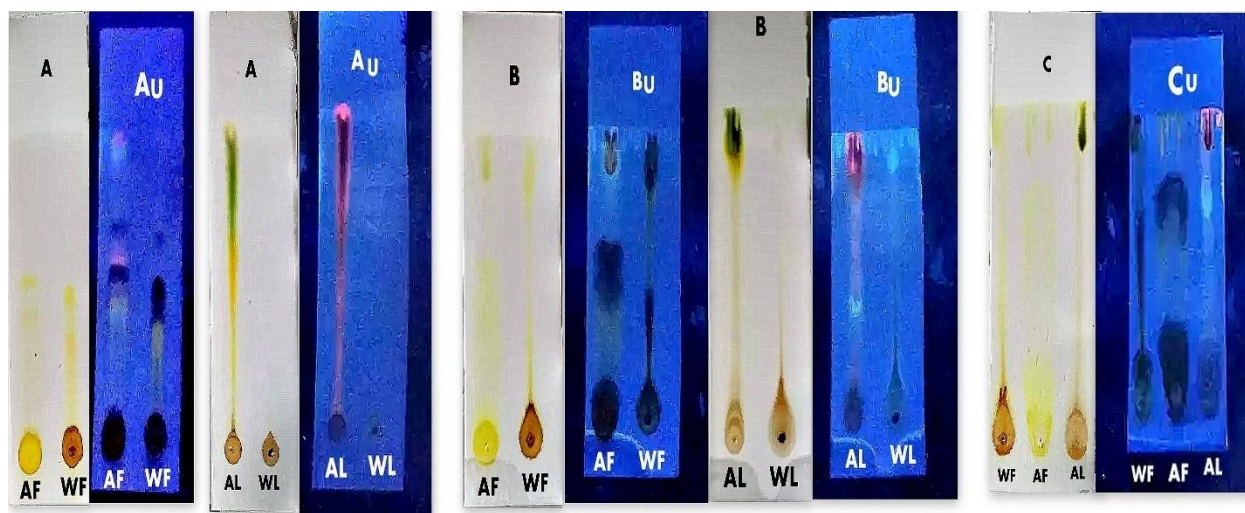


Figure 7: TLC data of various extracts of flower and leaves of *B. monosperma*

[A- Carbohydrates, B- tannins, C- Flavanoids, AU- carbohydrates TLC under UV, BU- Tannins TLC under UV CU- Flavanoids TLC under UV, AF- Alcohol extract of flower, WF- Water extract of flower, AL- Alcohol extract of leaves, WL- Water extract of leaves]

## CONCLUSION



The detailed histological study of *B. monosperma* has been carried out in the present study, which helps to identify this species without any confusion. The most convenient and cost effective method of identification of a medicinal herb would be the use of microscopic characteristics, it has been the foundation of conventional pharmacognosy and remains a fundamental module of the contemporary monograph. Powder microscopy also plays an important role in pharmacognostical evaluation and sometimes may be an identifying parameter of herbal drugs. As there is no pharmacognostical anatomical work on records for this traditionally much valued shrub, present work is taken up in the view to lay down the macroscopic and microscopic standards, which could be used in deciding the genuineness of the herb, irrespective of their collection from different sources. The colored photographs of the leaves, petiole and stem of the above mentioned plant might facilitate the researcher for identification. The results of the phytochemical screening, histochemical tests can be considered as distinguishing parameters to identify and decide the authenticity of *B. monosperma* and thus can be used as standards for reference purpose also. TLC study of above mentioned extract revealed that flower has more constituents present in comparison to

leaves of plant. Column chromatography & HPTLC profile helps in standardization and also for undertaking work on isolating and identifying the bioactive compounds.

## ACKNOWLEDGEMENT

The authors are thankful to Dr. Harsha V. Patel, Principal, Indukaka Ipcowala College of Pharmacy, New V. V. Nagar to provide facilities to perform this research work.

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Author Surname Initials. Title: subtitle. Edition (if not the first). Place of publication: Publisher: Year.

