

MORPHOLOGY, PHYTOCHEMISTRY AND ANTIFUNGAL ACTIVITY OF *Butea monosperma* Lam.

Nafeesa Zahid Malik¹, Mubsher Mazher², Muhammad Riaz³*, Bashir Ahmad⁴ and Qum Qum Noshad⁵

¹Department of Botany, Mirpur University of Science and Technology (MUST), Pakistan

²Department of Botany, Mirpur University of Science and Technology (MUST), Pakistan

³International Islamic University Islamabad PK

⁴International Islamic University Islamabad PK

⁵Department of Botany, Mirpur University of Science and Technology (MUST), Pakistan

*For correspondence: hassan.biotech.iiui1974@gmail.com

ABSTRACT

Extracts of *Butea monosperma* Lam. Were prepared in methanol. Phytochemical analysis of these extracts showed presence of alkaloids and flavonoids in the leaves and saponin, tannin, steroids and falovoids were present in the methanolic extracts of flowers. Antimicrobial activity was accessed by the agar well diffusion method against different fungal pathogens viz. *F. solani*, *C. albicans* and *S. cervisiae*. Methanolic extracts of leaves of *B. monosperma* used at 1mg/ml concentration showed no detectable zone of inhibition. When a concentration of 5mg/ml was used it showed 5.07 ± 0.66 mm, 6.04 ± 0.16 mm and 7.03 ± 0.12 mm zones of inhibition against *F. solani*, *C. albicans* and *S. cervisiae* respectively. At the concentration of 20mg/ml was used it showed 10.0 ± 0.26 mm, 9.0 ± 0.13 mm and 9.66 ± 0.26 mm zones of inhibition against *F. solani*, *C. albicans* and *S. cervisiae* respectively.

Keywords: *Butea monosperma*, Leaf and flower extract, Phytochemical analysis, Antifungal activity.

INTROUCTION

Butea monosperma L. is native to tropical and sub-tropical parts of the Indian Subcontinent and Southeast Asia, ranging across India, Pakistan, Bangladesh, Nepal, Sri Lanka, Myanmar, Thailand, Laos, Cambodia, Vietnam, Malaysia and Western Indonesia. It is common throughout the greater part of India and Burma, up to 1000m (Rana *et al.*, 2012).

Stem of *B. monosperma* is used as aphrodisiac, antidysentery, antiulcer, antitumor, antimicrobial, antifungal, antipyretic, blood purifier and anti-asthmatic. It is also used in bleeding hemorrhoid disorder, dysmenorrhea, hydrocele, liver disorders, gonorrhoea, wound, worm infections, scorpion sting, cough and cold (Pal *et al.*, 2011). The orange dye obtained from the leaves of *B. monosperma* is used as an insecticide. The leaves are further used to pack parcel materials. The seeds are used in Ayurvedic and Unani medicine for treating different disorders (Orwa *et al.*, 2009).

B. monosperma is traditionally used as antitumor, anti-inflammatory, antidiabetic, antimicrobial, diuretic, anthelmintic, appetizer, astringent carminative, aphrodisiac, stomach disorders, diabetic sore throat, irregular bleeding during menstruation, flatulent colic, cough and cold (Malpani *et al.*, 2011). Seed of *B. monosperma* is used in inflammation, bleeding piles, urinary stones, skin and eye diseases, abdominal troubles, intestinal worms and tumour. When seeds are pounded with lemon juice and applied to the skin, they act as a rubefacient. Gum is used in stomatitis, corneal apacitis, ring worm, leucorrhoea, septic sore throat, excessive perspiration (Mazumder *et al.*, 2011). The leaves of *B. monosperma* contain glucoside, kino-oil containing oleic and linoleic acid, palmitic and lignoceric acid (Nadkarni *et al.*, 2002).

OBJECTIVES

- To evaluate the morphology of *B. monosperma*.
- To estimate the presence of different chemical compounds in *B. monosperma*.
- To study antifungal activity of *B. monosperma*.

MATERIAL AND METHODS

PHYTOCHEMICAL ASSAY

The methods of phytochemical analysis were those described by Kandelwal (2007) and Kokate (2000) for the presence of various active compounds.

Drying of leaves and flowers

The leaves and flowers of *B. monosperma* were shade dried until all the water molecules evaporated and they became well dried for grinding. After drying the plant materials were ground well using mechanical blender into fine powder and transfer into airtight containers with proper labeling for future use.

Preparation of Crude Extracts

The 20gm of powdered leaves and flowers of *B. monosperma* were separately soaked in 100ml of methanol solvent in two conical flasks. The flasks containing the powder were shaken, corked and left to stand for 48 hours at room temperature. Then each extract of flowers and leaves of *B. monosperma* were screened for the presence of various secondary metabolites (phytochemicals).

ANTIFUNGAL ACTIVITY

The extracts were also screened for their antifungal activity in-vitro by using standard well diffusion method given by Toit and Rautenbach, (2000).

Sterilization of Materials

All glassware were washed with detergent and rinsed with distilled water properly 2-3 times. These were then air dried before wrapping with aluminum foil and sterilized in autoclave.

Preparation of PDA

Suspend 9.8 grams of PDA medium in 250 ml of distilled water into a beaker. Mix well and dissolve by heating with frequent agitation. Boil for one minute until complete dissolution. Dispense into appropriate container and sterilize in autoclave at 121 °C for 15 minutes. The prepared medium should be stored at 2-8 °C. The colour is amber, slightly opalescent. The dehydrated medium should be homogeneous, free-flowing and beige in colour. If there are any physical changes, discard the medium.

Collection of Plant Material

The leaves of *B. monosperma* were collected from different regions of district Bhimber. The fresh *B. monosperma* leaves were washed thoroughly 2-3 times with tap water and with distilled water, chopped into small pieces, air-dried and grinded into fine powder before use.

Preparation of Plant Material

The leaves of *B. monosperma* were extracted with cold methanol. This was prepared by using the method as described by (Oyagade *et al.*, 1999). This was carried out by suspending 10 grams of the finely ground leaves in 95% methanol. The methanolic extraction was done at $28 \pm 1^\circ\text{C}$ for 120 hours. The extract was then decanted and filtered through a Whatmann filter paper. The filtered extract was sterilized using a membrane filter and evaporated to dryness at 45°C . The residues obtained were reconstituted in 95% ethanol at stock concentration of 10 mg/mm. The extract solution was then stored in refrigerator at $4 \pm 2^\circ\text{C}$ (Omojosola and Awe, 2004).

Antifungal activity

The extracts were screened for its antifungal activity in comparison with standard antibiotic ketoconazole (mg/ml) in-vitro by well diffusion method. Lawn culture was prepared using the test organism on potato dextrose broth (PDA). The inoculated plates were kept aside for a few minutes. Using sterilized micropipettes 30 gm/L of solvent with selected *B. monosperma* leaves extract was added in to the well. The plates with yeast like fungi were incubated at 37°C for overnight.

RESULTS

Table 1: Qualitative Morphological characters of Leaf of *Butea monosperma* Lam.

Plant Part	Morphological Characters	Observation	
		Fresh	Dried

Leaves	Colour	Green	Light Green
	Arrangement	Compound	Compound
	Margins	Undulate	Undulate
	Venation	Reticulate	Reticulate
	Base	Cuneate	Cuneate
	Apex	Obtuse	Obtuse

Table illustrates the morphology of fresh and dried leaves of *B. monosperma*.

Table 2: Qualitative Morphological characters of Flower of *Butea monoaperma* Lam.

Plant part	Morphological characters	Observation	
		Fresh	Dried
Flower	Colour	Orange red	Light Orange
	Type	Asymmetric	Asymmetric
	Inflorescence	Receme	Receme
	Colour of petals	Orange	Light Orange

	Colour of sepals	Olive green-brown	Olive green-brown
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Table illustrates the morphology of fresh and dried leaves of *B. monosperma*.

Table 4: Phytochemicals from methanolic extracts of leaves and flowers of *B. monosperma*

Sr. no	Phytochemicals	Leaves	Flowers
1	Alkaloids	+	–
2	Saponins	–	+
3	Tanins	–	+
4	Steroids	–	+
5	Flavonoids	+	+
6	Terpenoids	+	–
7	Glycosides	+	+
8	Carbohydrates	+	+

Key: + = present and - = absent

Table 5: Antifungal activity of methanolic extracts of *B. monosperma* Leaves

Microorganisms	Zone of inhibition after 48 hours		
	Methanolic extract		
	Concentration		
	1mg/ml	5mg/ml	20mg/ml
<i>F. solani</i>	0.00±0.00	5.07±0.66	10.0±0.26
<i>C. albicans</i>	0.00±00	6.03±0.16	9.0±0.13
<i>S. cerevisiae</i>	0.00±00	7.03±0.12	9.66±0.26

Mean ± S.E.

95% Confidence level ($\alpha = 0.05$)

DISCUSSION

Plant parts were collected and morphological character of fresh and dried *B. monosperma* plant parts i.e. flowers and leaves were observed. Fresh leaves are green in colour while the colour of dried leaves was dull green. The length of both fresh and dried leaves was measured as the length of fresh leaves have an average of 12.8cm while the length of dried leaves was 11.6cm. So the

dried leaves shrink in size as compared to the fresh leaves. Moreover the leaves of this plant are compound having three leaflets which have undulate margins, reticulate venation, cuneate base and obtuse apex. Findings of present study are in accordance with Das *et al.*, 2011.

The flowers are asymmetric and colour is orange-red while the dried flowers were light orange in colour. The calyx of both fresh flower and dried flower were olive green to brown and velvety from outside. The corolla is 3.8-5cm in length. Present findings are coherent with Burli *et al.*, 2007.

Qualitative phytochemical analysis of *B. monosperma* confirmed the presence of various phytochemicals like saponins, terpenoids, steroids, alkaloids and tannins. These results are in accordance with Deshmukh *et al.*, 2014. However present study showed no anthraquinones whereas Deshmukh *et al.*, 2014 found anthraquinones too.

The methanolic extract of *B.* showed best antifungal activity against tested organisms. No activity was shown by the extracts at 1mg/ml, while at the concentration of 5mg/ml and 20mg/ml *F. solani*, *C. albican* and *S. cerevisiae* showed the inhibition zones ranging from 5.07±0.66 cm to 10.0±0.26 cm. Methanolic extract was more potent against the tested pathogens at the concentration of 20mg/ml. The results shown by present research are significantly different from Chintan *et al.*, 2014. Present research found no zones of inhibition at 1mg/ml concentrations whereas Chintan *et al.*, 2014 showed some zones of inhibition at 1mg/ml concentrations. Present research has shown high zones of inhibition at 20mg/ml concentrations as compared to Chintan *et al.*, 2014.

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