A novel approach towards evaluation of primary metabolite and antimicrobial screening in *Butea monosperma* (Lamarck.) Kuntze.

Divya Fageria, D.V. Rao and Renu Kumari

Laboratory of Bioactive compounds and Biotechnology, Department of Botany, University of Rajasthan, Jaipur-302004, India.

**ABSTRACT**

The quantification of primary metabolites and antimicrobial activities of medicinal plant *Butea monosperma* (Lam.) kuntze against clinical isolates was evaluated. The present study was aimed to determine the antibacterial and antifungal activities of the chloroform and ethanolic extract of leaf, flower, root and seed of the *Butea monosperma* (Lam.) kuntze. Antimicrobial activity was determined by using agar well diffusion method. Ethanolic extracts of leaves and flowers possessed highest antibacterial activity against *E.coli* and highest antifungal activity against *Trichoderma reesei* and *Fusarium oxysporum*. Chloroform extracts of seeds have the highest antibacterial activity against *Bacillus subtilis* and highest antifungal activity was observed in chloroform extracts against *Fusarium oxysporum*. The present results showed potential of this medicinal plant which can be used as herbal drug as therapeutic ventures in future aspects.

**Keywords**

*Butea monosperma* (lam.) Kuntze, Primary metabolites, Antimicrobial activity, Activity index.

**Article history:**

Received: 28 July 2017;
Received in revised form: 23 September 2017;
Accepted: 6 October 2017;

**Keywords**

*Butea monosperma* (lam.) Kuntze, Primary metabolites, Antimicrobial activity, Activity index.

**Article history:**

Received: 28 July 2017;
Received in revised form: 23 September 2017;
Accepted: 6 October 2017;

**Material and Methodology**

**Plant material:** Healthy plants of *B. monosperma* were collected from Amer, Delhi road, Jaipur and authenticated by the Herbarium (RUBL.211650), University of Rajasthan, Jaipur, Rajasthan, India;

- **RUBL**—Rajasthan University Botanical Library

**Chemical:** All the chemicals and growth regulators used are analytical grade and purchased from Hi Media Pvt. Ltd., Mumbai, India.

**Quantification of Primary metabolites**

The flower, root, seed and leaf parts of *B. monosperma* were evaluated quantitatively to estimate the total levels of soluble sugars, Loomis and Shull(1937), starch, McCready (1950), proteins, Lowery et al.,(1951), lipids, Jayaraman (1981) and phenols, Bray and Thorpe(1954) following the established methods for the sugars, starch, lipid, protein and phenol. All experiments were repeated five times for precision and values were expressed in mean ± standard deviation in terms of shade dried material.

**Antimicrobial activity of *B. monosperma***

Antibacterial and Antifungal activities of the plant extracts were tested using Agar well diffusion method of Perez et al.,(1990) and Bonzar et al., (2005) respectively. The zone of inhibition was calculated by measuring the diameter of the inhibition zone around the well (in mm) including the well diameter.

**Preparation of Extracts:**

The flowers, leaves, seeds and roots of *B. monosperma* were dried and powdered for experimentation. The resultant was then subjected to extraction with methanol in Soxhlet apparatus. The extracts were then concentrated in vacuum under plates were incubated at 27°C for 24 hrs and fungal plates at 24°C for 72hrs. The diameter of the minimum zone of inhibition was recorded and calculated in mm.
Result and Discussion:

All the plant parts of Butea monosperma were evaluated quantitatively for the analysis of total soluble sugars, starch, protein, phenol, lipid and ascorbic acid. Table 1:

Table 1. Estimation of primary metabolites (mg/gdw) in different parts of Butea monosperma (mg/gdw).

<table>
<thead>
<tr>
<th>Experiments</th>
<th>Plant Parts</th>
<th>Sugar</th>
<th>Starch</th>
<th>Protein</th>
<th>Phenol</th>
<th>Lipid</th>
</tr>
</thead>
<tbody>
<tr>
<td>Root</td>
<td>5</td>
<td>7</td>
<td>36</td>
<td>1.98</td>
<td>46</td>
<td></td>
</tr>
<tr>
<td>Flower</td>
<td>14</td>
<td>16</td>
<td>250</td>
<td>7.56</td>
<td>12</td>
<td></td>
</tr>
<tr>
<td>Leaf</td>
<td>10.4</td>
<td>22</td>
<td>290</td>
<td>9.8</td>
<td>8</td>
<td></td>
</tr>
<tr>
<td>Seed</td>
<td>9.6</td>
<td>24</td>
<td>100</td>
<td>3.41</td>
<td>32</td>
<td></td>
</tr>
</tbody>
</table>

mg/gdw = milli gram per gram dry weight

Biochemical studies of the individual plant parts is a necessary prerequisite in order to evaluate their importance in the overall metabolism of the plant, as well as the role of specific substances that may be produced as direct or indirect products of metabolism in same physiological processes. Hence, carbohydrates, proteins, amino acids, chlorophyll, vitamins, hormones, phenols etc., are very essential for plant without which the plant life is hampered. In the present investigation among various primary metabolities maximum total soluble sugars were observed in flowers (14mg/gdw) and starch were observed in seeds (24 mg/gdw), while minimum in root (7mg/gdw). Leaves had maximum protein content (290 mg/gdw) and roots had minimum (36mg/gdw). Maximum lipids were found in roots (46 mg/gdw), while content in leaves was minimum (8mg/gdw). Phenols were maximum in seeds (7.56mg/gdw) and minimum in roots (1.98mg/gdw).

Antimicrobial activity

The maximum zone of inhibition in bacteria culture was observed in chloroform extract of seed against Bacillus subtilis (24 mm) and minimum was observed in ethanolic extract of Bacillus subtilis (10 mm) while in the case of fungal strains the highest zone of inhibition was shown against Fusarium oxysporum in chloroform extract (18mm) of leaves whereas minimum in chloroform extract of roots against Aspergillus niger (4mm).

Evaluation of antibacterial activity

Table 2: Antimicrobial activity of chloroform extracts of B. monosperma.

<table>
<thead>
<tr>
<th>S.No</th>
<th>Bacterial colonies</th>
<th>Bacillus Subtilis</th>
<th>E. coli</th>
<th>Streptomyces griseus</th>
<th>Staphylococcus aureus</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flowers</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>Leaves</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>Seeds</td>
<td>IZ-24</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>Roots</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
</tbody>
</table>

Table 3. Antimicrobial activity of Ethanol Extract of B. monosperma.

<table>
<thead>
<tr>
<th>S.No</th>
<th>Bacterial colonies</th>
<th>Bacillus Subtilis</th>
<th>E. coli</th>
<th>Streptomyces griseus</th>
<th>Staphylococcus aureus</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flowers</td>
<td>IZ-11</td>
<td>IZ-16</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>Leaves</td>
<td>NA</td>
<td>IZ-12</td>
<td>NA</td>
<td>IZ-18</td>
<td></td>
</tr>
<tr>
<td>Seeds</td>
<td>IZ-10</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>Roots</td>
<td>IZ-11</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
</tbody>
</table>

Standard: Ciprofloxacin -20mm IZ= Inhibition zone (in mm) AI- activity index = IZ of test sample / IZ of standard.

Table 4. Antifungal activity of Chloroform extract of B. monosperma.

<table>
<thead>
<tr>
<th>S.No</th>
<th>Fungal colonies</th>
<th>Trichoderma reesii</th>
<th>Aspergillus niger</th>
<th>Penicillium funiculosum</th>
<th>Fusarium oxysporium</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flowers</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>IZ-12</td>
<td></td>
</tr>
<tr>
<td>Leaves</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>IZ-18</td>
<td></td>
</tr>
<tr>
<td>Seeds</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>Roots</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
</tbody>
</table>

Table 5. Antifungal activity of Ethanolic extract of B. monosperma.

<table>
<thead>
<tr>
<th>S.No</th>
<th>Fungal colonies</th>
<th>Trichoderma reesii</th>
<th>Aspergillus niger</th>
<th>Penicillium funiculosum</th>
<th>Fusarium oxysporium</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flowers</td>
<td>IZ-16</td>
<td>NA</td>
<td>NA</td>
<td>IZ-16</td>
<td></td>
</tr>
<tr>
<td>Leaves</td>
<td>IZ-14</td>
<td>NA</td>
<td>NA</td>
<td>IZ-12</td>
<td></td>
</tr>
<tr>
<td>Seeds</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>Roots</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td>NA</td>
<td></td>
</tr>
</tbody>
</table>

Standard: Ketokonazole-22mm (fungus) IZ= Inhibition zone (in mm) AI- activity index = IZ of test sample / IZ of standard.

Discussion

Herbal drugs have gained importance in recent years because of their efficacy and cost effectiveness. The continuous evolution in bacterial resistance to currently available antibiotics has necessitated the search for novel and effective antimicrobial compounds. The first step towards these goals is the in vitro antibacterial activity assay Tanwer et al., (2010). Many primary metabolites lie in their impact as pharmacologically active metabolites in pharmaceutical compounds (Sagwan et al., 2011).

Plant synthesizes primary metabolites (lipid, protein, starch, phenol etc for the normal growth and development of itself. Polysaccharides extracted from Chinese medicinal herbs possess immunnunomodulatory and antimicrobial activity (Wong et al., 1994). Bioactivity of carbohydrates derivatives has also been reported by Nobmann (2009).Further work to isolate and characterize the active compounds responsible for this activity in the plant is recommended in future.

Conclusion

The results of the present investigation clearly indicate that the study of primary metabolites and antimicrobial activity of this plant could consider it as a natural herbal source. Thus, the study ascertains the value of plants used in Ayurveda, which could be of considerable interest to the development of pharmaceutical drugs.

References

Trichostrongylid nematodes in sheep. Fitoterapia, 77, 2006, 137–140.


Bandara, B. M. R., Kumar, N. S., Wimalasiri, K.M.S. Journal of the National Science Council of Sri Lanka 18, 97- 103.