Antibacterial Activity in Methanolic Leaves Extract of *Madhuca indica* L.

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1. Introduction

*Madhuca indica* belonging to family Sapotaceae is an important economic tree growing throughout INDIA. Traditionally *Madhuca indica*. Leaves have been used as an Anti-diabetic, Rheumatism, Ulcers, Bleedings and Tonsillitis. The flowers, seeds and seed oil of *Madhuca* have great medicinal value.Externally, the seed oil massage is very effective to alleviate pain. In skin diseases, the juice of flowers is rubbed for oleation. It is also beneficial as a nasya (nasal drops) in diseases of the head due to pitta, like sinusitis. The purpose of the present study was to evaluate the in vitro antibacterial activity of the methanol leaves extract of *Madhuca indica*. The external applications are skin effections, analgesic anti pyretic, antioxidant and anti diabetic.

2. Materials and Methods

2.1 Plant material  
Disease free leaves were collected from the campus of Government Institute of Science, Aurangabad. The collected leaves were surface sterilized with 0.1% mercuric chloride & then washed with D/W 2-3 times separately & shade dried. Fine powder were made after complete drying and used for the experimental work.

2.2 Solvent Extraction of Leaves  
Extracts were made in 80% methanol at room temperature by simple extraction method (*Deshpande et al*). 10 gm dried powder of leaves mixed with 100ml solvent in 250 ml flask and were kept on shaker for 24 hrs. Then it was allowed to stand for the 30 min to stand the plant material. Thereafter it was filtered & centrifuged at 5000 rpm for 15 min .The supernatant was collected & solvent was evaporated at 45 °C in vacuum evaporator to make the final volume 1/5 of the original volume.

2.3 Determination of Antibacterial activity  
2.3.1 Culture media  
For antibacterial test Nutrient Agar/broth was purchased from Hi-Media Pvt. Ltd Bombay, India.

2.4 Inoculums Preparation  
The bacteria were inoculated into Nutrient broth &Incubated at 37°C for 18 hrs & suspension was checked to provide approximately 3x 10⁷ cfu/ml.

2.5 Microorganism Used  
The pure culture of test microorganism Bacillus cereus, *E. coli*. Staphylococcus aures, *P. auroginosa* was isolated from different samples in
2.6 Antibacterial assay
The Antibacterial activity of methanolic extract analysed by using disc diffusion assay (Dugler and Gomez, 2004) sterile disc was used for the present investigation. The extracts were incorporated to the sterile disc individually with 100, 250, 500, and 1000 µl using micropipette. Precautions were taken to prevent the flow of solvent extract from the disc outer surface. commercial antibiotic streptomycin (20 ml) was used foe the positive control. The Disc was placed on the nutrient agar plates in which the bacteria were inoculated and spread uniformly and incubated at 37°C + 1°C for 24 hrs. The diameter of zone of inhibition was measured in mm.

Table 3: Antimicrobial activity of methanolic extract of leaves of Madhuca indica

<table>
<thead>
<tr>
<th>Sr.No.</th>
<th>Test Microorganism</th>
<th>Zone of inhibition (mm)</th>
<th>Zone of inhibition (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Conc. of methanolic leaf extract (µl)</td>
<td>Streptomycin (µl)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>250</td>
<td>500</td>
</tr>
<tr>
<td>1</td>
<td>E.coli.</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>2</td>
<td>S.aures</td>
<td>+++</td>
<td>+++</td>
</tr>
<tr>
<td>3</td>
<td>B. cereus</td>
<td>+++</td>
<td>+++</td>
</tr>
<tr>
<td>4</td>
<td>P.auroginosa</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

3. Results and Discussion
Alcoholic extracts of leaves of Madhuca indica, were screened for antimicrobial activities against Staphylococcus aureus, Bacillus subtilis, Escherichia coli, Pseudomonas aeruginosa, and at dose level ranging from 100 µg/ml, 250 µg/ml, 500 µg/ml and 1000 µg/ml. (Table 1) methanolic extracts of Madhuca indica, leaves inhibited all the bacterial strains tested. All the doses (100 µg/ml to 1000 µg/ml) showed zone of inhibition against all the bacteria, even at a dose of 100µg/ml of extract exhibited significant zone of inhibition comparable to standard antibiotic (streptomycin) against S.
aureus. For leaf extract B. cereus exhibited maximum inhibition (18 mm) which was greater than that of standard. For all other bacteria 100 µg/ml concentration of the extract was sufficient to produce effective inhibition. Concentration which were comparable to standard antifungal agent Streptomycin. Thus leaves of alcoholic extracts of Madhuca indica, were found to be inhibitory against all the bacteria tested. Antibacterial activities of alcoholic extracts of leaves of Madhuca indica, could be attributed to the presence of biological compounds like 2-Furan methanol, 4H pyran 4-one, 2,3-dihydro 3,5-dihydroxy-6-methyl, Thiophene, 2-Furan carboxy aldehyde-5-(hydroxymethyl) and 1,4-tetra decanediol. The use of medicinal plants play a vital role in covering the basic health needs in developing countries and these plants may offer a new sources of antibacterial, antifungal and antiviral agents with significant activity against infective microorganisms.

**Conclusion**

The present study indicates that Madhuca indica, extracts have broad inhibitory activities to pathogenic microorganism and to act as potential antibacterial agent from natural sources. In general, commercial antibiotic and antifungal drugs causes side effects such as liver, kidney and gastrointestinal tract toxicity. Severe hepatotoxicity had also been reported in patients undergoing antifungal drug therapy. However, herbal remedies often do not produce any side effects. Therefore, alternative medicine become popular remedy to various types of ailments In conclusion, Madhuca indica extracts have revealed significant antibacterial activities against test organisms used for the study.

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