Antibacterial screening of *Ficus palmata* Forsk. pure latex and its methanolic and ethanolic extracts

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Received 9 September 2012; Accepted 24 January 2013

The antibacterial activity of various extracts of the latex of *Ficus palmata* Forsk. was studied against two bacteria (*Escherichia coli* and *Staphylococcus aureus*) using disc diffusion method. Methanolic extract of latex showed significant antibacterial activity against both the test organisms. The zone of inhibition by latex was found to be significantly higher than the zone of inhibition shown by tetracycline. The magnitude of antibacterial activity of latex and its extracts found to be more against *Escherichia coli* (Gram-negative bacterium) as compared to *Staphylococcus aureus* (Gram-positive bacterium).

**Keywords:** *Ficus palmata*, Antibacterial activity, Latex, Zone of inhibition, Activity Index.

**IPC code; Int. cl. (2011.01)−** A61K 36/00.

**Introduction**

Human beings have been utilizing plants for basic preventive and curative health care since time immemorial. Over 9,000 plants have been known for medicinal applications in various cultures and countries and this is without having conducted comprehensive research amongst several indigenous and other communities1. *F. palmata* is a small tree belonging to family Moraceae (Plate 1a). This plant is distributed in Nepal, Somalia, South Egypt, Peninsula and India2. The plant is commonly found in the lower and hot areas of Himachal Pradesh. *F. palmata* Forsk. is used as fuelwood and traditionally used for the effective treatment of many diseases3,4, viz. skin diseases, ringworm, wound infections and haemorrhoid5-9. In last couple of years a lot of research work has been carried out in order to find the pharmacological proves of traditionally used medicinal plants. Several workers have studied antibacterial activities exhibited by extracts of various parts of different species belonging to genus *Ficus* L. such as leaf extract, stem bark, crude extract, flavonoids etc.10-13 To the best of our knowledge no attempt has been made yet to study the antibacterial activity of latex of *F. palmata*. Keeping in view the importance of latex of this plant, the present research work was aimed to study the antibacterial activity of pure latex and its extracts (methanolic and ethanolic) against two bacterial strains causing various diseases in human beings *i.e. Escherichia coli* and *Staphylococcus aureus*.

**Materials and Methods**

The collected plant specimen was authenticated by Dr. Suresh Kumar Dept. of Botany, Abhilashi Institute of Life Sciences and the specimen was deposited in herbarium section of the institute. The fresh latex of *F. palmata* was collected from village Tanda, Distt. Mandi, Himachal Pradesh (N: 31°35’27.73”, E: 76°53’21.76”, H: 746 m msl) (Plate 1b). The latex was collected in the form of liquid exudates from stem and branches of tree in the month of April, 2012. The fresh latex was stored in sterilized coloured reagent bottles at 4°C.

**Preparation of plant extract**

Latex collected from *F. palmata* stem was used for preparing extracts. Further, the known quantity of latex (1 mL) was mixed with varying amount of ethanol and methanol (1, 2, 3, 4, 5 and 6 mL) for
preparing extracts. Then the mixtures were placed in autoshaker with very low speed for over night and filtered through Whatman’s filter paper (No. 1) for using against the test micro-organisms.

**Bacterial Culture Preparation**

In the present study two bacterial strains *E. coli* and *S. aureus* were procured from Department of Microbiology, Abhilashi Institute of Life Sciences, Tanda, Distt. Mandi, Himachal Pradesh. Bacterial strains used in this study were firstly cultured in nutrient broth, incubated for 24 h in incubator shaker at 120 rpm at 37°C ± 1°C.

**Determination of antibacterial activity**

The disc of Whattman’s filter paper having diam. of 6 mm was saturated with extract of known quantity. Sterile nutrient agar and Eosin Methyl Blue plates were inoculated with tested organism and allowed the plates to dry and then discs were placed. These plates were incubated at 37°C ± 1°C for 24 h in incubator, after which the zone of inhibition measured and compared with the zone of inhibition shown by tetracycline.

**Determination of activity index**

The activity index \(^{14}\) of the crude plant extract was calculated as

\[
\text{Activity index (A.I.)} = \frac{\text{Mean of zone of inhibition}}{\text{Zone of inhibition obtained for standard antibiotic drug}}
\]

**Statistical Analysis**

All experiments were carried out in triplicate. Data analysis was done by using MS office 2010. Data are presented as arithmetic mean and the results obtained were analyzed in terms of standard deviation.

**Results and Discussion**

Pure latex of *F. palmata* showed inhibition zone of 8 mm against *E. coli* with activity index 1.14 and did not show antibacterial activity against *S. aureus*. Methanolic extract of latex showed antibacterial activity against both the test organisms. It is evident from the result that the methanolic extract against *E. coli* at concentration 1:4 mL (latex: methanol) exhibited maximum inhibition zone 13.3 mm (Plate 2a), whereas extract concentrations 1:2 mL and 1:6 mL showed 9.3 mm (Plate 2b) and 7 mm zone of inhibition, respectively as compared to standard tetracycline i.e. 7 mm zone of inhibition (Plate 2c). Minimum inhibition zone 5.3 mm has been observed at extract concentration 1:1 mL. Activity index for various methanolic extracts against *E. coli* varied from 1.90 (maximum) to 0.76 (minimum).

In case of *S. aureus* methanolic extract 1:5 mL exhibited maximum antibacterial activity with 7.3 mm zone of inhibition (Plate 2d) as compared to standard tetracycline (5 mm zone of inhibition) (Plate 2e). Inhibition zone of 6.0, 5.0 and 4.0 mm has been observed with latex concentrations 1:2 mL, 1:3 mL (Plate 2f) and 1:4 mL, respectively. Methanolic extract of concentrations 1:1 mL and 1:6 mL found to be ineffective against *S. aureus* and activity index varies from 1.46 to 0.80 (Table 1).
Plate 2 (a-f) – Antibacterial activity of *Ficus palmata* Forsk. latex and its extracts against *E. coli* and *S. aureus*: a. Methanolic latex extract (1:4 mL); b. Methanolic latex extract (1:2 mL); c. Tetracycline; d. Methanolic latex extract (1:5 mL); e. Tetracycline; f. Methanolic latex extract (1:3 mL).

Table 1—Antibacterial activity of the latex and its extracts of *Ficus palmata* Forsk.

<table>
<thead>
<tr>
<th>S. No</th>
<th>Concentration (latex: methanol) Or (latex: ethanol) (mL/mL)</th>
<th>Test Bacterial spp.</th>
<th>Escherichia coli</th>
<th>Staphylococcus aureus</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Zone of Inhibition (mm) Mean (± SD)</td>
<td>Activity Index</td>
<td>Zone of inhibition (mm) Mean (± SD)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>7.0 (± 0.0)</td>
<td>-</td>
<td>5.0 (± 0.0)</td>
</tr>
<tr>
<td>Tetracycline Pure latex</td>
<td></td>
<td>8.0 (± 0.8)</td>
<td>1.14</td>
<td>6.0 (± 0.8)</td>
</tr>
<tr>
<td>1</td>
<td>1:1</td>
<td>5.3 (± 0.9)</td>
<td>0.76</td>
<td>5.0 (± 0.8)</td>
</tr>
<tr>
<td>2</td>
<td>1:2</td>
<td>9.3 (± 1.2)</td>
<td>1.32</td>
<td>4.0 (± 0.8)</td>
</tr>
<tr>
<td>3</td>
<td>1:3</td>
<td>6.3 (± 0.4)</td>
<td>0.90</td>
<td>7.3 (± 1.2)</td>
</tr>
<tr>
<td>4</td>
<td>1:4</td>
<td>13.3 (± 3.3)</td>
<td>1.90</td>
<td>NIZ</td>
</tr>
<tr>
<td>5</td>
<td>1:5</td>
<td>6.3 (± 0.4)</td>
<td>0.90</td>
<td>NIZ</td>
</tr>
<tr>
<td>6</td>
<td>1:6</td>
<td>7.0 (± 0.4)</td>
<td>1.00</td>
<td>NIZ</td>
</tr>
<tr>
<td>Methanolic extract of latex</td>
<td></td>
<td>7.0 (± 2.1)</td>
<td>1.00</td>
<td>NIZ</td>
</tr>
<tr>
<td>1</td>
<td>1:1</td>
<td>7.3 (± 0.4)</td>
<td>1.04</td>
<td>NIZ</td>
</tr>
<tr>
<td>2</td>
<td>1:2</td>
<td>7.6 (± 0.4)</td>
<td>1.08</td>
<td>NIZ</td>
</tr>
<tr>
<td>3</td>
<td>1:3</td>
<td>4.6 (± 0.9)</td>
<td>0.66</td>
<td>NIZ</td>
</tr>
<tr>
<td>4</td>
<td>1:4</td>
<td>7.0 (± 1.4)</td>
<td>1.00</td>
<td>NIZ</td>
</tr>
<tr>
<td>5</td>
<td>1:5</td>
<td>5.6 (± 1.2)</td>
<td>0.80</td>
<td>NIZ</td>
</tr>
<tr>
<td>6</td>
<td>1:6</td>
<td>6.0 (± 0.8)</td>
<td>1.20</td>
<td>NIZ</td>
</tr>
</tbody>
</table>

*Data are the arithmetic mean ± S.D. n = 3, **NIZ = No Inhibition Zone
Ethanolic latex extract of *F. palmata* showed antibacterial activity only against *E. coli*. Ethanolic extract of concentration 1:3 mL exhibited maximum antibacterial activity with 7.6 mm zone of inhibition, where as concentrations 1:3 mL, 1:2 mL, 1:1 mL and 1:5 mL showed inhibition zones of 7.6, 7.3, 7.0, 7.0 mm and 5.6 mm as compared to standard tetracycline (7 mm zone of inhibition). The extract concentration 1:4 mL found to be least effective against *E. coli* with 4.6 mm zone of inhibition. The value of activity index of ethanolic extract against *E. coli* varies from 1.08 (maximum) to 0.66 (minimum) (Table 1).

The magnitude of antibacterial activity of pure latex and its methanolic and ethanolic extracts found to be more against *E. coli* (Gram-negative) then the *S. aureus* (Gram-positive) where as pure latex and its ethanolic extract not show any antibacterial activity against *S. aureus*. As activity index clearly showed that in the latex extracts of this plant, antibacterial activity found to be more than tetracycline a well known synthetic antibiotics indicating that the latex of this plant have significant antibacterial activity.

**Acknowledgements**

Authors are thankful to Department of Botany, Abhilashi Institute of Life Sciences, Mandi for providing necessary facilities to carry out the research work.

**References**