Detection of Antimicrobial Activity of *Oscimum sanctum* (Tulsi) & *Trigonella foenum graecum* (Methi) against some selected bacterial & fungal strains

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ABSTRACT

Plants are very useful, self-generating machines, producing a variety of useful bioactive products. Keeping in view this idea, the crude aqueous extract of two medicinal plants were subjected to *in vitro* antimicrobial assay against human pathogenic *Escherichia coli*, *Pseudomonas putida* (bacteria) and *Malassezia furfur* (fungus) by employing disc diffusion method. The zones of inhibition, minimum inhibitory concentration (MIC) were also determined. Among the plants tested *Oscimum sanctum* (Tulsi) was found to be the most effective against fungal strain in comparison to bacterial strain. *Trigonella foenum graecum* (Methi) extract showed maximum effect on *E. coli*. *Trigonella* and *Oscimum* both were found ineffective against *Pseudomonas putida*. Largest zone of inhibition (12 mm) is obtained with *M. furfur* at 100% conc. when treated with tulsi. In case of methi, largest zone of inhibition (14mm) is obtained with *E. coli* at 50% conc. Minimum Inhibitory Concentration (MIC) value of 5 mg/ml was obtained with *Oscimum sanctum*.

Keywords: Antimicrobial, disc diffusion, zone of inhibition, MIC.

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INTRODUCTION

The search for agents to cure infectious diseases began long before people were aware of the existence of microbes (1). Plants are the richest resource of drugs of traditional systems of medicine, modern medicines, nutraceuticals, food supplements, folk medicines, pharmaceutical intermediates and chemical entities for synthetic drugs (2). About 60% of the world’s populations exclusively rely on traditional medicine for their primary health-care needs (3). The major part of traditional therapy involves the use of plant extract and their active constituents (4).

Tulsi belongs to plant family Lamiaceae. The genus *Ocimum*, a member of the Lamiaceae family contains 200 sps of herbs and shrubs (5), a source of aroma compounds and essential oils containing biologically active constituents that possess insecticidal and nematicidal properties (6,7).

Fenugreek (*Trigonella Foenum-Graecum*) found in nature and is cultivated in India and Pakistan is a well known medicinal plant having properties of reducing blood sugar level (8), anthelmentic, antibacterial (9), anti-inflammatory, antipyretic (10).

This study was designed to explore the antimicrobial efficacy of *Ocimum sanctum* & *Trigonella foenum graecum* on selected human pathogens. The leaves of oscimum and seeds of trigonella were used for the present study.

MATERIAL AND METHODS

Preperation of Extracts

The aqueous extract was prepared by adding 10 g of crushed leaves of tulsi and 10 g of milled methi seeds in 50 ml of distilled water separately. It was allowed to stand for 24 h after which it was filtered using a Whatman No. 1 filter paper. The filtrate was directly used as crude extract with 100% concentration. Further dilutions were made by adding appropriate amount of distilled water.

Microbial Culture

The standard bacterial and fungal cultures were procured from IMTECH, Chandigarh, India and used in the present study. The bacteria rejuvenated in Mueller-Hinton broth (Hi-media laboratories, Mumbai, India) at 37°C for 18 h and then stocked at 4°C in Mueller-Hinton Agar. Subcultures were prepared from the stock for bioassay. The inoculum size of the bacterial culture was standardized according to the National committee for Clinical Laboratory Standards (11) guideline. The bacterial culture was inoculated into sterile nutrient broth and incubated at 37°C. Sabouraud’s dextrose agar medium is used for the culturing of *M. Furfur* with addition of butter in the medium.
Preperation of disc for Antibacterial activities

The aqueous extracts were prepared and the sterile blotting paper disc (5 mm) were soaked in the diluted extract in different concentrations (25%, 50%, 75% and 100%) . The prepared disc were dried in controlled temperature to remove excess of solvent and used for study.

Antibacterial activity using disc diffusion method

The modified paper disc diffusion (12) was employed to determine the antimicrobial activity of aqueous extract of the herbal preparations. Inoculum was spread over the agar plate using a sterile cotton swab in order to obtain uniform microbial growth. Then the prepared antimicrobial discs were kept over the lawn and pressed slightly along with control. Streptomycin 10 µg/disc (Hi-Media) were used as positive control. The plates were incubated for 18 h at 37°C. The antimicrobial activity was evaluated and diameter of inhibition zones was measured. Experiment was carried out in triplicate and the averages diameter of zone of inhibition was recorded. The antibacterial activity was classified as highly active (>10 mm), mild active (7-10) mm) and slightly active (6-7 mm) and less than 6 mm was taken as inactive.

RESULTS AND DISCUSSION

In the present study, two common herbs namely tulasi and methi were tested for their antimicrobial properties against selected human pathogens.

Table I: Antimicrobial activity of the different extracts on the selected pathogenic bacteria and fungi

<table>
<thead>
<tr>
<th>Plant</th>
<th>Part Used</th>
<th>Strains</th>
<th>Water Extract</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>100%</td>
</tr>
<tr>
<td>Tulsi</td>
<td>Leaf</td>
<td>Escherichia coli</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Pseudomonas putida</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Malassezia furfur</td>
<td>+++</td>
</tr>
<tr>
<td>Methi</td>
<td>Seed</td>
<td>Escherichia coli</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Pseudomonas putida</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Malassezia furfur</td>
<td>+</td>
</tr>
</tbody>
</table>

Table 1 shows the antimicrobial activity of the different extracts on the selected pathogenic bacteria and fungi.

In case of oscimum, Water extract with 75% conc. inhibited maximum the growth of the E.Coli and 100% conc. inhibited maximum the growth of M. furfur. No effect was observed in case of pseudomonas putida.

In case of trigonella Water extract with 50% conc. inhibited maximum the growth of the E.Coli and M. furfur. No effect was observed in case of pseudomonas putida.
Table II Zone of inhibition produced by different concentration of extracts

<table>
<thead>
<tr>
<th>Plant</th>
<th>Part Used</th>
<th>Strains</th>
<th>Zone of Inhibition (mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>100%</td>
</tr>
<tr>
<td>Tulsi</td>
<td>Leaf</td>
<td><em>Escherichia coli</em></td>
<td>7mm</td>
</tr>
<tr>
<td></td>
<td></td>
<td><em>Pseudomonas putida</em></td>
<td>6mm</td>
</tr>
<tr>
<td></td>
<td></td>
<td><em>Malassezia furfur</em></td>
<td>12mm</td>
</tr>
<tr>
<td>Methi</td>
<td>Seed</td>
<td><em>Escherichia coli</em></td>
<td>7mm</td>
</tr>
<tr>
<td></td>
<td></td>
<td><em>Pseudomonas putida</em></td>
<td>6mm</td>
</tr>
<tr>
<td></td>
<td></td>
<td><em>Malassezia furfur</em></td>
<td>7mm</td>
</tr>
</tbody>
</table>

Largest zone of inhibition (12 mm) is obtained with *M. furfur* at 100% conc when treated with tulsi. In case of methi, largest zone of inhibition (14mm) is obtained with *E. coli* at 50% conc.

Minimum Inhibitory Concentration (MIC) value of 5 mg/ml was obtained with *Oscimum sanctum*.

CONCLUSION

Our findings suggested that herbal preparation extracts have great potential as antimicrobial agent against microbes taken. Hence, it may be recommended that these two plants could be used in the treatment of human diseases caused by the above mentioned organisms. The study also supports the use of these herbal preparations not only as the dietary supplement but also as agent to prevent or control the microbial infections.

REFERENCES