ANTIMICROBIAL ACTIVITY OF NYCTANTHES ARBOR-TRISTIS LINN ON FEW CLINICAL ISOLATES

PRADIP HIRAPURE1, MUKESH POTE2
1. State Reference laboratory, Dept. of Microbiology, Govt. Medical College, Latur (MS).
2. Department of Pathology, Govt. Medical college, Ambajogai, Dist- Beed, (MS).
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Abstract: Nyctanthes arbor-tristis Linn. belonging to family Oleaceae is a well known medicinal plant. Present work taken to study the comparative antimicrobial activity of stem bark extract, root bark extract and leaf extract of Nyctanthes arbor-tristis on few clinical isolates. The test organisms were Staphylococcus aureus, Micrococcus luteus, Bacillus subtilis, Escherichia coli, Pseudomonas aeruginosa, Candida albicans and Aspergillus fumigatus. The zone of inhibition of the various plant part extracts were determined and compared with the standard drugs ciprofloxacin and fluconazole. The chloroform extract was found to have both antibacterial and antifungal activity whereas the ethanol extracts possess only antibacterial activity. Antimicrobial activity of leaf extract more effective for bacteria followed by root bark extract and stem bark extract while root bark extract most effective on fungal pathogens as compare to leaf extract and stem bark extract.

Keywords: Antimicrobial activity, Candida albicans, Oleaceae, ciprofloxacin, fluconazole

Corresponding Author: MR. PRADIP HIRAPURE
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INTRODUCTION

The frequency of life-threatening infections caused by pathogenic microorganisms has increased worldwide and is becoming an important cause of morbidity and mortality in immune compromised patients in developing countries and many infectious microorganisms are resistant to synthetic drugs; hence an alternative therapy is very much needed. In India, from ancient times, different parts of medicinal plants have been used to cure specific ailments. Today, there is widespread interest in drugs derived from plants. This interest primarily stems from the belief that green medicine is safe and dependable, compared with costly synthetic drugs that have adverse effects. Natural antimicrobials can be derived from plants, animal tissues, or microorganisms. The shortcomings of the drugs available today, propel the discovery of new pharmacotherapeutic agents in medicinal plants. To determine the potential and promote the use of herbal medicine, it is essential to intensify the study of medicinal plants that find place in folklore.

*Nyctanthes arbortristis* Linn. commonly known as harsinghar, parijat or night jasmine is one of the well known medicinal plants used in traditional system of medicine. Different parts of *N. arbortristis* are known to possess various ailments by rural and tribal people of India. Juice of the leaves is used as digestives, antidote to reptile venoms, mild bitter tonic, laxative, diaphoretic and diuretic. Leaves are also used in the enlargement of spleen. Traditionally the powdered bark is given in rheumatic joint pain, in treatment of malaria and also used as an expectorant. The plant have been screened for antihistaminic activity, CNS activities (viz. hypnotic, tranquilizing, local anesthetics), analgesic, anti-inflamatory, antipyretic, antilulcer, amoebicidal, anthelmintic, antitrypanosomal to antidepressant, antiviral and immunomodulatory. Leaves extracts was found to have antimicrobial activity as well as steam and roots extracts also found antimicrobial activity but no proper work was carried on comparative antimicrobial activity on the root bark, stem bark part and leaf of *Nyctanthes arbortristis* Linn, therefore the present work was conceived.

MATERIAL AND METHODS:

Plant materials

Leaves, Stem and Root of *Nyctanthes arbortristis* were collected from latur, Maharashtra in the month of April 2011 and were identified by the Department of Botany, Dayanand College of science latur. The plant parts were shade dried at room temperature (30-40 °C).
Preparation of extracts

The plant material was coarsely powdered and extracted sequentially with ethanol (35-40°), chloroform (95%) using Soxhlet apparatus. The extracts were filtered and allowed to evaporate to dry. Each extract was transferred into clean and dried airtight vials until ready for use. 11

Preparation of microorganisms for experiment

All the microorganisms were isolated from patients samples collected in dept. of microbiology government medical college, latur. For use in experiments, the organisms were sub-cultured in agar media.

Evaluation of antimicrobial activity

I. Culture media

Nutrient agar (NA) (Himedia) containing bromocresol purple was used for the activation of Bacillus glucose agar (Himedia) was used for the activation of the fungi.

II. Chemicals for antimicrobial assay

Ciprofloxacin and Fluconazole (Central Drug House (P). LTD., New Delhi 110002. India) were used as positive reference standards (RA) for all bacterial and fungal strains respectively. The ethanol and chloroform (Qualigenis) was used as solvent for the tested samples.

III. Antimicrobial assay:

All the experimentation was done in aseptic area under laminar air-flow cabinet. The agar diffusion method 12 was adopted for the study. Broth cultures of the test isolates (0.1 ml) containing 1.0 X 10^5 CFU/ml of organism was introduced into a sterile petridish and 15 ml of molten nutrient agar were added. The content was thoroughly mixed and then allowed to solidify. The extracts were dissolved in solvent (ethanol and chloroform) and used in concentrations 10, 20, and 40 mg/ml. Ciprofloxacin (5μg/ml) was used as standard for antibacterial activity and Fluconazole (5μg/ml) was used for antifungal activity. Holes were bored in the plates, using a standard sterile cork borer of 6 mm diameters and equal volumes of the plant extracts (500 μl) were transferred into the wells with the aid of micropipette. The experiments were carried out in triplicate. The plates were kept for 1hr for pre-diffusion and incubated at 37°C/24hr (plates containing bacterial cultures), 25°C/3days (plates containing Candida albicans culture) and 25°C/3 days (for plates containing Aspergillus fumigatus culture). At the end of incubation, zone of inhibition was measured in all the plates.

RESULT AND DISCUSSION:

In this study the antimicrobial potential of ethanol and chloroform extract of leaf, stem bark, roots bark of N. arbortristis Linn. was assessed in term of zone of inhibition of bacterial and
fungal growth. Table 1 shows the results of antibacterial activity against the tested microorganisms. Both Ethanol as well as chloroform extracts showed varying degrees of inhibition against all the bacterial stains but only chloroform extracts possess antifungal activity. Chloroform extract shows higher zone of inhibition than ethanol extract found in table-1.

Graph-1 shows the comparison of antimicrobial activity of ethanol extract of leaf, stem bark and root bark at concentration of 40 mg/ml. Antimicrobial activity of leaf extract more effective for bacteria followed by root bark extract and stem bark extract but antifungal activity not found in ethanol extract. In other hand graph- 2 shows antimicrobial activity of chloroform extract of leaf found higher for bacteria followed by root bark and stem bark extracts while in case of antifungal activity root bark extract more active followed by leaf extract and stem bark extract. From the present study it is concluded that the the leaf extract of both ethanolic extract as well as chloroform extract of leaf is more active against bacteria while chloroform extract of root bark most active against fungi Candida albicans and Aspergillus fumigates.

Table: 1 comparative antimicrobial activity of Nyctanthes arbor-tristis linn

<table>
<thead>
<tr>
<th>Zone of Inhibition of Growth (mm)</th>
<th>Extract / Conc.(mg/ml)</th>
<th>Ethanol Extract (500μl)</th>
<th>Chloroform Extracts(500μl)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>References</td>
<td>Leaf</td>
<td>Stem bark</td>
<td>Root bark</td>
</tr>
<tr>
<td>Bacterial strains</td>
<td></td>
<td>10</td>
<td>20</td>
<td>40</td>
</tr>
<tr>
<td>S. aureus</td>
<td></td>
<td>5</td>
<td>6</td>
<td>9</td>
</tr>
<tr>
<td>B. subtilis</td>
<td></td>
<td>4</td>
<td>5</td>
<td>8</td>
</tr>
<tr>
<td>M. luteus</td>
<td></td>
<td>3</td>
<td>5</td>
<td>9</td>
</tr>
<tr>
<td>P. aeruginosa</td>
<td></td>
<td>4</td>
<td>7</td>
<td>11</td>
</tr>
<tr>
<td>E. coli</td>
<td></td>
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<td>10</td>
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<tr>
<td>Fungal strains</td>
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<td>9</td>
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<tr>
<td>C. albicans</td>
<td></td>
<td>4</td>
<td>6</td>
<td>8</td>
</tr>
<tr>
<td>A. fumigates</td>
<td></td>
<td>4</td>
<td>6</td>
<td>8</td>
</tr>
</tbody>
</table>

- R1- Fluconazole , R2- Ciprofloxacin
Graph: 1 Antimicrobial activity of ethanolic extracts (leaf, stem bark, root bark) of *N. arbortristis* Linn. at a concentration 40mg/ml

Graph: 2 Antimicrobial activity of chloroform extracts of *N. arbortristis* Linn. at a concentration 40mg/ml

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