Antibacterial Activity of Fresh leaves of “Piper betle Linn”

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ABSTRACT

The Leaf of Piper betle Linn. is also called as Paan, belonging to family Piperaceae is used as masticatory in any time of maturity with areca nut and lime. Antimicrobial activity of the successive extract of the fresh leaves of Piper betle Linn. was evaluated against both Gram positive and Gram-negative bacterial strains by disc diffusion method. The results revealed that all extracts showed effective inhibitory action against S. aureas. The aqueous, ethyl acetate and pet. Ether extracts showed very effective as compared to standard penicillin. Aqueous extract was also found significantly effective against Bacillus and P.aureginosa as compared to standard penicillin.

Keywords: Antibacterial activity, Zone of inhibition, Piper betle Linn.

1. INTRODUCTION

Leaf of Piper betle is also called as Paan, belonging to family Piperaceae. Piper betle contains Diosgenin1, Essential oil i.e. eugenol, methyl eugenol, p-cymene, α-terpine2. Triterpenes & β-sitosterol are isolated & shows antiplatelet & anti-inflammatory effects3. It has been seen that betle quid have Alkaloids & Nitrosoguvacoline as mutagenicities4. Propenylphenols i.e. chavicol, chavibetol, allylpyrocatechol is isolated from chloroform extract of leaves of Piper betle, exhibited significant antifungal activity5. Piperbetol, methylpiperbetol, piperal A, and piperal B, isolated from Piper betle and are effective PAF receptor antagonists6. Reversible antifertility effect is shown by Piper betle stalk in Swiss albino male mice7. Aqueous & ethanolic extracts of Piper betle leaves have antidiabetic activity8. Radio protective property in ethanolic extract of leaf is reported9. It has antioxidative & antiplatelet effects in aqueous inflorescence of Piper betle leaf extract10. Inhibitory action of Piper betle is shown on the initiation of 7, 12-dimethylbenz[a]anthracene-induced mammary carcinogenesis in rats11.

In the present investigation, the successive extracts were subjected for antimicrobial activity against the strains of Staphylococcus aureus, P. aeruginosa, E.coli, and Bacillus.

2. MATERIALS AND METHODS:
The authenticated Leaf drugs of Piper betle were obtained from by CCRAS, Bangalore and used for further proposed studies.

2.1 Extraction:

Fresh leaves were collected and chopped into small pieces then it was subjected to successive extraction with petether, benzene, chloroform, ethyl acetate, ethanol, and aqueous. All the successive
extract were collected, filtered and concentrated in vacuum under reduced pressure and dried and stored in desiccator.

2.2 Test Organisms

The pure cultures of bacteria maintained in the microbiology Laboratory were used for the microbiological work. The test organisms were maintained on Nutrient agar medium. The test organism were used for work are, Staphylococcus aureus, Escherichia coli, P.aurenginosa , Bacillus.

2.3 Preparation of inoculums

Stock cultures were maintained at 4°C on slopes of nutrient agar. Active cultures for experiments were prepared by transferring a loopful of microorganism from the stock cultures to test tubes of Nutrient broth, and incubated for 24 hrs at 37°C. The cultures were diluted with fresh Nutrient broth.

2.4 Preparation of Media

The medium was prepared by dissolving the different ingredients in water and autoclaved at 121°C for 15 minutes. This was used for preliminary antibacterial studies.

2.5 Antibacterial susceptibility test

The disc diffusion method was used to screen the antibacterial activity. In vitro antibacterial activity was screened by using Nutrient agar (NA) obtained from Himedia (Mumbai). The NA plates were prepared by pouring 15 ml of molten media into sterile petriplates. The plates were allowed to solidify and 0.1 % inoculum suspension was swabbed uniformly and the inoculum was allowed to dry for 5 minutes. The different extracts were loaded on 3mm sterile disc till saturation. The loaded disc was placed on the surface of medium and the compound was allowed to diffuse for 5 minutes and the plates were kept for incubation at 37°C for 24 hrs. At the end of incubation, zone of inhibition formed around the disc were measured with transparent ruler in millimeter. These studies were performed in triplicate by using standard drugs (10 mcg/disc Penicillin).

3. RESULTS AND DISCUSSIONS

The zone of inhibition of different extract and standard are recorded in table 1. All the extracts were effective against S.aureus. The significant antimicrobial effect was observed with respect to aqueous, ethyl acetate and petroleum ether extracts. The aqueous extract was also found to have very significant antimicrobial effect against bacillus; where as the other extracts were found to have resistant

<table>
<thead>
<tr>
<th>Organisms</th>
<th>Zone of inhibition of extracts in mm</th>
<th>STD Penicillin (10mcg/disc)</th>
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<tbody>
<tr>
<td></td>
<td>P.E</td>
<td>B.E</td>
</tr>
<tr>
<td><strong>1. E.coli</strong></td>
<td>R</td>
<td>R</td>
</tr>
<tr>
<td><strong>2.P.aureginosa</strong></td>
<td>R</td>
<td>R</td>
</tr>
<tr>
<td><strong>3. S.aureus</strong></td>
<td>R</td>
<td>R</td>
</tr>
<tr>
<td><strong>4.Bacillus</strong></td>
<td>R</td>
<td>R</td>
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</tbody>
</table>
Graph: 1

Zone of inhibition of different extracts of Piper betle

![Graph showing zone of inhibition in different extracts](image)

R=Resistant, P.E= Petroleum ether extract, B.E= Benzene extract, C.E= Chloroform extract, E.Ac= Ethyl acetate, A.E= Alcoholic extract, Aq.E=Aqueous Extract, STD=Standard (Penicillin)

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REFERENCES

8. Arambewela LS, Arawwawala LD, Ratnasooriya WD. Antidiabetic activity of

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