Research Article

ANTIMICROBIAL ACTIVITY AND PHYTOCHEMICAL STUDY OF PHYLLANTHUS EMBLICA LINN

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ABSTRACT

In India, fruits of Phyllanthus emblica L are the most common ingredients of almost all Ayurvedic preparations like Lehya, Choorna etc. Phyllanthus emblica L fruits have been used for various disorders. This has led to the investigation of antimicrobial activity of Phyllanthus emblica L. In this study, both gram positive and gram negative organisms were used. The results show that Phyllanthus emblica L Methanolic extract exhibited a significant antimicrobial activity. The Minimum Inhibitory Concentration (MIC) exhibited by Phyllanthus emblica L methanolic extract against the tested organisms ranges between 0.261 to 0.342. The methanolic extract show more antimicrobial activity than the chloroform and Diethyl ether extracts.

KEY WORDS: Phyllanthus emblica, antimicrobial activity, Ayurvedic preparations.

INTRODUCTION

Molecular biology and genetic engineering and the computational chemistry have created the considerable potential within the pharmaceutical industry without the need to explore the nature’s chemical diversity. In the synthetic drug development of compound either a search is made through the inventory of substance earlier synthesized to find relatives to the therapeutical molecule or therapeutical molecules and their analogues are synthesized. In spite of this organic chemists have realized that plant species contain a bewildering diversity of the secondary metabolites. In the indigenous cultures higher plants have formed the basis for the treatment of the diseases since the earliest time. Accuracy in recording or observing the medical use of a plant determining whether the ethanomedicinal use can be demonstrated under the specific condition in the laboratory. Chemical characterization of the compounds and the role of the placebo effect are important issues that need to be verified in the development of drugs of plant origin.

During the last two decades there has been an up search in search for new plant derived drugs containing the medically useful alkaloids, glycosides, polyphenolics, steroids and terpenoids derivatives. Researchers identified 119 secondary metabolites isolated from the plant that are being used globally as drugs. It has been estimated that 80% of the world’s population still use the traditional medicines for their primary health care needs.

Many important modern plant drugs such as the winblastin and wincristiene have been discovered by following from traditional medicines.

The plant genus Phyllanthus L (Euphorbiaceae) is widely distributed in most of tropical and subtropical countries. It is very large genus consisting of approximately 550 to 750 species and is subdivided in to ten or eleven subgenera.
Phyllanthus emblica L is a tree of small or moderate size with a greenish-grey bark and greenish-yellow flowers formed in auxiliary clusters. The feathery leaves are linear oblong with a rounded base and obtuse or acute apex. The tended fruits are green fleshy, lobos and shining and changes to light yellow or brick red when mature. It grows in topical and subtropical parts of the China, India, Indonesia and on the Malay Peninsula. The origin of the name is from the Sanskrit (Malacca, molaka). In Hindi the tree is known as “Amla”, and “Bettada Nelli” in Kannada. Phyllanthus emblica L has been used for the anti-inflammatory and anti-pyretic treatments by the rural population. Phyllanthus emblica has been used for the treatment of several disorders such as the Scurvy, Cancer and Heart diseases. The important constituent of plant leaves have the anti-neutrophilic activity and anti-platelet properties in vitro. The extracts also posses several pharmacological properties like anti-viral (HIV, AIDS, HERPES VIRUS, CMV) anti-mutagenic, anti-allergic, anti-bacterial activities. (Khopde et al, 2000). Phyllanthus emblica L contains different class of secondary metabolites (Calixto et al, 1998).

MATERIALS AND METHODS

Plant materials:
The fruits of Phyllanthus emblica L were collected from Laxmi farms, sagara, Karnataka, India in the month November and were identified by the botany department, Kuvempu University, Karnataka, India. The fruits were peeled off and cut in to small pieces and are then stored at -20°C.

Preparation of Extracts:
Small pieces of Phyllanthus emblica L fruit material were taken and dried under shade, then grinded by mechanical grinder. Exactly 100g of coarse powdered material was successfully extracted in soxhlet extractor with chloroform, diethyl ether, and methanol. The extracts were dried in vacuum using a rotatory evaporator. The dried extracts were stored in desicator for further use.

Preparation of Micro organism for experiment:
The following micro organisms were used Klebsiella pneumoniae (LIO, locally isolated organism, obtained from the department of microbiology Kuvempu University), Staphylococcus aureus (LIO, locally isolated organism, obtained from the department of microbiology, Kuvempu University).

Phytochemical analysis of the extract:
A small portion of dried extract was used for the phytochemical analysis (Trease etal 1983, Harbourne 1973). Mayer’s reagents, Hager’s reagent, Wagner’s reagent were used to test alkaloids, Ferric chloride for Tannins, Trill-Hill reagent for Eridoids, Benedict’s solution was used to test for Saponins.

Sensitivity testing
The sensitivity testing of the extracts were determined using agar well diffusion method (Russell and Furr, 1977). The MIC of the extract was also determined using a two-fold dilution method (Russell and Furr, 1977). The bacterial were first grown in nutrient agar for 18 hour before use. The inoculum suspensions were standardized and then tested against the effect of the plant extracts at the concentration of 5 mg/ml, 10 mg/ml, 20 mg/ml. the plates later incubated at 37°C±
0.5°C for 24 hours after which they were observed for zones of inhibition (Table 1). The effects were compared with that of the standard anti biotic Streptomycin at a concentration of 1mg/ml (Khan and Omotoso, 2003).

RESULTS AND DISCUSSION:
In this study the result of the investigation showed that the chloroform, methanol, diethyl ether extracts from the fruits of Phyllanthus emblica L possess anti microbial activity against Klebsiella pneumoniae and Staphylococcus aureus. The methanol extract exhibited significant anti microbial activity than chloroform and diethyl extracts (Table 1). The MIC of Phyllanthus emblica L extracts against the tested organisms varied between 0.261 mg/ml to 0.631 mg/ml. The standard streptomycin had MIC value varying between 0.0313 mg/ml to 0.612 mg/ml. The results indicated that standard antibiotic streptomycin has stronger activity than the plant extracts (Table 2). The phytochemical analysis of Phyllanthus emblica L reveals the presence of alkaloids, tannins and saponins. These compounds are known to be biologically active. Tannins have been found to form irreversible complexes with proline-rich proteins (Hagerman and Butler, 1981) resulting in the inhibition of the cell protein synthesis.

Table 1.Antimicrobial activities of Phyllanthus emblica L fruit extracts.

<table>
<thead>
<tr>
<th>Extract</th>
<th>Concentration (mg/ml)</th>
<th>Staphylococcus aureus</th>
<th>Klebsiella pneumoniae</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Zones of inhibition(mm)*</td>
<td></td>
</tr>
<tr>
<td>Streptomycin</td>
<td>1</td>
<td>15</td>
<td>14</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>15</td>
<td>12</td>
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<td></td>
<td>10</td>
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<td>19</td>
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<td>20</td>
<td>28</td>
<td>26</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>8</td>
<td>5</td>
</tr>
<tr>
<td>Methanol</td>
<td>10</td>
<td>12</td>
<td>11</td>
</tr>
<tr>
<td></td>
<td>20</td>
<td>18</td>
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<td></td>
<td>5</td>
<td>6</td>
<td>4</td>
</tr>
<tr>
<td>Chloroform</td>
<td>10</td>
<td>10</td>
<td>9</td>
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<td></td>
<td>20</td>
<td>14</td>
<td>13</td>
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</tbody>
</table>

Table 2: The MIC of the Phyllanthus emblica L fruit extracts and Streptomycin against the bacterial isolates.

<table>
<thead>
<tr>
<th>Microorganism</th>
<th>MIC(mg/ml)</th>
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<tbody>
<tr>
<td></td>
<td>Phyllanthus emblica</td>
</tr>
<tr>
<td></td>
<td>Methanol</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>0.261</td>
</tr>
<tr>
<td>Klebsiella pneumoniae</td>
<td>0.342</td>
</tr>
</tbody>
</table>

Table 1.*= Mean of three replicates
Apart from antimicrobial activity exhibited by tannins, they also react with proteins to provide the typical tanning effect. Medicinally, this is important for the treatment of inflamed or ulcerated tissues (Mota et al, 1985). Tannins have important roles such as stable and potent antioxidants (Trease et al, 1983). Herbs that have tannins as their main component are astringent in nature and are used for treating intestinal disorders such as diarrhoea and dysentery (Dharmananda, 2003). The one of the largest group of chemical produced by plants are the alkaloids and their amazing effect on humans has let to the development of powerful pain killer medications (Raffauf, 1996).

*Phyllanthus emblica* L has been used for the anti inflammatory and anti pyretic treatments by the rural population in its growth areas in India. It is one of the common ingredient of many ayurvedic medicines. It is consumed as vegetable in pickles and other dishes in India. It is hoped that this study would lead to the establishment of some compounds that could be used to formulate new and more potent antimicrobial drugs of natural origin. Studies are in progress to further evaluate the mechanism of action of *Phyllanthus emblica* L extracts and some organism associated with human diseases.

**ACKNOWLEDGEMENT:**
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**REFERENCES:**