ANTIBACTERIAL, ANTIFUNGAL AND PESTICIDAL ACTIVITY OF PLANT MORUS ALBA - A NOVEL APPROACH IN POST HARVEST TECHNOLOGY

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ABSTRACT
A study was conducted to determine the antibacterial, antifungal & pesticidal activity of Plant Morus Alba seed oil extract on ethanol against bacteria, fungi & pest. Four bacterial species were used, of which - Escherichia coli, Pseudomonas aeruginosa, Bacillus subtilis & Staphylococcus aureus were most susceptible bacterial species to crude extract with MIC at concentration ranging from 0.2-26 mg/l. Among the tested fungal species Aspergillus Niger & Saccharomyces cerevisiae were most susceptible to oil extract with MIC at concentration ranging from 0.2-30 mg/l, respectively. The crude & seed oil extract show significant pesticidal activities against Sitophilus granaries, respectively. The above plants seed oil were shown by in vitro assays to be a potential source for natural antifungal, antibacterial & pesticidal agents.

KEYWORDS: Morus Alba, Seed Oil Extract, Ethanol, Fungi, Pest

INTRODUCTION
India is one of the largest producers of raw food materials in the world. Fruits and vegetables are living organisms and highly perishable commodities. These are affected by number of factors, main factor is post-harvest spoilage hence post-harvest losses are main source of human food loss. Post-harvest technology should comprise all the in between techniques, right from the field till the crops reach the hand of consumers either as a fresh commodity. All the methods or techniques which reduce the post-harvest losses and help economic utilization of crops to maximum as a fresh produce or to make nutritious safe and stable product (Gupta & Pathak, 1986).

Improving agricultural production is essential to achieve a sustainable development process that will contribute to reducing poverty and enhancing food security and income growth. Various researches at different level have contributed to make this development possible. High yielding varieties and new production technology have vastly increased the world’s agricultural potential and provide rural income sources and affordable food for large parts of the population. But the production of food and agricultural products does not end when the crop is harvested. Increasingly, agricultural products are not consumed in their raw form, and post harvest activities such as transport, storage, processing and marketing account for a growing part of their final value (Janardhana 1999; Chandra and sarbhoy 1997; Devi 2001). A recent estimate by the Ministry of Food and Civil supplies, Governments of India, puts the total preventable post harvest losses of food grains at 10 % of the total production or about 20 MT, which is equivalent to the total food grains produced in Australia annually.

Our study focused at minimizing the post harvest losses due to biodeterioration in grain storage system. The various traditional methods employed for grain storage inevitably provide favorable environment for growth of pests and microorganisms responsible for spoilage. Botanical antimicrobials and insecticides offer a more natural, environmentally friendly approach to control of microbes and pests growth than do synthetic ones( Satish 1999; Okigbo and Ogbonnaya 2006; Sheriff 2006; Bouamama 2006;Ergene 2006;kiran and raveessha 2006; Mohana and Raveesha 2006).
Morus is a genus of flowering plants in the family Moraceae. The whole plant is full of flavonoids like rutin, quercetin, isoquercitrin and Quercetin-3-Triglucoside, steroid, volatile oils, vitamins, trace elements, saccharides and amino acids. The plant is used for the treatment of ailments including diabetes (Naowaboot, 2009), constipation, treat grey hair, tonify the blood, bark is used to treat cough, wheezing, edema and to promote urination. It is also used to treat fever, headache, red dry and sore eyes, gout (Wang, 2010), Alba roots acts as antistress agent (Nada, 2009).

Keeping the tremendous value of plant a detailed study of plant Morus Alba was conducted which can be use to reduce post harvest losses. The aim of this study was to test various extracts from leaf and seed of Morus Alba plant against a diverse range of organisms comprising Gram positive, Gram negative and yeast to evaluate its antibacterial & antifungal activities.

MATERIALS AND METHODS

Fresh disease free leaves of Morus Alba plant were collected from New Delhi, UP, India.

PREPARATION OF EXTRACTS

Aqueous Extract

Leaf samples (100 g) of plant were thoroughly washed, blot dried and macerated with 100 ml sterile distilled water in a blender for 10 min. The macerate was first filtered through double layered muslin cloth and then centrifuged at 4000 g for 30 min. The supernatant was filtered through Whatmann. No.1 filter paper and sterilized at 120 ºC for 30 min., which serves as mother extract.

Solvent Extract

Thoroughly washed mature leaves of Morus plant were shade dried and then powdered with the help of a blender. Thirty grams of the powder was filled in the thimble and extracted successively with ethanol using Soxhlet extractor for 48 h at temperature not exceeding the boiling point of the solvent (LIN, 1999). The extract was concentrated using rotary flash evaporator and preserved at 5 ºC in airtight bottle until further use. The extract was subjected to antibacterial, antifungal & pesticidal activity.

Test Organism

Two strains of Gram negative bacteria- Escherichia coli, Pseudomonas aeruginosa, two strains of Gram positive bacteria Bacillus subtilis and Staphylococcus aureus, Aspergillus Niger and Saccharomyces cerevisiae were used. The culture of bacteria were maintained in their appropriate agar slants at 4 ºC throughout the study and used as stock cultures.

Growth Media & Conditions

Nutrient agar media pH-7.2, nutrient broth media pH-6.8 & Potato dextrose media pH- were used for antibacterial screening. MIC determination & antifungal screening determination respectively.

Minimum Inhibitory Concentration

A definition of the minimum inhibitory concentration is lowest concentration which resulted in maintenance or reduction of inoculums viability. Serial tube dilution technique was used to determine of MIC of extracts against Gram positive & negative & fungi. The plant extract (500mg) was dissolved in 2ml of distilled water to obtain stock solution. After preparation of suspension of test organism (10^5 organisms per ml), 1 drop of suspension (0.02ml) was added to each broth dilution. After 18 h incubation at 37 ºC, the tubes were then examined for the growth. The MIC of the extract was
taken as the lowest concentration that showed no growth. Growth was observed in those tubes where the concentration of the extract was below the inhibitory level & the broth medium was observed turbid (cloudy).

RESULT & DISCUSSIONS

Table 1: Minimum Inhibitory Concentration of Ethanol Extract

<table>
<thead>
<tr>
<th>S.No.</th>
<th>Test Organism</th>
<th>MIC Value of Leaf Extract</th>
<th>MIC Value of Seed Extract</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Gram +ve</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Bacillus subtilis</td>
<td>8</td>
<td>4</td>
</tr>
<tr>
<td></td>
<td>Staphylococcus aureus</td>
<td>7</td>
<td>3</td>
</tr>
<tr>
<td>2</td>
<td>Gram -ve</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Escherichia coli</td>
<td>8</td>
<td>4</td>
</tr>
<tr>
<td></td>
<td>Pseudomonas aeruginosa</td>
<td>6</td>
<td>3</td>
</tr>
<tr>
<td>3</td>
<td>Fungi</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Saccharomyces cerevisiae</td>
<td>8</td>
<td>3</td>
</tr>
</tbody>
</table>

Table 2: Diameter of Zone of Inhibition (mm)

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Test Organism</th>
<th>Ethanol Extract 150µg/disc (M± SE)</th>
<th>Ethanol Extract 300µg/disc (M± SE)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Gram +ve</td>
<td>14.2 ± 0.3</td>
<td>24 ± 0.3</td>
</tr>
<tr>
<td></td>
<td>Bacillus subtilis</td>
<td>14.4 ± 0.7</td>
<td>23 ± 0.4</td>
</tr>
<tr>
<td></td>
<td>Staphylococcus aureus</td>
<td>15 ± 0.0</td>
<td>24 ± 0.3</td>
</tr>
<tr>
<td>2</td>
<td>Gram -ve</td>
<td>14 ± 0.0</td>
<td>24 ± 0.3</td>
</tr>
<tr>
<td></td>
<td>Escherichia coli</td>
<td>14 ± 0.0</td>
<td>24 ± 0.3</td>
</tr>
<tr>
<td></td>
<td>Pseudomonas aeruginosa</td>
<td>15 ± 0.0</td>
<td>23 ± 0.0</td>
</tr>
<tr>
<td>3</td>
<td>Fungi</td>
<td>15 ± 0.0</td>
<td>23 ± 0.0</td>
</tr>
</tbody>
</table>

Note: The control disc used for solvent had no zone of inhibition so there data was omitted from the above data. Data are represented in the form of mean of the three test ± SE of the standard graph.

Pre and post harvest bio-deterioration and spoilage of grains, vegetables, fruits and agricultural produce due to infestation by insects and microorganisms may cause losses of up to 100%. Association of variety of fungi including species of Aspergillus causing significant loss in seed quality and nutritional quality of grains have been reported (Koirala, 2005). Excessive usage of pesticides in agriculture to overcome the pre-harvest and post-harvest problem was resulted in many toxic epidemics. Generally, toxic synthetic fungicides are not exploited to prevent biodeterioration of grains during storage (Harris, 2001) even through they are exploited for improving seed quality. Thus there is urgent need to develop for alternative method for prevention of biodeterioration of grains during storage without any toxicity to the consumer. Many higher plants produce economically important organic compounds, pharmaceuticals and pesticides. The most species of higher plants have never been described surveyed. Their chemical or biological active constituent which is potential to
be used as new sources of commercially valuable pesticides remain to be discovered (Balandrin, 1985). This is mainly due to the lack of information on the screening/evaluation of diverse plants for their antifungal potential. Biologically active plant derived pesticides are expected to play an increasingly significant role in crop protection strategies.

Considering these as a first step, in the present investigation Morus Alba plants were screened in vitro for antifungal activity. The finding of the present investigation is an important step towards crop protection strategies for antifungal, antibacterial activity. It was concluded that the ethanol extract of Morus Alba seed & leaf demonstrated a strong activity against gram positive, gram negative & fungi having strong pesticidal activity.

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REFERENCES


