Antibacterial activity of seed and bark extracts of Holarrhena antidysenterica

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ABSTRACT
In the present study of seed and bark of Holarrhena antidysenterica was screened for antibacterial activity. Air dried Seed and bark powder of plant was subjected to hot continuous extraction with various solvents like aqueous extracts and acetone extracts. Aqueous extract was prepared by cold maceration. Result: The antimicrobial activity of seed and bark of Holarrhena antidysenterica when tested have exhibited different degree of antimicrobial activity against tested organism. So it is found to be effective against all tested microorganisms with inhibition zone ranging from 18-25mm.

INTRODUCTION
Holarrhena antidysenterica a small shrub or deciduous tree, up to 13m in highest, with a milky latex. The bark peels off in flakes and it is grey to pale brown color. It is found all over India and other Asian countries up to an altitude of 1300m, especially in the sub Himalayan tract. In deciduous forests and open waste lands. Common names of this plant are Cavessi bark, common holarrhena, coyness bark, Easter tree, Ivory tree, Tellicherry bark, White angel. Kodaga pala, Indrayava, vennali and veppali. Around 30 alkaloids have been isolated from the plenteously from the bark[4]. The bark is used as an astrin gent, anthelmintic, antidental, stomachic, febrifuge, antdiropsical, diuretic in piles, colic, dyspepsia, chest affections, and as remedy in disease of the skin and spleen. It is a well known drug for amoebic dysentery and other gastric disorders[1]. It is also indicated in diarrhea, indigestion, flatulence and colic (Selected medicinal plants, CHEMEXCIL, Mumbai 1982). The antimicrobial activity of 18 ethno medical plant extracts against nine bacterial strains Bacillus subtilis, Staphylococcus aureus, Staphylococcus epidermis, Escherichia coli, Klebsiella pneumonia, Pseudomonas sp., and Proteus sp., Salmonella sp., and Vibrio cholerae was studied already[3]. In this study bark extract of Holarrhena antidysenterica was examined against those nine organisms.

MATERIALS AND METHODS
Collection of plant material
Plant parts such as bark, and seed of Holarrhena antidysenterica were collected from (koduuvayur) Palaghat, Kerala in December 2008. Plant material
was dried under shadow. The dried materials were fine powdered and stored in polythene bags at room temperature.

Preparation of plant extracts

Aqueous extracts

To obtain the aqueous extracts, weighed quantity powder of dried plant parts of *Holarrhena antidysenterica* was added to distilled water (1:15), boiled for 15 minutes on water bath cooled to room temperature and filtered. Filtrate was dried on tray dryer at 70°C (yield 24% w/w). The dry extract powder was dissolved in distilled water to prepare solution at concentration of 100mg/ml.

Preparation of media

Mueller Hinton Agar was prepared, and sterilized by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Whole process pH was maintained between 7.2 ±0.2° at room temperature.

Tested organisms

*Staphylococcus aureus, Escherichia coli, Klebsiella species, Proteus species, Pseudomonas species, Salmonella sp* and *Vibrio cholerae*. The cultures obtained from Clinical Microbiology Laboratory, Kovai Medical Centre and Hospital (KMCH), Coimbatore.

Antibacterial activity

Antibacterial activities of the aqueous extract of barks, and seed of *Holarrhena antidysenterica* were observed using well diffusion method. A loop full of test organisms was inoculated into peptone broth and incubated for 2 to 6 hours at 35°C until it achieved the turbidity of 0.5Mc Fairland’s standards. After incubation, the test organisms were swabbed on Mueller Hinton Agar plates. Wells were made using the sterile well cutter. Different concentrations of bark extracts, seed extracts and bark extracts (100µl to 400µl) were added to each well. The plates were incubated at 37°C for 24 hours.

RESULT AND DISCUSSION

The extracts were tested on a panel of positive and negative gram bacteria. The antimicrobial activities of bark of *Holarrhena antidysenterica* are presented in TABLE 1.

And the antimicrobial activities of seed of *Holarrhena antidysenterica* are presented in TABLE 2.

TABLE 1: Inhibitory effect of *holarrhena antidysenterica* bark against enteric bacteria

<table>
<thead>
<tr>
<th>No. Of enteric organisms</th>
<th>Bark extracts (mg/ml)</th>
<th>Aqueous</th>
<th>200 µg</th>
<th>300 µg</th>
<th>400 µg</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Staphylococcus aureus</em></td>
<td>17 mm  19 mm  20 mm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Escherichia coli</em></td>
<td>19 mm  19 mm  23 mm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Klebsiella sps</em></td>
<td>18 mm  18 mm  23 mm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Proteus sps</em></td>
<td>17 mm  18 mm  22 mm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Pseudomonas sps</em></td>
<td>16 mm  18 mm  18 mm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Bacillus sps</em></td>
<td>19 mm  19 mm  21 mm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Salmonella sps</em></td>
<td>15 mm  16 mm  19 mm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Vibrio cholerae</em></td>
<td>17 mm  19 mm  19 mm</td>
<td></td>
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</tr>
</tbody>
</table>

*Staphylococcus aureus* shows effective zone of inhibition of 20mm in aqueous extracts of bark and seed.

*Escherichia coli* shows effective zone of inhibition of 23mm in aqueous extracts of bark and 25mm in aqueous extracts of seed.

*Klebsiella species* shows effective zone of inhibition of 23mm in aqueous extracts of bark and 25mm in aqueous extracts of seed.

*Proteus species* shows 22mm in aqueous extracts of bark and in aqueous extracts seed.

*Pseudomonas species* shows 18mm in aqueous extracts of bark and 23mm in aqueous extracts of seed.

*Salmonella species* shows 19mm zone of inhibition in aqueous extracts of bark and 25mm in aqueous extracts.
Short Communication

extracts of seed. Vibrio cholera also shows effective result on 19mm in bark and 27mm in seed extract.

In case of bark extract it shows effective result on E.coli and Klebsiella sps and in seed extract Salmonella species, E.coli and Klebsiella sps shows equal response and Vibrio cholera shows highly inhibited.

It is evident that the present study states that the plant extract were active against tested organisms and it also shows activity against fungal plant pathogens[3].

In addition the results supports the use of this plant in traditional medicine for the treatment of various infections.

Now a day’s antibiotic may cause some adverse effects. As in the ancient times in which traditional medicines were used for treating many diseases. This present study Shows the antibacterial activity of bark extracts of Holarrhena antidysenterica against the test organisms. Hence it can be used for the treatment of the diseases caused by them.

In future various screening test, phytochemical analysis and animal studies has to be performed for using theses extracts as a medicine for treating various diseases.

REFERENCES