IN VITRO EVALUATION OF ANTI BACTERIAL ACTIVITY OF HEART WOOD EXTRACT OF ACACIA CATECHU WILLD ON ENTERIC PATHOGENS

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ABSTRACT

The aim of the present study was to evaluate the antibacterial activity of Acacia catechu willd on selected enteric pathogens. Acacia catechu willd (AC) is a moderate thorny deciduous tree growing in tropical countries commonly known as Black cutch. It has a diverse pharmacological and phytochemical importance and is a potent medicinal plant in the traditional Indian medicinal systems. Antibacterial activity of ethanolic and aqueous extract of heart wood of Acacia catechu was screened against Salmonella typhi, [Gram negative bacilli-GNB], Shigella flexneri [GNB], E.coli [GNB], Klebsiella pneumonia [GNB], Vibrio cholerae [GNB], Pseudomonas aeruginosa [GNB] and Staphylococcus aureus, [Gram positive cocci], using agar well diffusion technique. The results of this study showed that both the extracts at different concentrations exhibited antibacterial activity against the bacterial species tested. The ethanolic extract showed higher degree of activity than aqueous extract when compared with the standards.

Keywords: Acacia catechu willd, Anti bacterial evaluation, Mac Farland’s standard, Zone of inhibition.

INTRODUCTION

Enteric or diarrhoeal infections are major public health problems in developing countries. Enteric bacteria comprised of Salmonella sp., Shigella sp., Proteus sp., Klebsiella sp., E. coli, Pseudomonas sp., Vibrio cholerae, and S. aureus, which are major etiologic agents of sporadic and epidemic diarrhoea both in children and in adults. Recently, it has been demonstrated that many human pathogenic bacteria have developed resistance against several synthetic drugs. Available reports on lesser efficacy and more side effects of synthetic drugs need to search for alternative medicine. There are several reports on antimicrobial activity of crude extracts prepared from plants that inhibit various bacteria. However, a limited numbers antimicrobial activity of in vitro studies on herbal preparation has been published. Currently many studies are being conducted to know these herbs in depth. Screening of medicinal plants for antimicrobial activities are important for finding potential new compounds for therapeutic use. There have been numerous reports of the use of traditional plants and natural products for the treatment of enteric diseases. The present study was to evaluate the antibacterial activity of heart wood extract [ethanolic & aqueous] of Acacia catechu willd on selected enteric pathogens.

Acacia catechu Willd. (AC) (Family: Fabaceae and subfamily: Mimosoideae) known as Black cutch. AC is medium sized thorny deciduous tree mainly found in India and also found in deciduous forests around the world. It grows up to 13 meters in height. It is said that the name ‘catechu’ was given to it because its bristles resemble the claws of animals of the cat family or may be because its heart wood contains gummy extract called kath or cutch. The foliage is softly textured, light green and oval-shaped. The branches are thin and spike like due to tiny thorns grown around the exterior. The sap wood of AC is large and yellowish white and heart wood is small and red in colour. The important chemical constituents reported in the heartwood are catechin, catechutannic acid, epicatechin, catechin tetramer, dicatechin, galallocatechin, kaempferol, taxifolin, isorhamnetin, (+)azelechin, L-arabinose, D-galactose, D-rhamnose and aldobriuranic acid. Catechin is biologically highly active. It is used as a haemostatic. Another important constituent is taxifolin. Taxifolin has antibacterial, antifungal, antiviral, anti-inflammatory, and antioxidant activity.

The extract of Acacia catechu extract have been reported to have various pharmacological effects like immuno modulatory, anti pyretic, hypoglycaemic, anti diarrhoeal, hepatoprotective activity and anti inflammatory, and antioxidant activity. The extract of AC is large and yellowish white and he

MATERIALS AND METHODS

Plant material

The ethanolic and aqueous extract of heartwood of Acacia catechu willd was obtained from Green Chem Herbal Extract & Formulations. Bangalore.

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Test microorganisms

Bacterial strains used were Salmonella typhi, [Gram negative bacilli–GNB], Shigella flexneri [GNB], E.coli [GNB], Klebsiella pneumonia [GNB], Vibrio cholera [GNB], Pseudomonas aeruginosa [GNB], and Staphylococcus aureus, [Gram positive cocci]. The organisms were obtained from department of Microbiology, Saveetha Dental College and maintained in nutrient agar slope at 4°C.

Methodology

The extracts were prepared in the following concentrations in sterile water. 2mg/ml, 3mg/ml and 4mg/ml, so that 50µl of extract of different concentrations delivers 100µg, 150µg and 200 µg respectively.

Assay for antibacterial activity using agar well diffusion method

The screening of antibacterial activity of plant extracts was carried out using the agar well diffusion method. The bacterial strains were inoculated into tubes of nutrient broth and incubated at 37°C overnight. Each of the cultures were then adjusted to 0.5 McFarland turbidity standard. Lawn culture of the test organisms were made on the Muller Hinton agar [MHA-HI media M1084] plates using sterile cotton swab and the plates were dried for 15 minutes. A sterile cork borer was then used to make wells (6mm diameter) for different concentrations of the extracts on each of the plates containing cultures of the different bacterial strains. 50µl of the varying concentrations (100,150, 200µg) of the extracts were introduced into the wells with the help of micropipettes. The culture plates were allowed to stand on the working bench for 30 min for pre-diffusion and were then incubated in upright position at 37°C for 24 h. After 24 hrs, antibacterial activity was determined by measurement of diameter of zones of inhibition (mm). Standard antibiotic discs of amoxicillin (30mcg/disc) and Ciprofloxacin (30mcg/disc) were used as positive control. All the tests were done in triplicate to minimize the test error.

RESULTS AND DISCUSSION

The antibacterial activity of the extracts (Ethanolic and Aqueous) at different concentrations was screened by agar well diffusion technique. The zone of inhibition was measured in mm diameter and the results are given in table 1.

Table 1: Anti bacterial activity of heartwood extract of Acacia catechu wild

<table>
<thead>
<tr>
<th>Extract</th>
<th>Conc [µg]</th>
<th>Zone of inhibition [in mm diameter]</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>B1</td>
<td>B2</td>
</tr>
<tr>
<td><strong>Ethanolic</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>100</td>
<td>9</td>
<td>15</td>
</tr>
<tr>
<td>150</td>
<td>14</td>
<td>20</td>
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<tr>
<td>200</td>
<td>19</td>
<td>25</td>
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<tr>
<td><strong>Aqueous</strong></td>
<td></td>
<td></td>
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<tr>
<td>100</td>
<td>8</td>
<td>9</td>
</tr>
<tr>
<td>150</td>
<td>11</td>
<td>12</td>
</tr>
<tr>
<td>200</td>
<td>14</td>
<td>15</td>
</tr>
<tr>
<td><strong>Ciprofloxacin</strong></td>
<td>24</td>
<td>21</td>
</tr>
<tr>
<td>30mcg/disc</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Amoxycillin</strong></td>
<td>25</td>
<td>23</td>
</tr>
<tr>
<td>30mcg/disc</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

B1-Salmonella typhi, B2- Shigella flexneri, B3-Vibrio cholerae, B4- E.coli, B5- Klebsiella pneumoniae, B6- Psuedomonas aerugenosa, B7- Staphylococcus aureus.

Both the extracts at different concentration exhibited antibacterial activity against all bacterial strains tested. Ethanolic extract exhibited comparably a high degree of activity than the aqueous extract. The ethanolic extract was more effective against Shigella flexneri, Vibrio cholerae and Staphylococcus aureus with a zone of inhibition of 25mm, 25mm and 24 mm diameter (at conc200 µg.) respectively and was least effective against Pseudomonas aeruginosa with zone of inhibition of 10mm (at conc. 200 µg.) Among the other bacterial species studied E.coli and Salmonella typhi showed a zone of inhibition of 19mm diameter (at conc. 200 µg.) and Klebsiella pneumoniae showed inhibition zone of 16mm diameter (at conc. 200 µg.).

One way to prevent antibiotic resistance of pathogenic species is by using new compounds that are not based on existing synthetic antimicrobial agents. Traditional healers claim that some medicinal plants are more efficient to treat infectious diseases than synthetic antibiotics. It is necessary to evaluate, in a scientific base, the potential use of folk medicine for the treatment of infectious diseases produced by common pathogens. Medicinal plants might represent an alternative treatment in non-severe cases of infectious diseases. They can also be a possible source for new potent antimicrobial agent to which pathogen strains are not resistant.

The present study was to evaluate the antibacterial activity of Acacia catechu willd against enteric pathogens. All the extracts showed varying degrees of antimicrobial activity on the microorganisms tested. It was evident that antimicrobial activity was more apparent in ethanol than aqueous extracts. Further work is needed to isolate the secondary metabolites from the extracts studied in order to test specific antimicrobial activity.
CONCLUSION

This in vitro study demonstrated that folk medicine can be as effective as modern medicine to combat pathogenic microorganisms. The millenarian use of these plants in folk medicine suggests that they represent an economic and safe alternative to treat infectious diseases. It is clear from the results that, the extract acts as a good source of antimicrobial agent against various bacterial pathogens tested and exhibited broad spectrum of antibacterial activity.

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REFERENCES


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