ANTIFUNGAL ACTIVITY OF ACACIA CATECHU BARK ETHANOLIC EXTRACT AGAINST DERMATOPHYTES- AN INVITRO STUDY

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Abstract

The objective of the study is to evaluate in-vitro antifungal activity of Acacia catechu against three human pathogenic fungi, Trichophyton rubrum, Microsporum gypseum and Epidermophyton floccosum. Acacia catechu is an evergreen tree which possess lots of medicinal value. It exhibits diverse pharmacological actions like hepato protective, antibacterial, antifungal, anti-inflammatory, antioxidant activity. The herbal extract were tested against various concentrations adopting agar well diffusion method. The anti-dermatophytic activity indicated that the extract was ineffective and did not show any activity.

Key words: Acacia catechu, Anti fungal, dermatophytes, Zone of Inhibition.

Introduction

Dermatophytes are a group of filamentous fungi that cause infections of the skin. Diseases caused by dermatophytes include athlete’s foot, ringworm, nail infections. Dermatophyte infections in immune compromised patients can be quite severe. Dermatophytes are fungi that require keratin for growth. The organisms colonize the keratin tissues and inflammation is caused by host response to metabolic by-products. These infections are long lasting and are difficult to treat.

The incidence of dermatophytic infections have increased in the past decades. Dermatophytes are responsible for serious human pathogenic disorders in various parts of the world. The incidence of these infections are more in tropical countries, their humid climate, population and poor hygiene make an ideal condition for the growth of these organisms. Although control measures are available, they have limited effectiveness. Conventional antifungal agents such as chlorohexidine and imidazole derivatives have limited uses. Due to their common side effects such as hepatotoxicity, nausea, diarrhoea and impotency, the use is restricted in pregnant and the young people.
Dermatophytic infections can be treated either topically or systemically; the method chosen will depend on the type of infection, the severity of the infection, and the patient’s preferences.  

*Acacia catechu* commonly known as karungali in Tamil and khadira in Sanskrit is an evergreen tree with lots of nutritional and medicinal value. People in Kerala consume karungali water for relieving digestive disorders. It exhibits diverse pharmacological effects like antibacterial, antioxidant, hepatoprotective, anti-inflammatory, antiviral, analgesic, antipyretic, anti ulcer, anti cancer activity. The phytochemical constituents like Epigallocatechin, Epicatechin, rutin, Quercetin present in it produces antibacterial and antioxidant effects.

Keeping this in view, the present study was designed to evaluate the in vitro anti-dermatophytic activity of *ficus Racemosa* against *Microsporum gypseum*, *Trichophyton rubrum* and *Epidermophyton floccosum*.

**Materials & methods**

**Plant Material**

*Acacia catechu* Willd bark (AAE/9007) were collected from Hosur, Tamilnadu and was authenticated by Dr.H.B Singh, Raw materials herbarium & museum ,NISCAIR ,New Delhi. The voucher specimen is preserved for further use in Green Chem lab, Bangalore.

**Ethanol Extraction**

Barks were shade dried for a week. Dried barks were milled to fine powder. Powder was passed through 100 mesh sieve and stored in a sealed polythene bag. 2.5kg of powdered *Acacia catechu* bark were extracted with 10 liters of Ethanol, at 65°C temperature, for 1 hour, in a 20 liter round bottom flask with Graham condenser attached. Condenser was cooled circulating with chilled water. After 1 hour of extraction, round bottom flask was cooled to room temp and the extract were filtered and collected. The Marc was extracted repeatedly with 10 liters of Ethanol, twice. The extracts were filtered and collected. The combined extracts was evaporated to dryness under reduced pressure in a Buchi Rotar Evaporator (Switzerland) at 65°C, to obtain 150g of powder extract. The w/w yield of the prepared extract was 6%. The extract was stored at 4 °C until used.

**Fungal Cultures**

Three fungal pathogen used were procured from Institute of Microbial Type Culture Collection, Chandigarh (IMTECH) viz., *Microsporum gypseum* MTCC No. 2819, *Trichophyton rubrum* MTCC No.296 and *Epidermophyton floccosum* MTCC No.613 , and are maintained in Sabouraud Dextrose Agar.

**Antifungal Activity**
Well diffusion method

On sterile plates containing Sabouraud’s Dextrose Agar, the fungal cultures were swabbed. Wells of 6 mm diameter were bored in each plate. The wells were filled with varying concentrations of the sample. The plates were incubated at 28° C for 72 h for evaluation. The diameter of inhibition zones formed around the wells were measured in millimeter. The study was performed in duplicates for all the samples.²³

Result & Discussion

*M. gypseum* has been described as causing subcutaneous mycosis in humans and has been associated with opportunistic infections occurring in patients with Human Immunodeficiency Virus (HIV)²⁴,²⁵,²⁶

*Epidermophyton floccosum* is an anthropilic dermatophyte world wide in distribution. Humans and animals act as a host for this dermatophyte and the infection spreads by contact. These dermatophytes affect the cornified layers of epidermis. Their infection is more aggressive in immune compromised individuals.²⁷-²⁹

*Trichophyton rubrum* is an anthropilic saprotroph. They are usually restricted to the upper layers of epidermis, deeper infections may also occur. They manifest as both acute and chronic infections affecting men more commonly than women, these infections are known to form folliculitis which is characterized by foreign body giant cells and fungal elements. In patients with immune deficiency extensive granuloma formation is seen.³⁰-³¹

The study shows that there is no significant antifungal activity while testing against three dermatophytes in which the *Microsporum gypseum, Epidermophyton floccosum, Trichophyton rubrum* is most commonly affecting human and animal.

Table 1: Antifungal activity of *Acacia catechu* against dermatophytes

<table>
<thead>
<tr>
<th>S.No</th>
<th>Micro Organism</th>
<th>15mg/ml</th>
<th>25mg/ml</th>
<th>50mg/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td><em>Microsporum gypseum</em></td>
<td>No activity</td>
<td>No activity</td>
<td>No activity</td>
</tr>
<tr>
<td>2</td>
<td><em>Epidermophyton floccosum</em></td>
<td>No activity</td>
<td>No activity</td>
<td>No activity</td>
</tr>
<tr>
<td>3</td>
<td><em>Trichophyton rubrum</em></td>
<td>No activity</td>
<td>No activity</td>
<td>No activity</td>
</tr>
</tbody>
</table>

Conclusion

Dermatophytoses are refractory to treatment, and the spectrum of antifungal for treating dermatophytoses is narrow. However, we suggest that *Acacia catechu bark* extract do not exhibit pharmacological effects and couldnot be employed in management of cutaneous infections.

Acknowledgement

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