Anti-dermatophyte activity of Cryptolepis buchanani Roem. & Schult

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INTRODUCTION

The superficial fungal infections in humans (dermatomycoses) are caused by dermatophytes, a group of filamentous fungi that invade and draw nutrients from the keratinized tissues such as skin, hair and nails. Among the dermatophyte genera, Trichophyton, Microsporum and Epidermophyton are most important. As the dermatophytes have developed resistance to antimycotic drugs, there is an urgent need for nontoxic, safe and cost effective antifungal agents (1, 2). Biologically active compounds present in the medicinal plants have always been of great interest to scientists working in this field. In recent years this interest to evaluate plants possessing activity for various diseases is growing (3). Plants readily synthesize substances for defense against attack by insects, herbivores and microorganisms. Cryptolepis buchanani Roem. & Schutt, locally called Kareballi, belongs to the family Asclepiadaceae. It is a large climbing or straggling shrub. Leaves 2-5 cm, opposite, elliptic oblong, green shining above with a pale whitish beneath. Flowers yellowish green in axillary cymes and flowering occurs in June. In ayurvedic practice, the root is used as a substitute for that of Hemidesmus indicus to treat gout, polyuria, wounds and leprosy. It is considered alterative refrigerant and tonic (4).

MATERIAL AND METHODS

Collection and identification of plant material

The plant material was collected from Bhadra wildlife sanctuary of Central Western Ghats of Karnataka and authenticated in Department of Studies and Research in Applied Botany, Jnana Sahyadri, Shankaraghatta. The voucher specimen (voucher no. KU/AB/ KSV/31) was deposited in the department for future reference.

Extraction and Phytochemical analysis of solvent extracts

For extraction, about 50g of the shade dried and powdered leaf materials were taken and added to 100ml of methanol separately. The mixtures were sonicated for 30 min, and then left at room temperature overnight. The extracts were filtered over Whatman No 1 filter paper, and the filtrates were concentrated under reduced pressure to pasty mass (5). The methanol extract was subjected to preliminary phytochemical screening to screen the presence of various secondary metabolites (6).
**Antifungal activity of methanol and aqueous extracts**

Fungi namely *Microsporum gypsinum* (MTCC2819), *Chrysosporium keratinophilum* (MTCC1367), *Trichophyton rubrum* (MTCC3272) and *Chrysosporium indicum* (MTCC4965) were tested for their susceptibility to the solvent extracts by Agar well diffusion method (7). The fungal cultures were obtained from IMTECH, Chandigarh, India. The fungal inocula were aseptically swabbed on sterile and solidified Sabouraud dextrose agar plates. Then, aseptically wells of 6mm diameter were bored in the inoculated plates with the help of gel puncher and the extracts (10mg/ml of 10% DMSO), Standard (Amphotericin B, 1mg/ml) and Control (10% DMSO) were added into the respectively labeled wells. The plates were incubated at 28°C for 72 hours in upright position and the zone of inhibition formed around the well was recorded. The experiment was carried in triplicates to get average reading.

**RESULTS AND DISCUSSION:**

The preliminary phytochemical analysis showed the presence of phytoconstituents namely saponins, alkaloids and tannins in both the solvent extracts. In addition to these, flavonoids were detected in aqueous extract (Table-1). Table-2 shows antifungal activity of methanol and aqueous extracts of *C. buchanani* against human dermatophytic fungi. *T. rubrum* was found to be more susceptible to methanol extract followed by *C. keratinophilum*, *C. indicum* and *M. gypsinum*. Aqueous extract caused marked inhibition of *C. keratinophilum*. The inhibitory activity of standard was greater when compared to both the extracts. Control (10% DMSO) did not reveal any inhibition of test fungi.

The use of plant extracts to treat infections is an age-old practice in a large part of the world, especially in developing countries, where there is dependence of traditional medicine for a variety of diseases (8). In developing countries like India where poverty and malnutrition is rampant, knowledge of plant derived metabolites could reduce the cost of health care. India has a rich history of using various herbs and herbal components for treating various diseases (9). Infectious diseases caused by bacteria, fungi, viruses, and parasites remain a major threat to public health, despite tremendous progress in human medicine. Their impact is particularly great in developing countries because of the relative unavailability of medicines and the emergence of widespread drug resistance (10). Interest in plants with antimicrobial properties has revived as a result of current problems associated with the use of antibiotics (11). *C. buchanani* is a woody twiner found in Eastern Ghats and is widely used as demulcent, diaphoretic, diuretic, cure for paralysis (12), rickets in children (13); combined with *Euphorbia microphylla* is given to women as galactogogue when milk supply is deficient or ills (14). The roots and leaves of the plant are reported to consist of cardiac glycosides and anticarcinogens (15). Sarverogenin and isosarverogenins of the plant (16) possess potent cytotoxic activities against tumor cells and shows antibacterial and antiparasitic properties (17–18). The plant is used in Ayurveda for antidiarrhoeal, antiulcerative, anti-inflammatory, blood purifier, cough treatment, curing rickets in children and antibacterial (19). The alcoholic extract of stem has been used for

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**Table-1: Phytoconstituents in the methanol and aqueous extracts of *C. buchanani***

<table>
<thead>
<tr>
<th>Phytoconstituent</th>
<th>Methanol extract</th>
<th>Aqueous extract</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tannins</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Terpenoids</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Alkaloids</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Steroids</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Saponins</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>–</td>
<td>+</td>
</tr>
</tbody>
</table>

‘+’ Detected; ‘−’ Not detected

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**Table 2: In vitro antifungal activity of solvent extracts of *C. buchanani***

<table>
<thead>
<tr>
<th>Test fungi</th>
<th>Zone of inhibition in cm</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Methanol extract</td>
</tr>
<tr>
<td><em>M. gypsinum</em></td>
<td>10</td>
</tr>
<tr>
<td><em>C. keratinophilum</em></td>
<td>12</td>
</tr>
<tr>
<td><em>T. rubrum</em></td>
<td>14</td>
</tr>
<tr>
<td><em>C. indicum</em></td>
<td>11</td>
</tr>
</tbody>
</table>

Results are average of three trials
treatment of inflammatory conditions such as arthritis, muscle and joint pain (20). Anti-inflammatory activity of stem of *C. buchanani* using *in vitro* systems showed no relevant activity in any of the *in vitro* systems tested (21). Immunostimulatory and immunorestorative properties of *C. buchanani* were investigated (22). The aqueous extract of *C. buchanani* leaves showed inhibitory effect against *S. aureus*, *E. coli*, *S. typhimurium*, *K. pneumoniae*, *P. vulgaris*, *B. subtilis*, *L. plantarum* and *S. epidermidis* (23).

**CONCLUSION**

The traditional medicines hold a great promise as a source of easily available effective antifungal agents to the people, particularly in developing countries, including India. Indigenous system of medicine reports a number of plants for their antifungal efficacy. However, their scientific evaluation as compared to commercial agents is limited. The extracts of the plant used in this study were found to be effective against the human dermatophytes tested. The results of this investigation are in justification of traditional use of the plant. Further studies on isolation of active constituents from solvent extracts and *in vivo* studies may possibly reveal the potential of plant to inhibit human dermatophytic fungi.

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**REFERENCES**