

EVALUATION OF ANTIDERMATOPHYTIC ACTIVITY OF *PEDALIUM MUREX* L.

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ABSTRACT

Dermatophyte infections are very common in India where hot and humid climate along with poor hygienic conditions play an important role in the growth of these fungi. *Trichophyton* and *Microsporum* species forms the commonest etiological agent of dermatophytosis. In the present investigation, different solvent extracts of *Pedaliium murex* L. were screened against two dermatophytes viz., *Trichophyton rubrum* and *Microsporum gypseum*. The prominent zone of inhibitions was observed in methanol and petroleum ether extract of *Pedaliium murex* against in *Microsporum gypseum* at 200 µg/ml concentration. The distilled water and methanol extracts of *Pedaliium murex* showed significant activity against *Trichophyton rubrum*. The standard drugs

griseofulvin, fluconazole and ketoconazole were used as the positive control and zones of inhibition were from 10 to 20 mm.

KEYWORD: Dermatophytes, *Pedaliium murex* L., antidermatophytic activity, methanol extracts.

INTRODUCTION

Dermatophyte infections are one of the earliest known fungal infections of mankind and are very common throughout the world. Dermatophytosis are the most common types of cutaneous fungal infections seen in man affecting skin, hair, and nails, in both developed and developing countries due to advent of immunosuppressive drugs and diseases. Hot and humid climate in the tropical and subtropical countries like India makes dermatophytosis infection as a very common superficial fungal infection (Singh and Beena, 2003).

Griseofulvin, the only antifungal drug which had been in use for many years for treatment of dermatophytoses is still the preferred drug, though it is reported that the fungus has already developed resistance to the said drug (Huang, 2004). In this context, the new antifungals of plant origin could be useful alternatives for the treatment of dermatophytoses. Therefore, in recent years, there has been growing interest in search of suitable medicinal plants for skin ailments.

Pedaliium murex is a small herb distributed in tropical Africa, Ceylon, India and Mexico. It is commonly found in Coastal areas of Southern India. It is commonly called Gokhru (Yaana Nerinji) a member of family Pedaliaceae. It is traditionally used for the treatment of puerperal diseases, digestive tonics, ulcers, fevers, wounds, other ailments and general debility.

Infusion of the leaves and stem in cold water is demulcent, diuretic and used for the treatment of gonorrhea. Leaves are used for ulcers. Fresh leaves and young shoots dipped and kept for few minutes in boiling milk, such milk is used as an aphrodisiac. Root is used for antibilious (Nadkarni, 1992; Chopra *et al.*, 1999; Ashok kumar and Narayana, 2005). Fruits are considered demulcent, diuretic, antispasmodic, antiseptic and aphrodisiac. Juice of fruit is believed to dissolve the kidney stone (Kirtikar and Basu, 1933; Rastogi and Mehrotra, 1991; Satyavati and Gupta, 1987; Ebadi, 2004).

It contains several flavonoids like pedalitin, diosmetin, dinatin, pedalin dinatin-7-glucuronide (Subramanian and Nair, 1972). The stem contains herman and seeds contain an alkaloid harmine and also possesses some saponins. It contains alkaloids, a greenish fatty oil, small amount of resin and ash. Fruit contains a mucilagenous alkaloid 3.5 to 5 % stable oil, aromatic oil, tannins, glycosides, sterol and some nitrates, fat, resin, and gum, Caffeic acid, cumaric acid, daucosterol, ferulic acid, hepatatriacontonic acid, vanillic acid (Mithal and Sagar, 1974), ursolic acid and sitosterol were isolated from this plant. Flavonoids, triterpenoids, steroids, lipids, fatty acids, phenolic acids, amino acids and carbohydrates of *Pedaliium murex* were also reported (Khanuja and Shukla, 2004).

Pharmacological activities i.e antiulcer activity (Banji *et al.*, 2010), aphrodisiac activity (Balamurugan *et al.*, 2010; Patel *et al.*, 2012), anti inflammatory activity (Parimala Devi *et al.*, 2010), antioxidant activity (Thakkar *et al.*, 2011; Madhu Babu *et al.*, 2011; Patel *et al.*, 2011), antimicrobial activity (Muruganantham, 2011a & b; Shelke *et al.*, 2011; Nalini *et al.*,

2011), nephroprotective activity (Sreedevi *et al.*, 2011) and antidiabetic activity (Ravikumar and Krishnamoorthy, 2011) of *P. murex* L. were also reported.

The numerous ethnic tribes in India have developed their own herbal remedies for all their diseases including the skin ailments. Survey of ethno botanical literature in India, reveals 269 plant species used by various ethnic groups for skin disorders in India (Jain, 1991). Therefore, in the present study we aim to evaluate the antidermatophytic activity of *Pedaliium murex* L.

MATERIALS AND METHODS

Collection of medicinal plant

The selected medicinal plant *Pedaliium murex* L. was collected from in and around area of Thanjavur District, Tamil Nadu, India (Figure 1).

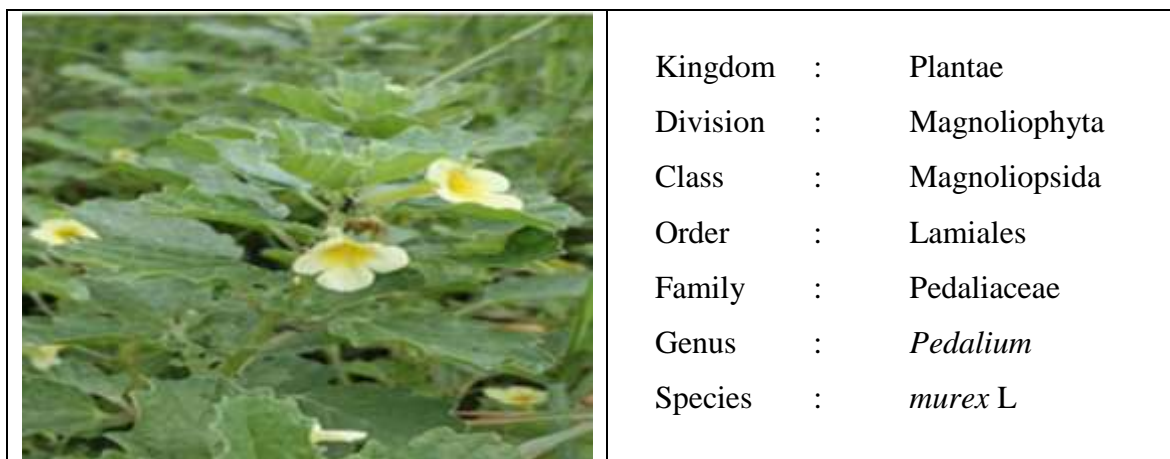


Figure 1. Scientific classification of *Pedaliium murex* L.

Identification of medicinal plant

The plant were carefully examined and identified with the help of regional flora (Gamble, 1935 and Mathew, 1983). Specimen was further confirmed with reference to Herbarium sheets available in the Rapinat Herbarium, St, Joseph College, Tiruchirappalli.

Sterilization of plant

Fresh and healthy plants were taken and sterilized with running tap water and soaked in 0.1% mercuric chloride solution for few seconds. Finally, the herb was washed with distilled water (three times) and shade dried was subjected to pulverization to get coarse powder. The coarse powdered material was stored at normal room temperature in sterilized vessels.

Preparation of extracts

The plant extracts were prepared using petroleum ether, benzene, chloroform, methanol and distilled water at different concentrations such as 100, 150 and 200 µg/ml.

Isolation of dermatophytes

The dermatophytes were isolated from the patients who outpatients of dermatology sections in Raja Mirasudhar Government Hospital, Thanjavur, Tamil Nadu using potato dextrose agar (PDA) medium (Himedia, Mumbai). The isolates were identified by using standard manuals (Rebell and Taplin, 1964; Emmons *et al.*, 1970; Rippons, 1988; Padhye and Weitzman, 1998).

Antidermatophytic activity

The antidermatophytic activity of *Pedaliium murex* was tested against isolated dermatophytic fungi *Microsporum gypseum* and *Trichophyton rubrum* by well agar method (Perez *et al.*, 1990).

In the freshly prepared and sterilized potato dextrose agar medium (Himedia, Mumbai), a pinch amount of streptomycin was added for preventing bacterial contamination and mixed well. The medium was poured into each petriplate and allowed to solidify. The test fungal cultures were evenly spread over the media by using sterile cotton swab. Then wells 5 mm was made in the medium by using sterile cork borer, 200 µl of the each concentration of petroleum ether, benzene, chloroform, methanol and aqueous plant extracts were transferred into separate wells. Then these plates were incubated at 28°C for 48-72 hours. After incubation period the results were observed and measure the diameter of inhibition zone around the each well. Fluconazole, Griseofulvin and Ketoconazole were used as positive control and petroleum ether, benzene, chloroform, methanol and aqueous were used as negative control.

RESULTS AND DISCUSSION

Dermatophytosis is one of the most commonly encountered cutaneous fungal infections worldwide. *Trichophyton* and *Microsporum* species forms the commonest etiological agent of dermatophytosis (Sen and Rasul, 2006; Venkatesan *et al.*, 2007; Ahmad *et al.*, 2010; Doddamani *et al.*, 2013). In the present study, two dermatophytes such as *Microsporum gypseum* and *Trichophyton rubrum* were isolated and identified. The macroscopic and microscopic views of isolates were presented in Plate 1.

Scientific classification of *Microsporum gypseum*

Kingdom	:	Fungi
Division	:	Ascomycota
Class	:	Euascomycetes
Order	:	Onygenales
Family	:	Arthrodermataceae
Genus	:	<i>Microsporum</i>
Species	:	<i>gypseum</i>

Macroscopic Features

Microsporum gypseum grows relatively rapidly and matures in 6 to 10 days. The texture of the colony is powdery to granular and the color is beige. From the reverse, it is yellow to brownish red.

Microscopic Features

Microsporum gypseum produces septate hyphae, macroconidia and microconidia. Macroconidia are abundant, measure 8-16 x 22-60 µm, contain 3-6 compartments and are fusoid (tapered at both ends) and symmetrical in shape with rounded ends. The walls of macroconidia are thin and rough. Microconidia are described as drop shaped, pyriform club shaped, moderate in number and located laterally along the hyphae.

Scientific classification of *Trichophyton rubrum*

Kingdom	:	Fungi
Division	:	Ascomycota
Class	:	Euascomycetes
Order	:	Onygenales
Family	:	Arthrodermataceae
Genus	:	<i>Trichophyton</i>
Species	:	<i>rubrum</i>

Macroscopic features

The growth rate of *Trichophyton rubrum* colonies is slow to moderately rapid. The texture is waxy, glabrous to cottony. From the front, the color is white to bright yellowish beige. Reverse is pale, yellowish brown.

Microscopic Features

Microscopically, the granular type is characterized by the production of moderate to abundant numbers of clavate to pyriform microconidia and moderate to abundant numbers of thin walled, cigar shaped macroconidia.

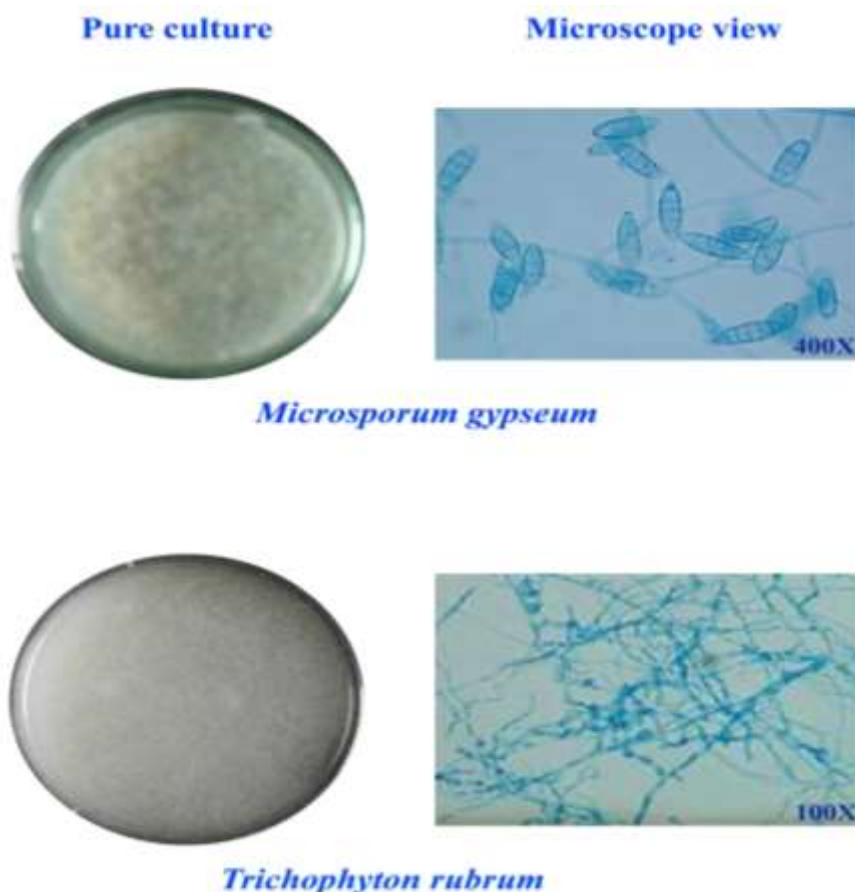


Plate 1. Isolated dermatophytic fungi

Antidermatophytic activity of *Pedaliium murex*

In the present study, the prominent zone of inhibitions was observed (22 and 20 mm) in methanol and petroleum ether extract of *Pedaliium murex* against in *Microsporium gypseum* at 200 µg/ml. The benzene and chloroform extracts did not show any inhibition against *Microsporium gypseum* (Table 1; Plate 2). The distilled water and methanol extracts of *Pedaliium murex* showed significant activity (20 mm to 24 mm) against *Trichophyton rubrum*. The benzene extract exhibited moderate activity against *Trichophyton rubrum*. The minimum zone of inhibition (5 mm) was observed in petroleum ether and benzene extract against *Trichophyton rubrum*. Similar work was done by Caceres *et al.* (1991) who reported

Cassia occidentalis exhibited antifungal activity against *T. rubrum*, *T. mentagrophytes* and *M. gypseum*.

Recently, Kavitha Sagar and Vidyasagar (2013) also studied antidermatophytic activity of some traditionally used medicinal plants of North Karnataka Region. They reported that *Cassia tora* showed good antifungal activity against the dermatophytes *T. tonsurans*, *T. mentagrophytes*, *T. rubrum*, *M. gypseum* and *Epidermophyton floccosum*.

Table 1. Antidermatophytic activity of *Pedaliium murex* L.

S. No.	Name of the organisms	Zone of inhibition at different concentration (µg/ml) (diameter in mm)														
		Petroleum ether			Benzene			Chloroform			Methanol			Distilled Water		
		100	150	200	100	150	200	100	150	200	100	150	200	100	150	200
1.	<i>Microsporum</i>	-	-	20	-	-	-	-	-	-	10	15	22	10	13	20
2.	<i>Trichophyton rubrum</i>	-	-	5	5	15	20	-	-	15	12	15	24	18	20	25

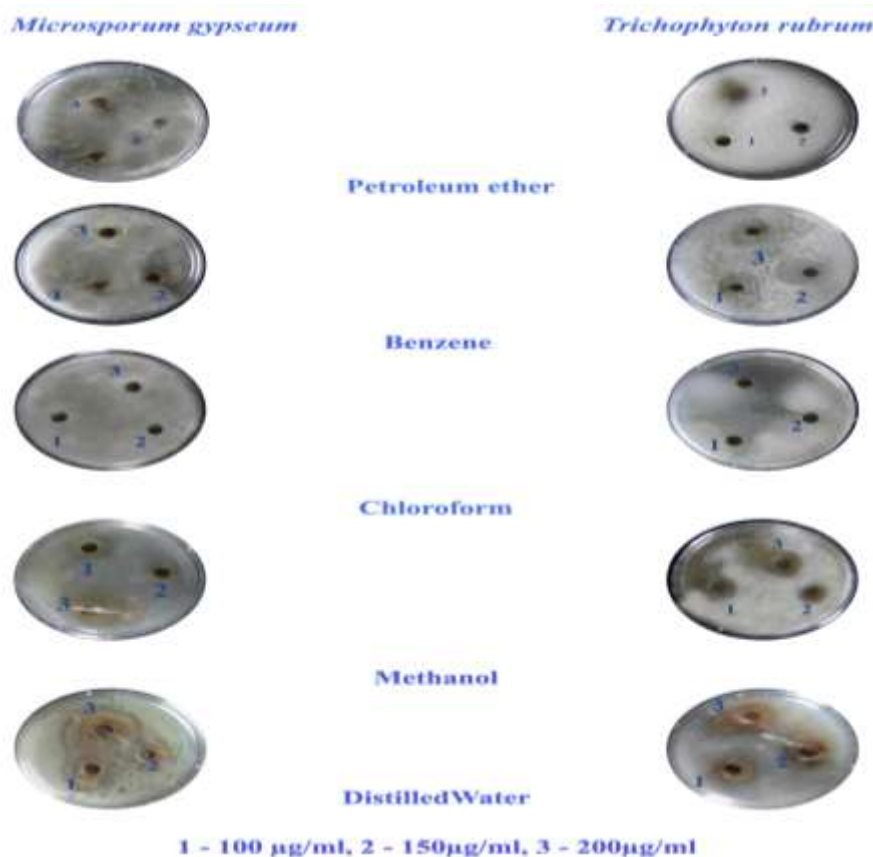


Plate 2. Antidermatophytic activity of *Pedaliium murex*

Antibiotic sensitivity test

The antibiotic sensitivity test using standard antibiotics such as fluconazole, ketoconazole and griseofulvin were tested against *Microsporum gypseum* and *Trichophyton rubrum*. The maximum activity was observed in griseofulvin (15 mm) followed by fluconazole (12 mm) and ketoconazole (10 mm) against *Microsporum gypseum*. The fluconazole antibiotic showed promising activity against *Trichophyton rubrum* (20 mm) (Figure 2; Plate 3). All the solvents (petroleum ether, benzene, chloroform, methanol and distilled water) showed no antidermatophytic activity against *Microsporum gypseum* and *Trichophyton rubrum*.

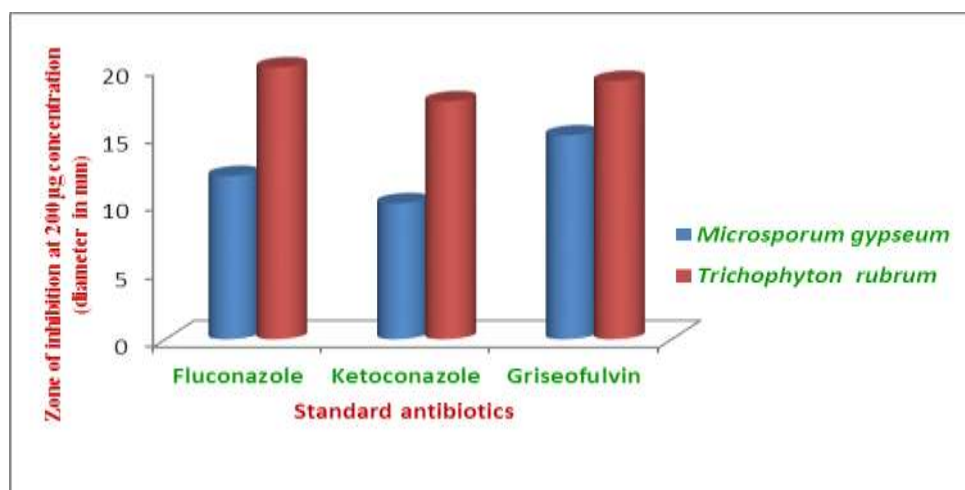


Figure 2. Antibiotic sensitivity test on *Microsporum gypseum* and *Trichophyton rubrum*



Plate 3. Antibiotic sensitivity test on *Microsporum gypseum* and *Trichophyton rubrum*

CONCLUSION

The overall investigation can be concluded that the different extracts of *Pedaliium murex* L. showed significant antidermatophytic activity. It is further recommended for isolation of

antifungal principles and their fractions for establishing cheaper, affordable and acceptable herbal products for future use.

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