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Isolation and Identification of Keratinophilic Fungi from Different Soil Samples in Jhansi City (India)

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Abstract: A total of 54 soil samples were examined from Jhansi city for the isolation and identification of keratinophilic fungi. Different sites were selected which include gardens, schools, poultry farms, rivers, hospitals and garbage dumping sites. A total of 23 species belonging to 11 genera were isolated and identified using hair bating technique. The fungi so isolated belongs to 5 species from genus *Trichophyton*, 4 species from *Chrysosporium*, 3 species each from *Microsporum* and *Aspergillus,* 2 species from *Fusarium* and one species from each of the following genera: *Rhizopus*, *Alternaria*, *Trichoderma*, *Candida*, *Penicillium* and *Paecilomyces*. The present study revealed that among the different sites selected the soil from gardens (65%) is rich in keratinophilic fungi followed by schools (52%), poultry farms (43%), garbage dumping sites (34%), hospitals (30%) and rivers (21%).

Key words: Keratinophilic fungi, prevalence, hair-baiting technique, Jhansi

INTRODUCTION

Nature has provided planet earth with a variety of beneficial organisms. Keratinophilic fungi are one of the nature's gifts which have the ability to decompose even the hardest substance like keratin. Keratineous material in or on soil are degraded by these fungi (biodegradation). In India lot of work has been done on distribution of keratinophilic fungi and related dermatophytes in the soil, which include the reports on isolation of fungi from soils of Jaipur and Mount Abu, (Garg, 1966), Mumbai, (Deshmukh, 1999), Chilka-Lake, (Ghosh and Bhatt, 2000) and Mussoorie, (Deshmukh *et al*., 2000), Damoh, (Khanam and Jain, 2002) and Mussoorie (Deshmukh and Agrawal, 1985). The keratinophilic fungi in India were isolated from various habitats viz., public parks and soils or floor dust of primary schools (Ramesh and Hilda, 1998), lake side soils (Ghosh and Bhatt, 2000), birds and their environment (Sur and Ghosh, 1980) and saltpans (Deshmukh, 2004). Places like schools, play grounds, public parks are often invaded by animals and humans there by leaving organic residues which may contaminate the soil with keratinaceous debris and mainly propagules of keratinophilic fungi. These soils become a potential source of infection for human beings (Marchisio, 1986; Matovani, 1978). Studies on the ecology and epidemiology of human dermatophytoses in the West Bank of Jordan (Al-Mussllam, 1989; Ali-Shtayeh and Arda, 1986) showed that about 36% of the patients with dermatophytoses were school children in the age group of 6-14. Studies of keratinophilic fungi are of considerable significance for their role in the breakdown of keratinous debris of man and animals in nature and they have a worldwide distribution (Al-Mussllam, 1989; Karam El-Din *et al*., 1996; Sur and Ghosh, 1980).

Geographically the climate of Jhansi region is characterized by long and intense hot summer, low and irregular rain fall with short and mild winter. The maximum temperature of 49°C has been recorded in the month of June. So far, no study has been conducted for isolation of keratinophilic fungi from Jhansi city. Hence the present study reports the occurrence of keratinophilic fungi in this region.

MATERIALS AND METHODS

During 2007, fifty four soil samples were collected randomly from different sites viz., gardens, schools, poultry farms, rivers, hospitals and garbage dumping sites. Each site was further divided in to three respective sites and as such a total of eighteen sites were selected for the isolation and identification of keratinophilic fungi. The soil samples were collected from the depth not exceeding 3-5 cm. Soil samples were brought to the

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laboratory in sterile, tightly closed polythene bags for further processing. The modified hair-baiting technique of Vanbreuseghem (Vanbreuseghem, 1952) was employed to isolate keratinophilic fungi. Inoculation of soil sample in sterilized petriplates was done with the help of sterilized spatula in laminar flow. Whole experiment was conducted in aseptic chamber. Petriplates were half-filled with the soil samples, moistened with distilled water and baited with sterile human hair (not exceeding the age of 20 years). The plates were wrapped in papers, incubated at 28°C ±1 and examined for a period of 7-8 weeks at regular intervals for the development of fungal growth on baited hair. After proper growth of fungi they were isolated and inoculated on Sabouraud's Dextrose Agar (SDA) medium supplemented with chloramphenicol 50 mg L^{-1} and streptomycin sulfate 100 mg L^{-1}. Petriplates were incubated for 3 weeks at 28°C and the developed colonies were send to the National Center of Fungal Taxonomy, New Delhi (India) for identification of genera and species.

RESULTS AND DISCUSSION

The results of isolation of keratinophilic fungi are presented in Table 1, the data reveals that out of 54 soil samples so collected a total of 23 species of Keratinophilic fungi belonging to 11 genera were isolated. The maximum numbers of Keratinophilic fungi were isolated from soil samples of gardens (65%). Similar results were obtained by Chmel *et al.* (1972), who reported that garden soils with its high organic debris are rich sources of keratinophilic fungi. Next to gardens stand schools (52%), poultry (43%) and garbage dumping sites (34%) and hospitals (30%). 26.9% of keratiniphilic fungi were isolated from soil samples of poultry in Sindh, Pakistan (Soomro *et al.*, 2007). Moreover, in the present study the least number of keratinophilic fungi were isolated from soil samples of rivers (21%). The maximum number of keratinophilic fungi in the soil samples of gardens, schools, poultry, garbage dumping sites and hospitals may be attributed due to the excessive presence of hair (keratin) at these sites. In the present study most of the isolated keratinophilic fungi, viz., species of *Chrysosporium*, *Fusarium*, *Aspergillus*, *Alternaria* and *Rhizopus* are common saprophytes in soil and plant debris. Some of them are often recovered as laboratory contaminants.

The data present in the Table 2 reveals the percentage occurrence of keratinophilic fungi from different soil samples, among the isolated species the most predominant is *Microsporum gypseum* (59%). *Microsporum gypseum* has been isolated from five places out of six. This species was also reported as the second most common dermatophyte from soils of Madras and Mumbai (Deshmukh and Agrawal, 1983; Ramesh and Hilda, 1998). The other species of genus *Microsporum* which were found in less abundance are *M. canis* (22%) and *M. nanum* (20%). The second most common species isolated was *Aspergillus flavus* (57%). *Aspergillus flavus* has also been previously reported as the second dominant species in soils of Gorgan (19.5%) and Gonbad-e Kavus (19%) areas in Iran (Moallaei *et al.*, 2006). The other species of genus *Aspergillus* were *A. niger* (18%) and *A. fumigatus* (24%).

Table 1: Isolation of Keratinophilic fungi from different places in Jhansi (India)

Fungi isolated	Gardens	Schools	Poultry	Rivers	Hospitals	Garbage sites
Microsporum gypseum	+	+	+	–	+	+
M. canis	+	–	–	–	+	–
M. nanum	--	+	–	+	--	+
Chrysosporium tropicum	+	+	+	–	--	–
C. anum	--	–	–	+	+	–
C. lobatum	--	+	–	+	--	–
C. indicum	+	–	–	–	+	–
Aspergillus flavus	+	+	–	–	--	+
A. niger	--	+	–	+	--	–
A. fumigatus	--	+	–	–	+	+
Tricophyton mentagrophytes	+	–	+	–	--	–
T. equinum	--	+	+	–	+	–
T. rubrum	+	–	–	–	--	–
T. ajelloi	--	+	–	–	--	+
T. interdigitale	+	–	+	–	--	–
Fusarium oxysporum	+	–	–	+	--	+
F. solani	+	+	–	–	--	–
Rhizopus stolonifer	+	–	+	–	+	–
Alternaria alternata	+	+	–	–	--	+
Trichoderma viride	+	–	+	–	--	–
Candida albicans	+	–	+	–	--	–
Penicillium funiculosum	+	–	+	–	--	–
Paecilomyces fusisporuss	--	+	+	–	--	+
Total No. of species isolated	15	12	10	5	7	8
% of occurrence	65	52	43	21	30	34

+: Presence, -: Absence

Table 2: Frequency occurrence of keratinophilic fungi isolated from different soil samples in Jhansi (India)

Fungi isolated	Total No. of samples observed	No. of positive samples	Percentage value
Microsporum gypseum	54	32	59
M. canis	54	12	22
M. nanum	54	11	20
Chrysosporium tropicum	54	30	55
C. anum	54	8	14
C. lobatum	54	12	22
C. indicum	54	9	16
Aspergillus flavus	54	31	57
A. niger	54	10	18
A. fumigatus	54	13	24
Tricophyton mentagrophytes	54	12	22
T. equinum	54	16	29
T. rubrum	54	10	18
T. ajelloi	54	9	16
T. interdigitale	54	10	18
Fusarium oxysporum	54	15	27
F. solani	54	14	25
Rhizopus stolonifer	54	17	31
Alternaria alternata	54	14	25
Trichoderma viride	54	6	11
Candida albicans	54	8	14
Penicillium funiculosum	54	7	12
Paecilomyces fusisporuss	54	13	24

Chrysosporium tropicum (55%) was the third most abundant species. *Chrysosporium* species were also earlier reported from Indian soils (Deshmukh, 2004; Deshmukh and Agrawal, 1983; Kushwaha and Agrawal, 1976; Randhawa and Sandhu 1965; Sur and Ghosh, 1980). *Chrysosporium* species were previously recovered from many other countries (Al-Mussllam, 1989; Bagy and Abdel Malek, 1991; Van Oorschot, 1980; Al-Mussllam, 1989). In the present investigation genus *Tricophyton* is most predominant in terms of number of species. Five species of genus *Tricophyton* were isolated namely *T. mentagrophytes* (22%), *T. equinum* (29%), *T. rubrum* (18%), *T. ajelloi* (16%) and *T. interdigitale* (18%). The other species which were isolated in the present investigation include *Alternaria alternata* (23%), *Fusarium oxysporum* (27%), *F. solani* (25%), *Rhizopus stolonifer* (31%), *Candida albicans* (14%), *Trichoderma viride* (11%), *Penicillium funiculosum* (12%) and *Paecilomyces fusisporuss* (24%).

The isolation of keratinophilic from different sites (Table 1) is not uniform this could be due to difference in the organic matter of the soil. Organic matter content of soil is one of the major factors affecting the presence of keratinophilic fungi in soil (Chmel *et al*., 1972). The present study reveals that soils of gardens, schools, poultry and garbage dumping sites are rich in keratinophilic fungi.

The data adds the information on the flora of keratinophilic fungi of india. Although, the study of keratinophilic fungi is a challenging task we have put our efforts to isolate keratinophilic fungi from few places of Jhansi. Immense need of work is yet to be done on this particular aspect in this specific area.

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