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# Occurrence of Keratinophillic Fungi and Other Dermatophytes from Soils of Various Habitats of Hyderabad, A.P (India)

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**The main goal of the present study was to isolate and identify keratinophilic fungi and related dermatophytes in different soil of Hyderabad. Soil of school playground, public parks, agricultural area and hospitals, which are frequently visited by children, adults and occupational workers, were chosen for the study. A total of 112 soil samples of various habitats were screened for the revalence of these fungi by hair-baiting technique, of which 99 samples showed positive growth on hair baits. Public parks showed maximum number of positive samples (97%) followed by school playgrounds (88%), agricultural soils (84%) and hospitals (78%) respectively. Our findings revealed the presence of 242 fungal isolates from soils of various habitats. These isolates represented 30 species of 16 genera, predominated by six *Chrysopsorium* species followed by four species of *Trichophyton* and three species of *Microsporum* and *Aspergillus* each, besides a single species representing the genera Alternaria, Aphanoascus, Bipolaris, Fusarium, Cladosporium, Ctenomyces, Geotrichum, *Malabranchea, Mucor, Rhizopus, Paecilomyces, Penicillium and Scopulariopsis.*The high prevalence of keratinophilic fungi along with dermatophytes are important bioindicators of soil contamination with keratin remnants.Most of these fungi are potential pathogens causing mycoses and are a threat to all those individuals visiting or working in such environments. To our knowledge, this appears to be the first report concerning the isolation of keratinophilic fungi from Hyderabad soils.**

**Key words**: Keratinophilic fungi, Dermatophytes, Public parks, School playgrounds, Agricultural soil, Hospital soil.

The ubiquity of fungi in soil and various other environments is well recognized. However,certain groups of fungi are very specific about their substrate and nutritional requirement, among them keratinophilic fungi occupy an important place. Majority of these represent a host of filamentous fungi comprising hyalo-hyphomycetes and many other taxonomic groups[1]. Hyphomycetes include the dermatophytes, and many other nondermatophytic fungi mainly occurring as saprophytes in soil, and some are known plant pathogens[2]. They have the ability to colonize and degrade various keratinous substrates like skin, nail, hair, hooves, wool and other cornified appendages in various soil environment completely. Hence, they are important ecologically, as they cycle one of the most abundant and highly stable animal proteins on earth i.e. keratins[3]. Keratinolytic fungi occur in many natural and manmade habitats and exist in communities together with keratinophilic fungi, that have weaker affinity to keratin and utilize chiefly the products of its decomposition[4]. While dermatophytes are known to cause dermatophytosis which are superficial cutaneous infections of keratinized tissues i.e skin, hair and nails of humans, animals and birds. They create inflammation, pruritus and desquamation by invading the keratin of the stratum corneum.These infections are contagious

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and are have been reported from different parts of the world.

The modern Hyderabad is spread over an area of 650 km$^2$ (250 sq mi), making it one of the largest metros in India.The city is nestled on Deccan Plateau and rises to an average height of 540 m above the sea level. The city lies at 17.366° N latitude and 78.476° E longitude. Summer day temperatures rise to a maximum of 44°C often in May and night temperatures in winters may fall to a minimum of 12 °C. Thus the metropolitian is quite dry and hot from February to May, much humid from end of June to September and mildly cool from October to January. Soils of locations with such tropical climatic conditions are known to serve as good habitat for the growth of keratinophilic fungi and dermatophytes. Several researchers have explored soils of India from different states .These fungi were isolated from Indian soils from different habitats viz. public places[5,3]. Agricultural soils and parks[6], water sediments[7], sewage waters[8], Hilly areas[9] and birds and their environment[10]. However, to the best of our knowledge, soils of Hyderabad city has not been explored yet for the prevelance of keratinophilic fungi. Our attempt in the present work was to explore soil of different habitats, for the occurrence of keratinophilic fungi and related dermatophytes. This study will help children, adults, farmers and gardeners who frequent these places and are at a risk of exposure to these potential pathogens through direct soil contact. To our knowledge this is the first report on the distribution of keratinophilc fungi and related dermatophytes from Hyderabad soil.

## MATERIALS AND METHODS

### Collection of soil samples

One hundered and twelve soil samples were collected from various locations.The superficial layer of soil at a depth not exceeding 2-5 cm was scooped and placed in sterile polyethylene bags, brought to the laboratory, and stored at 4°C for a maximum of two weeks. I thank Mr. Ibrahim and Mr. Ashok for their assistance during soil samples collection.

### Isolation of Keratinophilic fungi

Keratinophilic fungi were isolated using the hair baiting technique[11]. Sterile Petri dishes were half filled with thoroughly homogenised soil samples and the soil was moistened with sterile distilled water; the amount of water added varied from sample to sample, depending on the moisture content of the sample. It was then baited with short strands of sterilized human hair. Strands of human hair were spread uniformly as baits on the surface of moistened soil samples. The plates were then incubated at 28±2°C and then examined daily from day 5 for any fungal growth over a period of 4 -8 weeks. Five replicates were set for each sample. Samples of hair showing fungal growth were carefully observed, and then aseptically picked up and cultured on Sabourauds dextrose agar (SDA, Scharlau Chemie, Spain) supplemented with chloramphenicol (0.05mg/ml) alone and another set with chloramphenicol (0.05 mg/ml) and cycloheximide (0.5 mg/ml) respectively.They were sub-cultured to get pure cultures.

### Identification

Identification of the purified fungal isolates was based on the monographs of (12-15) using macro- and micro-morphological features.

## RESULTS

The results of isolations on hair baits are presented in Table 1. Out of 112 soil samples collected only 99 samples showed positive growth. Public parks showed maximum number of positive samples (97%) followed by school playgrounds (88.88%), agricultural soils (78.26%) and hospitals (84.21%) respectively. Our findings revealed the presence of 242 fungal isolates from soils of various habitats. These fungi represented 30 species of 16 genera. Public parks showed presence of 80 fungal isolates followed by 61 isolates from school playground , 58 isolates from agricultural area and 43 isolates from hospital soils. Among the important genera dominating Hyderabad soils, *Chrysosporium* was the most common with 97 isolates; the others being 31 isolates of *Trichophyton* ,*Microsporum*,*Aspergillus* 25 isolates each and 18 isolates of *Fusarium*.Table 2, Fungal isolates were predominated by six *Chrysopsorium* species followed by four species of *Trichophyton* and three species of *Microsporum* and *Aspergillus* each. graph-1,2 The percentage distribution of the keratinophillic fungi and dermatopytes are shown in Graphs( 1,2).Soils of Hyderabad harboured a variety of keratinophillic

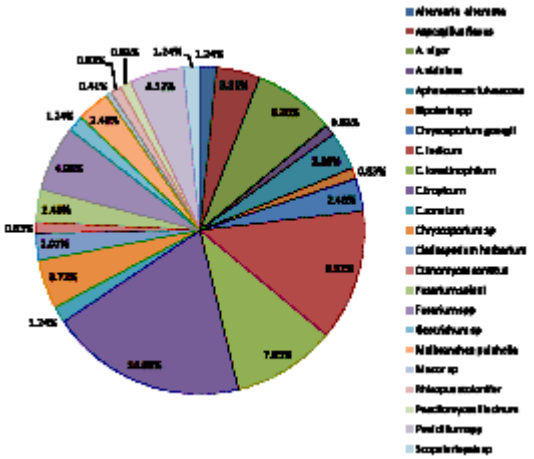
fungi in comparison to dermatophytes .*C.tropicum (14.88%)* was found to be the most dominating species representing keratinophillic fungi while it was *M.gypseum* (7.02%) amongst the dermatophytes.Percentage distribution of some important fungi among others are as follows; *C .indicum (9.92%),C.keratinophillum(7.85%)*, *Aspergillus niger* (6.20%), *Trichophyto mentagrophytes* (5.79%), *Penicillium spp* (4.13%), *Aspergillus flavus* (3.31%), *Aphanoascus fulvescens* and *Trichophyton rubrum* (2.89%) each. Almost all the fungi isolated were present in Public parks. C.*tropicum*, *C.indicum* and *C.keratinophillium* was present in large numbers in all soils screened.

**Table 1.** Percent prevalence of keratinophilic fungi and related dermatophytes in soil samples of various habitats

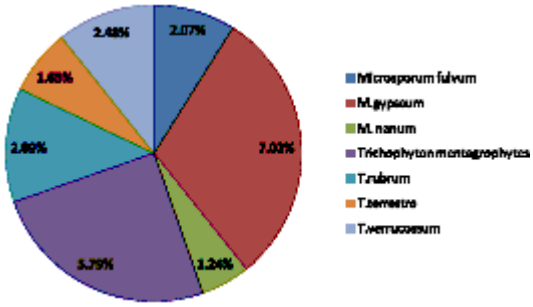
| Soil collection site | Public parks | School playground | Hospital | Agricultural area | Total |
|---|---|---|---|---|---|
| No .of samples examined | 34 | 36 | 23 | 19 | 112 |
| No .of samples positive | 33 | 32 | 18 | 16 | 99 |
| Distribution (%) | 97 | 88.88 | 76.26 | 84.21 | 88.39 |

**Table 2.** Frequency distribution of keratinophillic fungi and related dermatophytes in various soils of Hyderabad

| Fungi isolated | Public parks | School playground | Hospital | Agricultural area | Total |
|---|---|---|---|---|---|
| *Alternaria alternata* | 1 | 0 | 0 | 2 | 3 |
| *Aspergillus flavus* | 2 | 3 | 1 | 2 | 8 |
| *A. niger* | 5 | 3 | 5 | 2 | 15 |
| *A.nidulans* | 1 | 0 | 0 | 1 | 2 |
| *Aphanoascus fulvescens* | 2 | 1 | 4 | 0 | 7 |
| *Bipolaris spp* | 1 | 1 | 0 | 0 | 2 |
| *Chrysosporium georgii* | 2 | 1 | 1 | 2 | 6 |
| *C. indicum* | 7 | 5 | 4 | 8 | 24 |
| *C. keratinophilum* | 6 | 5 | 3 | 5 | 19 |
| *C.tropicum* | 10 | 12 | 6 | 8 | 36 |
| *C.zonatum* | 0 | 2 | 0 | 1 | 3 |
| *Chrysosporium sp* | 3 | 2 | 0 | 4 | 9 |
| *Cladosporium herbarium* | 2 | 3 | 0 | 0 | 5 |
| *Ctenomyces serratus* | 2 | 0 | 0 | 0 | 2 |
| *Fusarium solani* | 2 | 1 | 2 | 1 | 6 |
| *Fusarium spp* | 5 | 2 | 1 | 4 | 12 |
| *Geotrichum sp* | 2 | 0 | 0 | 1 | 3 |
| *Microsporum fulvum* | 2 | 2 | 0 | 1 | 5 |
| *M.gypseum* | 6 | 5 | 2 | 4 | 17 |
| *M. nanum* | 1 | 2 | 0 | 0 | 3 |
| *Malbranchea pulchella* | 2 | 0 | 0 | 4 | 6 |
| *Mucor sp* | 0 | 1 | 0 | 0 | 1 |
| *Paecilomyces lilacinum* | 1 | 1 | 0 | 0 | 2 |
| *Penicillium spp* | 4 | 2 | 1 | 3 | 10 |
| *Rhizopus stolonifer* | 0 | 0 | 2 | 0 | 2 |
| *Scopulariopsis sp* | 2 | 1 | 0 | 0 | 3 |
| *Trichophyton mentagrophytes* | 4 | 3 | 5 | 2 | 14 |
| *T.rubrum* | 2 | 2 | 3 | 0 | 7 |
| *T.terrestre* | 1 | 1 | 0 | 2 | 4 |
| *T.verrucossum* | 2 | 0 | 3 | 1 | 6 |
| *Total* | 80 | 61 | 43 | 58 | 242 |
| *Percent frequency* | 33.1% | 25.2% | 17.8% | 24.0% | 100.0% |



**Graph 1.** Percentage distribution of keratinophillic from various soils of Hyderabad




**Graph 2.** Percentage distribution of dermatophytes in soils of Hyderabad

## DISCUSSION

Throughout the world great interest is shown by research workers in soil mycoflora that can degrade keratinized residues. Every keratinophilic fungi can be considered a potential pathogen and its epidemiological study gains importance firstly because of the extreme resistance of keratin to biological attack, and secondly it's pathogenic potential[16].

Our findings reveal that soils of Hyderabad to be a rich source of both the keratinophilic fungi and dermatophytes.The high positivity rates of fungal isolation (97%) from Public parks and School playground (88%) could be attributed to abundant keratinic material mainly in the form of hairs and feathers. Plant litter, soil pH, moisture content, organic matter and manure besides the keratinic debris in the soil of Public parks and gardens provide the most congenial environment for these fungi to proliferate.

The high percentage prevalence of the genus *Chrysosporium* in our study is indicative of the fact that it can withstand wide range of temperatures and can survive in soils of varying keratin load. It has been reported that the wide spread distribution of the genus *Chrysosporium* could be attributed to its mesophilic, thermotolerant and hydrophilic nature[17]. Occurrence of species of *Chrysosporium was* reported from varied habitats of Indian soil,Srivasata[18], reported 14 species of the genus *Chrysosporium* from paddy fields, besides this there are reports of its occurrence from public parks, indoor and outdoor hospital soils and other habitats therefore it shows that this fungi dominates Indian soil mycobiota as it is well adapted to warmer conditions[19,20-21]. Incidence of infection with different species of *Chysoporium* have increased in the past few years. *Chrysosporium zonatum* was reported to cause disseminated infection in a patient with chronic granulomatous disease[22]. Non sporulating *Chrysopsorium sp* was found to cause invasive infection in a Neutropenic patient[23]. Lyskova[24] isolated *Chrysosporium queenslandicum, C. sulfureum, C. tropicum, Malbranchea pulchella, Myriodontium keratinophilum* from infections of the skin and nails of patients in the Moravian-Silesian Region (Czech Republic) .*Chrysosporium keratinophilum* a fungus present in cultivated soil was isolated from a case of fungal infection of skin and nail plates[25]. Saidi et al., reported comb lesion in two different breeds of chicken in India by*Chrysosporium tropicum*[26].

We isolated three species of *Aspergillus*, *Fusarium solani ,Cladosporium herbarium, Alternaria alternata ,Chaetomium sp,Mucor sp* and other keratinophilic fungi from soil public parks, Agricultural soils and school playground .However many of these fungal species were absent in soil of hospitals, which could be due to competitive utilization of keratinic material by dermatophytes and other competent keratinophillic fungi present in these soils.Indian soil habitats like parks ,school playground ,indoor and outdoor soils *of* hospitals have shown the presence of keratinophilic fungi like *Aspergillus, Mucor, Rhizopus Penicillum,Trichoderma,Malranchea fulva,Penicillium expansum and* and many others in varying numbers[20,27].

Worldwide fungal infections that were not previously recognised as pathogenic have become increasingly common[28,29]. *Fusarium* cause a broad spectrum of infections in humans including superficial infections, such as keratitis and onychomycosis, as well as locally invasive and disseminated infections[30,31,32]. *Aspergillus spp* are commonly isolated from the soil, plant debris, and the indoor environment, including hospitals.They may cause lifethreatening infections especially in immunocompromised hosts[33].Some species of *Aspergillus* cause cutaneous aspergillosis[34], while in most cases, *Aspergillus* is introduced to the lower respiratory tract by inhalation of the infectious spores and *Aspergillus* infection may also disseminate aematogenously to other organs, including the brain[35]. Some species of *Cladosporium* are known to be the cause of cerebral and cutaneous phaehyphomycoses[36]. Isolation of these fungi in our study from various soils is a sign of alarm to those individuals working in these environments.

Present study however revealed fungal isolates belonging to nondermatophyte to be more common than dermatophytes. Among the dermatophytes isolated, the geophilic dermatophyte *M .gypseum* was the most predominant followed by *Trichophyton mentagrophytes,T.rubrum , T.terestre and T.verrucossum,* although *M.fulvum and M .nanum* were isolated but in low numbers. Dermatophytic infections are common disorders worldwide and dermatophytes represent those type of fungi that cause infection of the hair, skin and nails[37-39]. Variety of clinical manifestations are represented by Tinea infections besides Onychomycosis. The high prevalence of superficial mycotic infections makes these one of the most frequent forms of infection.

Pathogens responsible for skin mycoses are primarily anthropophilic and zoophilic dermatophytes and few geophilic from the genera *Trichophyton, Microsporum* and *Epidermophyton* . *Trichophyton rubrum, T.* m*entagrophytes, M.gypseum M. canis, M. audouinii, T. tonsurans* and *T. verrucosum* are the most common, but the attack rates and incidence of specific mycoses can vary widely. *M.gypseum* causes ringworm of the scalp and glabrous skin in human and animals[40,41,42].

There appears to be considerable inter- and intra-continental variability in the global incidence of these fungal infections. Several workers have screened children's and adults of all ages for dermatophytosis all over the world . Ndako et al., screened school children in the age group of 5-13 years and reported infection from 91% children. Both dermatophytes and nondermatophytes were isolated. The etiological agents of dermatophycoses were *M. canis, T. verrucosum, T. rubrum, T. mentagrophyte* and many others. *A. flavus, A. niger, Penicillium* sp, *Mucor* sp, *Trichoderma* sp. and *A. fumigatus* constituted the non-dermatophytes associated with cutaneous infections. They reported a higher frequency of dermatophytosis occurred more in children with greater propensity for play, interaction with domestic animals[43]. Similarly several dermatophytes were isolated from 162 children of nomadic herdsmen who were in a continuous exposure to different animals[44]. Many zoophilic and geophilic fungi from genera *Acremonium, Fusarium* and *Aspergillus* were cultured occasionally from infected nail plate of farmers[45].

Hence amongst the wide spectrum of fungal species that was isolated from soils of Hyderabad,most are pathogens causing skin lesions and mycotic infections in birds,animals and humans. Therefore individuals like Farmers, Gardeners and children who are in constant exposure to soil habitats are exposed to these pathogens on regular basis. Additionally ocupations which cause prolong exposure to specific working conditions like humid environments with varying keratin loads in gardens and agricultural fields further expose them to a greater risk of invasion by these fungi .

## CONCLUSION

This study revealed that all the soils screened were contaminated with dermatophytes, Keratinophillic and keratinolytic fungi. The high percentage prevalence in public parks could be due to invasion of these places by animals like dogs, cats, pigs, cows, bullocks,various birds and of course humans activities. Thereby enriching these soil environments with keratinic materials. Our study highlights the distribution pattern of keratinophillic fungi as they are important bio

indicators of environmental pollution with keratin remnants. Both dermatophytes and nondermatophytes being potential pathogens, pose a threat to all those who are regularly in contact with such soil habitats. Special hygiene awareness programme and implementation of precautions will ensure safety of these individuals.

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