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Occurance of Keratinophilic Fungi from the Soils of Chhattisgarh

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Abstract: ***Soil is considered as the most complex media of microbial inhabitants including fungi. Soils differ in chemical composition and support the growth of specific fungal flora, however, majority of the soil fungi are autochthonous being indigenous to soil environment. Some of the soil fungi are associated with human and animal diseases and cause chronic problems. The soil containing keratin materials promotes the growth of keratinophilic fungi. Keratinophilic fungi grow and reproduce well on keratin materials such as skin, hair, feather, fur, horn, wool, hoop and nail. The distinction between the keratinolytic and keratinophilic fungi is based on keratin utilization and degradation in naturals and artificial condition and the dermatophytes have potency to invade keratinized tissue including hair, nail and skin. The dermatophytes have been further divided into three ecological groups as Geophiles, Zoophiles and Anthrophiles.The prevalence of dermatophytes varies according to the geographical location, climate or living conditions, and the environment to which the susceptible organism is exposed.In the present study, two techniques have been adopted in the qualitative and quantitative isolation of these fungi from the soil samples collected from various location of Chhattisgarh state viz. Ambikapur, Bilaspur, Jashpur, Korba, Raigarh and Raipur districts, by Surface Soil Dilution Plating (SSPD) and hair/feather Baiting Technique (HFBT). Colonies developed by this technique are easy to pick-up for subsequent purification. Purification was made with Sabouraud's Dextrose Agar medium at 32^0C. About one hundred and nineteen isolates belongs to twelve different genera were isolated from the soil. The dominating genera are: Microsporum pulchela, Microsporum gypseum, Trichophyton mentagrophytes, Arthroderma gypseum, Chrysosporium tropicum, Chrysosporium indicum, Chrysosporium pannicola, Malbranchea arcuata, M. pulchella, Acremonium sp., Aspergillus flavous, Aspergillus niger, Aspergillus terreus, Cladosporium cladosporoides, Geotricum sp. and Humicola griesea etc. The data obtained indicates that the soil contain large number of keratinophilic fungi which may cause dermatomycosis or keratinomycoses, in the person those are involved in building work, leather work or in the sowing rice.***

Keywords: Fungi, microbial inhabitants, Chhattisgarh

1. Introduction

Soil is considered as one of the most complex microbial habitats in which many fungi complete their life. Different soils have specific fungal floras, but the majority of species found in them are cosmopolitan in distribution. Some soil fungi are potential pathogenic to both human and animal. Soils that are rich in keratinous materials are the most conducive to the growth, development and occurrence of keratinophilic fungi (Randhawa & Sandhu 1965, Garg 1966, Marcanti et al., 1980, Ogbonna & Pugh 1987, Ramesh & Hilda 1989, Deshmukh & Shukla 2000, Deshmukh 2002, Nwaze & Okafor 2010 and Jain & Sharma 2011). Keratinophilic fungi grow well and reproduce on keratin substrates such as skin, hairs, nails, feathers, horns, wools and hoops etc. They utilized keratin as carbon source for their energy production (Graser et al.2006) Keratinophilic fungi are important, ecologically; because they are engaged in biodegradation of approximately two billion tons of keratinic waste dumped annually in Indian scenario and recycle them to simpler forms. Due to their variable distribution pattern and highly adaptive nature, they survive in diverse environmental conditions. Human and animals are the sudden hosts and cause dermal infections. Most of such infections are caused by the keratinophilic fungi known as dermatophytes (Singh et al. 2009 and Shukla & Chouhan 2011).

They have capacity to invade keratinized tissue of the body including skin hairs and nails. Dermatophytes have been divided into three ecological groups: Geophiles, Zoophiles and Anthropophyles. The prevalence of dermatophytes varies according to geographical location, season or living conditions, and the manipulation to which the susceptible human, animal or birds are exposed (Kawasaki 2011, Rippon 1988, Robert & Pihet 2008, Seebacher et al. 2008 and Sigler &Carnicheal 1976). However, they are more frequent in countries having hot and humid climatic conditions.

2. Materials & Methods

Collection of soil samples:

Soil samples were collected from various sites (Ambikapur, Bilaspur, Jashpur, Korba, Raigarh and Raipur Districts of Chhattisgarh, India) in presterilized polythene bags. The surface soil (depth not exceeding beyond two to three centimeter) was collected with the help of sterilized spatula. All the polythene bags were labeled indicating the date and the sites of collection etc. These samples were then tightly closed to maintain the original moisture contain and brought to laboratory then kept in refrigerator till further analysis (Saxena 1955).

Baiting of soil samples:

Each soil was thoroughly homogenized, and a sufficient amount each of the soil samples was taken in separate pre-sterilized Petri plates. Sterile human hairs, feathers and nails were used for the isolation of keratinophilic fungi by Baiting technique. In addition to these, the sterilized distilled water was added to provide RH and moisture to the soil. The hairs, nails and feathers were seeded uniform and at equal distance on Petri plates containing wet soil samples. After inoculation of baits, each Petri plates sealed with cellophane tape and

then incubated at 28^0C± 2^0C temperature for 4-5 weeks. Plates were regularly observed for any growth of fungi on the baits. After observing the growth under a stereoscopic binocular microscope, isolates cultured on Sabouraud´s Dextrose agar supplemented with chloramphenicol 50 mg/L and cycloheximide 500mg/L.

Isolation, Purification and identification of fungi:
Keratinophilic fungi were isolated by the selective isolation using hair bait technique (Vanbreuseghem, 1952).Fungal growth appeared on the hairs, nails and feathers baits after three to four weeks of incubation. The isolates were examined under the microscope especially their morphology, colonial characteristics (mounted with cotton blue and lacto phenol solution) and other factors. Identification was made on the basis of micro and macro stages of conidia on mycelium, nutritional requirement and biochemical characteristics etc. However, some fungi which are appear to be different on the basis of conventional typing method may be very closely related to gnomically similar. After preliminary examination, the fungi were transferred and maintained on Sabouraud´s Dextrose Agar medium supplemented with chloramphenicol 50 mg/L and cycloheximide 500mg/L to avoid the growth of gram-positive and gram-negative bacteria.

3. Results

About one hundred fifty different soils samples were analyzed for distribution of keratinophilic fungi. A total of 119 isolates belongs to twelve genera and fourteen species were isolated from the soil samples Each soil samples have different composition of fungal flora because of difference in soil pH that is ranging from 6.5 to 10.5, and available organic materials, that support the growth specific fungi. Present study shows an overall prevalence of keratinophilic fungi in soils of Surguja, Chhattisgarh (Saxena 1955, Pandey et al. 2009, Padhya et al. 1967, Rai et al. 1971). The result of the distribution of these fungi was presented in Table 1 & Table 2. The isolated fungi from soil samples are*: Arthroderma gypseum, Acremonium sp., Aspergillus flavous, A. niger, A. terreus, Cladosporium cladosporoides, Chrysosporium tropicum, Chrysosporium indicum, Chrysosporium pannicola Geotricum sp., Humicola griesea, Malbranchea arcuata, M. pulchella, Microsporum gypseum, Trichophyton mentagrophytes, Curvularia lunata and Bipolaris australiensis*, are distributed in the soils sample. A total of seventy eight samples were found positive out of one hundred fifty soil sample examined. The highest distribution of keratinophilic fungi (88%) were recorded in soil sample collected from Jashpur district. The frequency of occurrence of keratinophilic fungi in the soils sample of Ambikapur district was 52%, and in the samples of Bilaspur and Raipur with 48% of each. The samples of Raigarh and Korba were found with 36%-52% respectively. Other worker also recorded about similar results Jain & Sharma 2011, Kanbe 2008, Nigam & Kushwaha 1990, Saxena et al.2004 Verma et al. 1982 and Vidyasagar et al. 2005.

Table 1: Distribution of keratinophilic fungi in different Districts of Chhattisgarh state

S. No.	*Name of fungus*	*Distribution of keratinophilic fungi in different Districts of Chhattisgarh*					
		Ambikapur	Bilaspur	Jashpur	Raipur	Raigarh	Korba
1	*Arthroderma sp.*	02	--	03	03	--	05
2	*Acremonium sp*	--	02	--	04	01	--
3	*Aspergillus flavous*	01	03	02	01	--	--
4	*A. niger*	02	01	01	01	01	02
5	*A.terreus*	--	02	02	--	--	--
6	*Cladosporium cladosporoides*	--	--	--	01	01	02
7	*Chrysosporium tropicum*	01	02	04	--	02	03
8	*Chrysosporium indicum*	02	--	01	--	--	01
9	*Chrysosporium pannicola*	--	--	04	01	--	--
10	*Geotricum sp*	--	--	--	01	--	--
11	*Humicola griesea*	01	--	06	--	04	--
12	*Malbranchea arcuata*	04	02	02	04	--	--
13	*Microsporum gypseum*	--	--	04	--	--	04
14	*Trichophyton mentagrophytes*	01	02	05	--	04	05
15	Curvularia lunata	02	--	02	--	--	--
16	Bipolaris australiensis	--	01	--	02	--	--
Total colonies		16	15	36	18	13	21
Total no. of samples positive		13	12	22	12	09	10
Total no. of samples examined		25	25	25	25	25	25
Distribution in soils (%)		52%	48%	88%	48%	36%	40%

Table 2: Keratinophilic fungi isolated on different baits

S. No.	*Name of fungi*	*Baits used for isolation of fungi*			*Frequency*
		Nails	Hairs	Feathers	
01	***Arthroderma sp.***	---	---	+++	08.66
02	***Acremonium sp***	+++	---	---	04.66
03	***Aspergillus flavous***	---	---	+++	05.33
04	***A. niger***	+++	+++	---	05.33
05	***A.terreus***	---	+++	+++	02.66
06	***Cladosporium cladosporoides***	+++	---	---	02.66
07	***Chrysosporium tropicum***	---	+++	+++	08.00
08	***Chrysosporium indicum***	+++	---	---	02.66
09	***Chrysosporium pannicola***	+++	---	---	02.66
10	***Geotricum sp***	---	+++	---	00.66
11	***Humicola griesea***	+++	---	---	07.33
12	***Malbranchea arcuata***	---	+++	+++	08.00
13	***Microsporum gypseum***	+++	+++	---	05.33
14	***Trichophyton mentagrophytes***	---	+++	+++	11.33
15	**Curvularia lunata**	+++	---	---	02.66
16	**Bipolaris australiensis**	+++	---	---	02.00

4. Discussion

The occurrence of keratinophilic fungi in different soils have been reported from all over the world. Degradation of keratin waste in nature and recycling of materials is one of the important ecological mechanisms in soil. Due to spearing adaptability on keratin, these fungi may cause human and animal mycoses. However, the isolated fungi are generally considered as saprophytic in nature, but occasionally, they cause infection to human and animal. A total of 150 samples were tested, only 78 samples yielded keratinophilic fungi which were further categorized in sixteen species of twelve genera including *Trichophyton mentagrophytes*, *Chrysosporium tropicum* and *Humicola griesea* with 11.33%, 8.66% and 7.33% frequency in amongst the isolates, respectively.

Trichophyton mentagrophytes was found most dominating fungal species from the soil samples. It was also reported by various workers as dominant in the microbial community of the Indian soils because of its adaptability in tropical and sub tropical soils. *Arthroderma sp, Chrysosporium tropicum Malbranchea arcuata* and *Humicola griesea* were the next most frequent keratinophilic fungi also reported by various workers in Indian soils. Besides of these fungi, other dermatophytes were also occurred in Indian soils with low frequency.

5. Conclusion

Dermatophytes cause infections on the skin, hair and nails due to their ability to obtain nutrients from keratinized materials. The organisms colonized the keratin contained tissues and cause inflammation as a result of host response. Infections are usually restricted to the dead layer of the epidermis because of their inability to penetrate viable tissue of an immunocompetent host

The soils of Surguja district of Chhattisgarh state is also the source of different keratinophilic fungi. Chhattisgarh is well known as rice bowl of India, and it´s cultivation required comparatively high relative humidity in soils which favor the growth of keratinophilic fungi. The dermatophytes attack on epidermal tissue, which came into the contact of soil during showing of rice, hence, can be infected. However, the workers those who involved in showing of rice have threats to be infected by dermatophytes.

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