Prevalence of keratinophilic fungi in *usar* soils of Uttar Pradesh, India

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Abstract

Alkaline soils commonly called as Usar soil having pH 7.5 to 11.0 is commonly found in regions having poor drainage and little percolation. These soils occupy vast tracts of barren lands in the northern parts of India. These areas are frequently visited by man and animals, however there are no reports on the occurrence of keratinophilic fungi from the *Usar* soils. The purpose of this research was to study the occurrence of keratinophilic fungi in the alkaline Usar soil. To investigate the occurrence of soil keratinophilic fungi from Usar soil, 120 samples were collected from five districts of Uttar Pradesh viz. Lucknow, Bareilly, Azamgarh, Balia and Pratapgarh. Keratinophilic fungi were isolated by the hair baiting techniques using human hair as keratin bait. The cultures were identified using macro- and micro morphological features. Eighty-tow strains of keratinophilic fungi were recovered from 120 (68.33%) soil samples. The isolated fungi were composed of ten species of five genera viz. Chrysosporium indicum (19.16%), Microsporum gypseum (12.5%), Chrysosporium tropicum (10.0%), Chrysosporium pannicola (7.5%), Trichophyton terrestre (5.83%), Chrysosporium lucknowense (4.16%), Chrysosporium state of Ctenomyces serratus (3.33%), Gymnascella dankailensis (3.33%),Gymnoascus reessii (1.66 %) and Trichophyton mentagrophytes (1.66 %). The presence of keratinophilic fungi in the Usar soils of Uttar Pradesh, India indicates that these fungi do occur at higher alkalinity.

Introduction

Alkaline soils commonly termed as *Usar* soil occupy vast tracts of barren lands in India. Such soils are commonly found in regions having poor drainage and little percolation. The pH of such soil ranges from 7.5 to 11.0 and it is mostly white alkaline type. The salts generally present in these soils are the carbonates, bicarbonates, sulphate and chlorides of calcium and magnesium. The carbonates and bicarbonates of sodium are present in large quanti-

ties than the sulphates and chlorides. Thus the free exchangeable sodium ions make these soils highly alkaline and thus raise pH considerably. Such soils have either no vegetation at all or have patches of poor grass growth, interspread with areas which shows prominent whitish salt excrescences. The soil being devoid of vegetation coupled with low moisture content is subjected to high temperature, drought and intense solar radiations for greater part of the year which makes it an interesting habitat for such study. Various workers have isolated a large number of fungi from usar soil.¹⁻³ However, we have not found the reports on the occurrence of keratinophilic fungi from these alkaline soils, although these areas are frequently visited by man and animals. Therefore the present work was conducted to study the occurrence of keratinophilic fungi from Usar soil from five districts of Uttar Pradesh, India.

Materials and Methods

One hundred and twenty soil samples were collected from five sites of *Usar* soils in five districts of Uttar Pradesh viz. Lucknow, Bareilly, Azamgarh, Balia and Pratapgarh during March 2009 to February 2010 (Table 1). The samples were collected from the superficial layer of soil at a depth not exceeding 3-5 cm with a plastic spoon in sterile polyethylene bags, brought to the laboratory and stored at 15° C for a maximum of two weeks if not processed promptly.

Keratinophilic fungi were isolated by the hair baiting technique of Vanbreuseghem⁴ using human hair as keratin bait. For this purpose, sterile Petri dishes half filled with the soil samples and moistened with sterile water were baited by burying sterile human hair in the soil. These dishes were incubated at room temperature $(28\pm1^{\circ}C)$ and examined daily from the fifth day for fungal growth over a period of four weeks. After observing the growth under a stereoscopic binocular microscope, isolates were cultured on Sabouraud's dextrose agar supplemented with chloramphenicol (50 mg/L) and cycloheximide (500 mg/L).

These fungi were identified based on the monographs of Sigler and Carmichael,⁵ Oorchschot,⁶ Currah,⁷ von Arx,⁸ Cano and Guarro⁹ using macro- and micro morphological features.

Results and Discussion

The results of the isolations are presented in Table 1. They reveal that out of 120 samples Correspondence: Sunil Kumar Deshmukh, Department of Natural Products, Piramal Life Sciences Limited, 1, Nirlon Complex, Off Western Express Highway, Near NSE Complex, Goregaon (East), Mumbai 400 063, India. Tel. +91.22.3081.8819 - Fax+91.22.3081.8703. E-mail. sunil.deshmukh@piramal.com

Key words: *usar* soil, pH, keratinophilic fungi, Uttar Pradesh, India.

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only 82 yielded keratinophilic fungi which were categorized in ten species of five genera viz. Chrysosporium indicum (19.16%), Microsporum gypseum (12.5%), Chrysosporium tropicum (10.0%), Chrysosporium pannicola (7.5%), Trichophyton terrestre (5.83%), Chrysosporium lucknowense (4.16%), Chrysosporium state of Ctenomyces serratus (3.33%), Gymnascella dankailensis (3.33%), Gymnoascus reessii (1.66%) and Trichophyton mentagrophytes (1.66%).

Chrysosporium indicum was the most dominant fungal species isolated from *Usar* soils. It was reported from Indian soils by various workers¹⁰⁻¹² and dominates the mycobiota of the Indian soils because it is adapted to warmer conditions of India.¹⁰

Microsporum gypseum (12.5%) was the next most frequent isolate followed by cosmopolitan Chrysosporium tropicum (10.0%). Microsporum gypseum is a geophilic dermatophyte, universal in soil and has been encountered in one or more countries of each continent.13 It causes ringworm of the scalp and glabrous skin in human beings.14 It has been reported from Indian plains.^{10,15-17} Chrysosporium tropicum is reported from various parts of India.^{10,15,18} Chrysosporium pannicola was 7.5% in distribution and was reported from Indian soils as Trichophyton evolceanui by Randhawa and Sandhu,¹⁶ then Garg¹⁰ placed it in the genus Chrysosporium as C. evolceanui. While revising the genus Chrysosporium, Oorchschot⁶ placed it as C. pannicola. It is reported from Indian soils.^{10,16, 19-20}

Regarding other dermatophytes, we encountered *Trichophyton mentagrophytes* and *T. terrestre* which were recovered from 1.66% and 5.83% soil samples respectively. *Trichophyton*



species isolated The other were (4.16%), Chrysosporium lucknowense Chrysosporium state of Ctenomyces serratus (3.33%), Gymnascella dankailensis (3.33%), and G. reessii (1.66%). Chrysosporium state of Ctenomyces serratus was reported from Indian soils by various workers.^{16,19,28} Gymnoascus reessii was reported by Deshmukh²¹⁻²² and Ghosh and Bhatt²⁹ and G. dankailensis by Verekar and Deshmukh²⁰ from Indian soils. Chrysosporium lucknowense was first time reported from Lucknow by Garg¹⁰ followed by Deshmukh and Shukla,³⁰ Nigam and Kushawaha,31 Dixit and Kushwaha,32 Saxena et al.33 Species of Chrysosporium are known to cause diseases in man and animals.34³⁷.Similarly species of *Gymnascella i.e. G. hyalinospora* causes the infection in human being.³⁸ Thus, these fungi may be regarded as opportunistic pathogens.

Ghosh and Bhatt²⁹ reported many keratinophilic fungi like Aphanoascus fulvescens, Chrysosporium farinicola, C. keratinophilum, C. tropicum, C. xerophilum, C. zonatum, Gymnoascoideus petalosporus, Malbranchea aurantiaca, Microsporum gypseum, Rollandina hyalinospora, Uncinocarpus orissi, and U. reesii. from Chilka lake-side soil which is semi- alkaline/ alkaline in nature.

Aphanoascus durus, Aphanoascus punsolae, Auxarthron kuehnii, Chrysosporium indicum, Chrysosporium tropicum, Chrysosporium sp., Chrysosporium state of Ctenomyces serratus were reported from the vicinity of meteorite crater soils of Lonar (India).³⁹ The lake water is alkaline, having an average pH of 10.5-11.2 and temperature of 35°C. Salts and minerals like sodium, chloride, carbonates, fluorides and bicarbonates are found in the lake. Alkalinity of the lake is attributed to the high content of sodium carbonate.⁴⁰

Recently Jain and Sharma⁴¹ isolated Trichophyton rubrum, T. simmi, T. mentagrophytes, T. terrestre, Chrysosporium tropicum, Chrysosporium indicum, Chrysosporium pannicola as C. evolceanui, Gymnoascus reessii, Ctenomyces sp., Microsporum gypseum, M. canis, M. audouinii and Epidermophyton floccosum from the soils of Jaipur with reference to soil pH. From their study it appeared that the fungi preferred the soil pH of 7.0 to 8.5. The fungi like Trichophyton simmi, T. mentagrophytes, T. terrestre, Chrysosporium tropicum, Chrysosporium indicum, Chrysosporium pannicola, Microsporum gypseum and M. canis can survive even higher pH.

A perusal of the Table 2 indicates that these fungi preferred neutral to alkaline soil. The

Table 1. Distribution of fungi in soil samples.

	Source of soil samples	Lucknow	Bareilly	Azamgarh	Balia	Pratapgarh	Total	Percentage distribution
	No. of samples examined No. of samples positive Distribution (%)	22 14 63.63	20 15 75	25 16 64.0	23 15 65.21	30 22 73.33	120 82 68.33	
1	Chrysosporium indicum	3	5	5	3	7	23	19.16
2	Chrysosporium lucknowense	-	2	4	1	1	5	4.16
3	Chrysosporium pannicola	2	2) 1	2	2	9	7.5
4	Chrysosporium tropicum	1	3	1	3	4	12	10.0
5	Chrysosporium state of Ctenomyces serratus	-	2	-	-	2	4	3.33
6	Gymnascella dankailensis	1	-	-	1	1	3	2.5
7	Gymnoascus reessii	1	-	-	1	-	2	1.66
8	Microsporum gypseum	5	1	4	2	3	15	12.5
9	Trichophyton mentagrophytes	\mathbf{O}_1	-	-	1	-	2	1.66
10	Trichophyton terrestre	-	-	4	1	2	7	5.83
	Total	14	15	16	15	22	82	68.33

Table 2. Distribution of keratinophilic fungi in relation to different soil pH.

	soil pH									
	7.50	8.00	8.50	9.00	9.50	10.00	10.50	11.00	Total	%
Total No. of sample examined	18	32	25	17	12	7	5	4	120	
Positive soil samples	17 94.44	29 90.62	17 68.00	10 58.82	6 50.00	1 14.28	2 40.00	- 00.00	82 68.33	
Chrysosporium indicum	6	7	5	3	2	-	-	-	23	19.16
Chrysosporium lucknowense	1	2	1	1	-	-	-	-	5	4.16
Chrysosporium pannicola	2	3	3	1	-	-	-	-	9	7.5
Chrysosporium tropicum	3	4	2	2	1	-	-	-	12	10.00
Chrysosporium state of Ctenomyces serratus	2	2	-	-	-	-	-	-	4	3.33
Gymnascella dankailensis	-	1	1	1	-	-	-	-	3	2.5
Gymnoascus reessii	-	2	-	-	-	-	-	-	2	1.66
Microsporum gypseum	2	5	4	1	1	1	1	-	15	12.5
Trichophyton mentagrophytes	-	1	-	-	1	-	-	-	2	1.66
Trichophyton terrestre	1	2	1	1	1	-	1	-	7	5.83
Total No.	17	29	17	10	6	1	2	-	82	68.33



maximum numbers of fungi were recorded in the range of pH 7.5 to 8.5. Only few fungi viz. *Trichophyton terrestre, Microsporum gypseum* and *Chrysosporium indicum* were recorded at pH 9 and above. This indicates that these fungi are able to survive as resting spore in the soil under pH 9 or above and germinate when suitable condition arises.

Thus, the results of this study points to an influence of the alkaline pH (7.5-9.00) on the growth of most of the keratinophilic fungi. It is also recorded that the keratinophilic fungi grow much better in alkaline pH.⁴²⁻⁴⁴ However, the growth of some keratinophilic fungi in acid pH was also observed in other studies.⁴⁵

Conclusions

In our study it can be concluded that keratinophilic fungi occurs in the *Usar* soils of Uttar Pradesh and are surviving in harsh conditions like high pH and radiation.

These fungi may be playing an important role in decomposing the keratinic mater added to these uncultivated lands on one hand and may be a source of infection to the people visiting these areas. Garg¹⁰ also emphasized that the climate and perhaps other environmental factors are apparently important in determining the distribution of keratinophilic fungi.

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