

The occurrence of dermatophytes and other keratinophilic fungi from the soils of Himachal Pradesh (India)

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Results of a preliminary study of keratinophilic fungi from soils collected from a variety of sites in hilly areas of Himachal Pradesh, India, are reported. A total of 122 soil samples were examined, of which 101 samples were positive for keratinophilic fungi and related dermatophytes. Eleven species of five genera were isolated viz. *Chrysosporium indicum* (11 %), *C. keratinophilum* (4 %), *C. queenslandicum* (25 %), *C. tropicum* (19 %), *Chrysosporium* sp. (2 %), *C. xerophilum* (2 %), *Chrysosporium* state of *Ctenomyces serratus* (2 %), *Malbranchea gypsea* (7 %), *Microsporium canis* (2 %), *Microsporium gypseum* complex (9 %) and *Trichophyton mentagrophytes* (1 %).

Key words: Himachal Pradesh, hilly areas, soil fungi, keratinophilic fungi, India

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Článek shrnuje předběžné výsledky studia keratinofilních hub, sbíraných na různých místech v horských oblastech indického státu Himachal Pradesh. Celkem bylo studováno 122 půdních vzorků, z nichž 101 obsahovalo keratinofilní houby a příbuzné dermatofyty. Bylo izolováno 11 druhů z 5 rodů, a sice *Chrysosporium indicum* (11 %), *C. keratinophilum* (4 %), *C. queenslandicum* (25 %), *C. tropicum* (19 %), *Chrysosporium* sp. (2 %), *C. xerophilum* (2 %), anamorfa typu *Chrysosporium* od druhu *Ctenomyces serratus* (2 %), *Malbranchea gypsea* (7 %), *Microsporium canis* (2 %), *Microsporium gypseum* (9 %) a *Trichophyton mentagrophytes* (1 %).

INTRODUCTION

Keratinophilic fungi are present in the environment with variable distribution patterns that depend on different factors, one of which, of fundamental importance, is human and/or animal presence. Keratinophilic fungi play an important role in decomposing keratin. These fungi were isolated from Indian soils by various investigators from various habitats viz. public parks and soils or floor dust of primary schools (Ramesh and Hilda 1998-99), lake side soils (Ghosh and Bhatt 2000), house dust (Nigam and Kushwaha 1989), water sediments (Katiyar and Kushwaha 2000), a glacier bank (Deshmukh 2002), salt pans (Deshmukh 2004),

and birds and their environment (Sur and Ghosh 1980). However, hilly areas have attracted less attention (Deshmukh 1985, 2002; Bhadauria and Kushwaha 2003) and data on the distribution of keratinophilic fungi in such areas are therefore scanty. The present investigation reports the isolation of these fungi from soils of hilly areas in Himachal Pradesh, a Himalayan state. It is geographically diverse and the cold climate makes it a potentially interesting area to study the distribution of these fungi as it provides an unusual natural environment. There is great diversity in the climatic conditions of Himachal Pradesh due to variation in elevation (450–6500 m), rainfall (50–3400 mm) and temperature in winter (–3 °C to 14 °C) and summer (12 °C to 25 °C).

MATERIALS AND METHODS

One hundred and twenty-two samples were collected from 8 regions and consisted of 14 sites in the Northwestern state of Himachal Pradesh from May 2000 – July 2001 (Tab. 1, Fig. 1, 2). These regions were hilly areas such as around Shimla, Manali, Mandi, Chamba and Dharamshala. The approximate height of these region was 800–3500 m. The other regions were in the valleys such as Kullu, Pin Valley and Daranghati. The samples were collected from the superficial layer, depth not exceeding 3–5 cm, with a plastic spoon and transferred to sterile polythene bags, brought to the laboratory, stored at 15 °C and processed within two weeks time.

In the soils of the Himalayan area a lot of heterogeneity exists (Tab. 2). The garden soil is alluvial (pH 6.5–7.5), the soil of hilly slopes is brown in colour (pH 7.0–8.5), and the soil in the forest region has a greyish white colour in its upper leached layer and is acidic in nature (pH 5.5–6.5). Himachal Pradesh is a very popular hill station visited by large number of tourists.

The hair bait technique of Vanbreuseghem (1952) was used to isolate keratinophilic fungi. For this purpose, sterile Petri dishes were half filled with the soil samples and moistened with water and baited with burying sterile human hair in the soil. These dishes were incubated at room temperature (20 ± 1 °C) and examined for fungal growth over a period of four weeks. After observing the growth under a stereoscopic binocular microscope it was 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 Domsch et al. (1980), Oorschot (1980), Sigler and Carmichael (1976), Currah (1985), Cano and Guarro (1990), von Arx (1986).

Tab. 1. Distribution of soil samples.

Site	Height in metres	Number of samples examined	Number of samples positive
Shimla	2,195	14	14
Manali	1,915	13	10
Pin Valley National Park	3,500	29	19
Kullu	1,220	12	11
Mandi	800	18	16
Chamba	1,676	16	12
Dharamshala	1,830	10	10
Daranghati Sanctuary	2,195	10	9
Total		122	101

RESULTS AND DISCUSSION

The results of the isolations are presented in Tab. 2. From 122 samples 101 yielded keratinophilic fungi. A total of five genera with eleven species were isolated. *Chrysosporium queenslandicum* was the most frequently isolated species (25 %). *Chrysosporium tropicum* (19 %) and *Chrysosporium indicum* (11 %) were the next frequently isolated species. The other species isolated were *Chrysosporium keratinophilum* (4 %) and *Chrysosporium xerophilum* (2 %). Two isolates of *Chrysosporium* were obtained but could not be identified. Their systematic positions are yet to be confirmed. *Chrysosporium* species were also earlier reported from Indian soils (Randhawa and Sandhu 1965, Kushwaha and Agarwal 1976, Deshmukh and Agrawal 1983, Deshmukh 2004). *Chrysosporium indicum* was reported as the most dominant species (Deshmukh and Agrawal 1998, 2004).

The *Microsporium gypseum* complex had showed a frequency of 9 %. We also encountered *Nannizzia* sp. three times in association with the *Microsporium gypseum* complex, which was made up of the two teleomorph states *Nannizzia incurvata* and *N. gypsea*, but *N. fulva* was altogether absent. However, mating experiments were not conducted to confirm its specific identity. Other dermatophytes isolated were *Microsporium canis* (3 %), and *Trichophyton mentagrophytes* (1 %). These dermatophytes were also reported from Indian soils by various workers (Randhawa and Sandhu 1965, Garg 1966, Deshmukh and Agrawal 1983, Deshmukh 2004).

The keratinophilic species of *Malbranchea*, i.e. *Malbranchea gypsea* had a frequency of 1 %. Also various other workers have recorded some species of this genus from Indian soils in their study while investigating keratinophilic fungi (Kushwaha and Agrawal 1976, Deshmukh and Agrawal 1983, Singh et al. 1994).

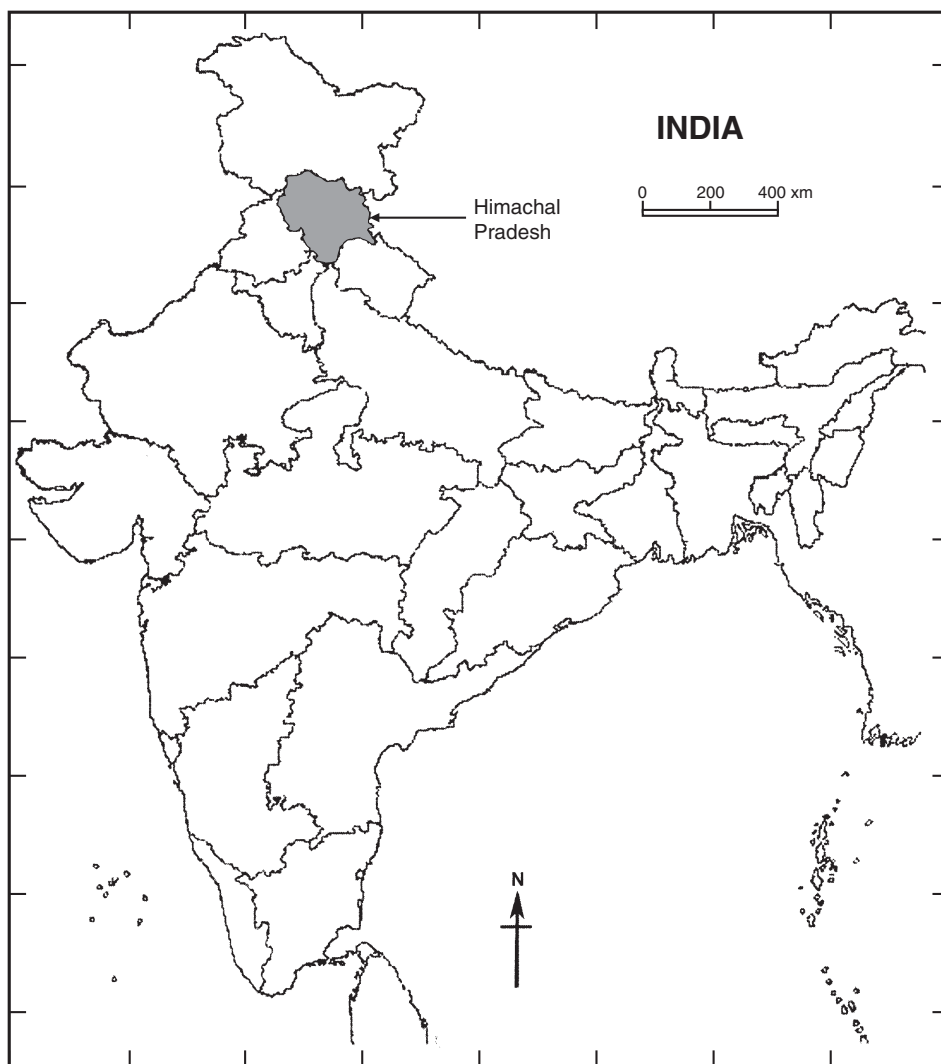


Fig. 1. Location of the State of Himachal Pradesh in India.

Fungi isolated from parks and play-grounds were *Chrysosporium indicum*, *C. keratinophilum*, *C. queenslandicum*, *C. tropicum*, *Malbranchea gypsea*, *Microsporium canis* and *M. gypseum* complex. *Trichophyton mentagrophytes* was exclusively isolated from parks. The parks and play-grounds in these areas are visited by tourists and occurrence of dermatophytes in these soils can be a threat to humans. A large number of keratinophilic fungi have been isolated from public parks and play grounds (Ramesh and Hilda 1998-99, Ali-Shtayeh



Fig. 2. Sites of collection of soil samples from Himachal Pradesh, India.

1989). In India, open school playgrounds, public parks and public places are often invaded by animals such as cows, bullocks, dogs, pigs, cats, and rats. These transit animals leave organic residues, which may contaminate the soil with keratinous debris and can be reservoirs for these fungi.

It is interesting to note that the maximum number of fungi was recorded from Daranghati sanctuary. *Chrysosporium indicum*, *C. queenslandicum*, *C. xerophilum*, *Malbranchea gypsea*, *Microsporium canis* and *M. gypseum* complex were isolated from this bird and animal sanctuary. Occurrence of these fungi in these areas may be due to the addition of feathers and keratinous material from birds and animals. The half-decomposed feathers along with plant litter which were collected showed that they can be a reservoir for these fungi.

Tab. 2. Distribution of keratinophilic fungi in different types of soil.

Regions	Shimla		Manali		Dharamshala		Mandi		Chamba		Kullu		Pin Valley National Park		Daranghati	Total	Frequency %
	Garden	Play ground	Garden	Play ground	Play ground	Pastures	Crop fields	Glacier sediments	Crop fields	Glacier sediments	Play ground	Garden	Pin Valley National Park	Pastures	Sanctuary		
Source of soil samples																	
Number of samples examined	10	4	13	7	3	3	13	5	4	12	8	4	17	12	10	122	
Number of samples positive	7	4	12	6	2	2	11	4	3	10	5	4	13	11	9	101	
Frequency %	70	100	92	86	67	67	85	80	75	83	63	100	76	92	90	83	
Fungi recorded																	
<i>Chryso sporium indicum</i>	1	-	2	-	-	-	1	-	1	1	2	1	-	2	2	13	11
<i>Chryso sporium keratinophilum</i>	-	1	1	-	-	-	-	-	-	1	-	-	-	1	1	5	4
<i>Chryso sporium queenslandicum</i>	2	1	1	3	2	2	2	1	1	5	1	1	5	2	3	30	25
<i>Chryso sporium</i> sp.	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2	2	2
<i>Chryso sporium tropicum</i>	2	2	2	-	-	-	4	1	1	2	2	1	-	6	-	23	19
<i>Chryso sporium cerophilum</i>	-	-	-	-	-	-	-	-	-	-	-	-	2	-	-	2	2
<i>Chryso sporium</i> state of <i>Ctenomyces serratus</i>	-	-	-	-	-	3	-	-	-	-	-	-	-	-	-	3	2
<i>Malbranchea gypsea</i>	1	-	1	-	-	-	1	1	-	1	-	-	2	-	1	8	7
<i>Microsporium canis</i>	-	-	1	1	-	-	-	-	-	-	-	1	-	-	-	3	2
<i>Microsporium gypseum</i> complex	1	-	3	2	-	-	-	1	-	-	-	-	4	-	-	11	9
<i>Trichophyton mentagrophytes</i>	-	-	1	-	-	-	-	-	-	-	-	-	-	-	-	1	1

The Pin Valley National Park yielded *C. queenslandicum*, *C. xerophilum* and *Malbranchea gypsea*. It is situated in the cold desert of Spiti Valley and is famous for large herds of ibex, bharal and snow leopard (*Capra ibex*, *Uncia uncia*, *Pseudois nayaur*). It seems that these fungi have adapted to the colder climate of this area and are associated with the animals and limited human presence.

Crop field soils were dominated by various keratinophilic fungi, which indicates that these fungi are adapted to crop field. *Ctenomyces serratus* was exclusively isolated from crop field soil along with *C. indicum*, *C. queenslandicum* and *Malbranchea gypsea*.

Chrysosporium indicum, *C. queenslandicum*, *C. tropicum* and *C. keratinophilum* were isolated from pastures. In the pastures, sheep and other domestic animals graze and shepherds spin the wool. The fungi isolated from glacier sediments were *C. indicum*, *C. queenslandicum*, *C. tropicum*, *C. keratinophilum*, *Malbranchea gypsea* and *Microsporium gypseum*. These fungi were also isolated from water sediments from Kanpur by Katiyar and Kushwaha (2000). The occurrence of these fungi in the sediments of glaciers makes this study more interesting. These fungi were also reported from glacier banks soils of Kashmir (Deshmukh 2002) and from the Antarctic environment (Mercantini et al. 1993, Caretta and Piontelli 2004).

In this paper we have presented the results of a study on the occurrence of keratinophilic fungi in Himachal Pradesh. Whether significant differences exist with those occurring in other climate zones of India cannot be concluded from this study and will need additional extensive investigations.

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REFERENCES

- ALI-SHTAYEH M. S. (1989): Keratinophilic fungi of school playgrounds in the Nablus area, West Bank of Jordan. – *Mycopathologia* 106(2): 103–108.
- ARX J. A. VON (1986): The ascomycetes genus *Gymnoascus*. – *Persoonia* 13: 173–183.
- BHADOURIA D. and KUSHWAHA R. K. S. (2003): Keratinophilic fungi from soils of hills and their keratinolytic activity. – In: Rao G. P., Manoharachari C., Bhat D.J., Rajak R.C., Lakhnupal T.N. (eds.), *Frontiers of fungal diversity in India* (Prof. Kamal Festschrift). Lucknow, India, p. 251–269.
- CANO J. and GUARRO J. (1990): The genus *Aphanoascus*. – *Mycol. Res.* 94: 355–377.
- CARETTA G. and PIONTELLI E. (2004) : Keratinophilous fungi from Antarctic terrestrial habitats. – In: Kushwaha R.K.S. (ed.), *Fungi in human and animal health*. Jodhpur, India, p. 29–38.
- CURRAH R. S. (1985): Taxonomy of *Onygenales: Arthrodermataceae, Gymnoasceae, Myxotrichaceae* and *Onygenaceae*. – *Mycotaxon* 24: 1–216.

- DESHMUKH S. K. (1985): Isolation of dermatophytes and other keratinophilic fungi from soil of Mussoorie (India). – *Mykosen* 28(2): 98–101.
- DESHMUKH S. K. (2002): Incidence of dermatophytes and other keratinophilic fungi in the glacier bank soils of Kashmir valley (India). – *Mycologist* 16(4): 165–167.
- DESHMUKH S. K. (2004): Isolation of dermatophytes and other keratinophilic fungi from the vicinity of salt pan soils of Mumbai (India). – *Mycopathologia* 157(3): 265–267.
- DESHMUKH S. K. and AGRAWAL S. C. (1983): Prevalence of dermatophytes and other keratinophilic fungi in soils of Madhya Pradesh (India). – *Mykosen* 26(11): 574–577.
- DESHMUKH S. K. and AGRAWAL S. C. (1998): Biology of keratinophilic fungi and related dermatophytes. – In: Varma A. (ed.), *Microbes: for health, wealth and sustainable environment*, New Delhi, India, p. 253–272.
- DESHMUKH S. K. and AGRAWAL S. C. (2004): Dermatophytes and keratinophilic fungi and their secondary metabolites. – In: Kushwaha R.K.S. (ed.), *Fungi in human and animal health*. Jodhpur, India, p. 85–111.
- DOMSCH K. H., GAMS W. and ANDERSON T.-H. (1980): *Compendium of soil fungi*. – 859 p. London.
- GARG A. K. (1966): Isolation of dermatophytes and other keratinophilic fungi from soils in India. – *Sabouraudia* 4: 259–264.
- GHOSH G. R. and BHATT S. (2000): Keratinophilic fungi from Chilka Lake-side soil Orissa (India). – *Ind. J. Microbiol.* 40: 247–254
- KATIYAR S. and KUSHWAHA R. K. S. (2000): Human hair colonizing fungi in water sediments of India. – *Mycopathologia* 152(2): 81–84.
- KUSHWAHA R. K. S. and AGRAWAL S. C. (1976): Some keratinophilic fungi and related dermatophytes from soils. – *Proc. Indian Natn. Sci. Acad.* 42 (B): 102–110.
- MERCANTINI R., MARSELLA R., MORETTO D. and FINOTTI E. (1993): Keratinophilic fungi in the Antarctic environment. – *Mycopathologia* 122: 169–175.
- NIGAM N. and KUSHWAHA R. K. S. (1989): Some new reports on keratinophilic fungi. – *Current Science* 58: 1374.
- OORSCHOT C. A. N. van (1980): A revision of *Chrysosporium* and allied genera. – *Studies in Mycology* 20: 1–89.
- RAMESH V. M. and HILDA A. (1998–99): Incidence of keratinophilic fungi in the soil of primary schools and public parks of Madras City, India. – *Mycopathologia* 143: 139–145.
- RANDHAWA H. S. and SANDHU R. S. (1965) : A survey of soil inhabiting dermatophytes and related keratinophilic fungi of India. – *Sabouraudia* 4: 71–79.
- SIGLER L. and CARMICHAEL J. W. (1976): Taxonomy of *Malbranchea* and some other hyphomycetes with arthroconidia. – *Mycotaxon* 4: 349–488.
- SINGH C. J., SINGH B. G. and SINGH B. S. (1994): Keratinophilic fungi of Ghana birds Sanctuary Bharatpur (Rajasthan). – *Ad. Plant Sci.* 7: 280–291.
- SUR B. and GHOSH G. R. (1980): Keratinophilic fungi from Orissa India I. Isolation from soils. – *Sabouraudia* 18: 269–274.
- VANBREUSEGHEM R. (1952): Technique biologique pour l'isolement des dermatophytes du sol. – *Ann. Soc. Belge. Med. Trop.* 32: 173–178.