J. Indian bot. Soc
Vol. 86 (3 & 4) 2007 : 10-13

COMPARATIVE INCIDENCE OF KERATINOFERS BETWEEN AGRICULTURAL FIELDS AND WASTELANDS

SHALINI UPADHYAYA, RICHA SHARMA & *B.S. SINGH

Department of Botany, St. John's College, Agra-282002.
**Head, Department of Botany, St. John's College, Agra-282002.*

During the present study, soil samples of six each agricultural fields and wastelands in and around Agra city were analyzed for the presence of keratinofers through bait-technique using peacock feathers, pigeon feathers, buffalo hair and human hair for direct isolation from soil medium kept at room temperature. Further, their percent frequency is determined and compared. A total of 15 genera and 30 species were found to inhabit both agricultural fields and wastelands. Maximum 210 of fungal encounters were observed in agricultural fields and 168 in that of wastelands. *Chrysosporium keratinophilum* predominated with maximum 8.10 % frequency in agricultural fields while *C. indicum* showed 8.93 % frequency in wasteland soils respectively. Variable percent frequency of fungal isolates occurs between agricultural fields and wastelands. Their occurrence in agricultural fields indicate pollution due to heavy manuring and fertilizers.

Key words: Keratinofers, agricultural fields, wastelands and pollution indicators.

Keratinofers occur in many natural and man made habitats and exhibit affinity to keratinous substrates while degrading them in nature. Ecological mapping of keratinofers in different unusual habitats has been carried on in the past (Srivastava,2002). In the soil environment, the factors influencing keratinolytic fungi have been relatively well recognized (Vollekova,1992). However, little information is available on the occurrence of these fungi in waste contaminated habitats. Studies on the isolation of keratinofers from agricultural fields where human interference and water-logging are encountered, is meagre (Ulfig,2000). Abundance of these microbes in the habitats with which keratin remnants of human and animal origin is well expected. Since these fungi display potentially pathogenic properties, studies on their presence in agricultural environment and wastelands become epidemiological significant. Indoor and outdoor dust within urban agglomerations has been surveyed for keratinolytic fungi and related species several times. (Mercantini *et al.*,1989). Since, Agra is highly populated and industrialized city, high organic and inorganic contamination considerably created impact on microbial communities including those of keratinolytic fungi.

The present study is therefore undertaken to review and assess keratinofers associated with agricultural fields and wastelands in and around Agra city. Besides this, a comparision is drawn between agricultural fields and wastelands in relation to their occurrence and incidence.

MATERIALS AND METHODS

Six agricultural fields and six wastelands in and around Dayalbagh, Sikandra, Tehra, Balkeshwar, Runakta and Kuberpur of Agra city were being surveyed. Animal or human visits to these places were also being noticed. 10 soil samples each of 500gm of depth 5" from these places were collected in presterilized polythene bags, which were brought to the laboratory for testing the presence of keratinofers. Half of the presterilized separate sets of Petriplates were filled with various samples. Four baits i.e.buffalo hair, pigeon feathers, peacock feathers and human hair were collected, cut into small pieces, washed with distilled water several times and with chloroform and washed with distilled water again and dried in an oven. The pieces of these baits were spread over the moistened soil samples present in the Petriplates under aseptic conditions.

Table-1: Percent frequency of fungal isolates from Agricultural fields and wastelands (A Comparative Account)

S. No.	Name of Fungus	Agricultural Fields		Wastelands	
		No. of ENC.	% Frquency	No. of ENC.	% Frequency
1	*Absidia glauca*	5	2.38	5	2.98
2	*Alternaria alternata*	4	1.90	2	1.19
3	*Alternaria tenuis*	7	3.33	6	3.57
4	*Aspergillus flavus*	7	3.33	3	1.79
5	*Aspergillus fumigatus*	6	2.86	4	2.38
6	*Aspergillus nidulans*	7	3.33	5	2.98
7	*Aspergillus niger*	6	2.86	4	2.38
8	*Aspergillus ustus*	6	2.86	10	5.95
9	*Candida albicans*	10	4.76	3	1.79
10	*Chrysosporium indicum*	6	2.86	15	8.93
11	*Chrysosporium keratinophilum*	17	8.10	4	2.38
12	*Chrysosporium tropicum*	5	2.38	11	6.55
13	*Fusarium oxysporum*	12	5.71	5	2.98
14	*Fusarium semitectum*	8	3.81	7	4.17
15	*Fusarium solani*	8	3.81	6	3.57
16	*Geomyces sp.*	6	2.86	8	4.76
17	*Geotrichum candidum*	7	3.33	7	4.17
18	*Helminthosporium sp.*	8	3.81	8	4.76
19	*Malbranchea auriantiaca*	7	3.33	9	5.36
20	*Malbranchea fulva*	8	3.81	5	2.98
21	*Mucor mucedo*	7	3.33	6	3.57
22	*Penicillium species*	6	2.86	5	2.98
23	*Rhizopus globosus*	6	2.86	4	2.38
24	*Rhizopus nigricans*	3	1.43	4	2.38
25	*Rhizopus nodosus*	5	2.38	3	1.79
26	*Trichophyton mentagrophytes*	8	3.81	3	1.79
27	*Trichophyton rubrum*	4	1.90	2	1.19
28	*Trichophyton simii*	7	3.33	4	2.38
29	*Trichophyton terrestre*	7	3.33	4	2.38
30	*Verticillium species*	7	3.33	6	3.57
	TOTAL ENCOUNTERS	210		168	

Distilled water was spread periodically to maintain moist conditions. The Petriplates were left at room temperature for about 35 days undisturbed and fungal growth was observed, and the results were recorded. After ensuring optimum fungal growth, the mycelial fragments from the margins of the fungal colonies were picked up by a sterilized inoculation needle and were transferred aseptically into presterilized Petriplates, containing Sabouraud's Dextrose Agar medium.

It was sterilized in autoclave at 15 lbs pressure , for 15 – 20 minutes and pH was adjusted to 6.7-7.00 for obtaining pure cultures, which were maintained on Sabouraud's Dextrose Agar slants. After ensuring a fairly good growth, they were preserved in refrigerators. The cultures were periodically transferred into fresh tubes to maintain their purity and to avoid pleomorphy, through repeated culturing. On the basis of macro and micromorphological characters, the isolates were assigned to their systematic position, with the help of standard references and manuals.

RESULTS AND DISCUSSION

All the soil samples investigated were found to be positive for fungal activity. The soils of agricultural fields showed maximum of 210 encounters in comparison to 168 fungal encounters

observed in that of wastelands. A total of 15 genera and 30 species were isolated through various baits in both the types of soils. Among which 3 species of *Chrysosporium*, 2 species of *Malbranchea* and 4 species of *Trichophyton* were present which are known for their pathogenicity. Maximum 8.10% frequency of *C. keratinophilum* in agricultural fields and 8.93% frequency of *C. indicum* in that of wastelands is being observed. While the former i.e. *C. keratinophilum* showed decreased percent frequency of 2.38 in wastelands and the latter i.e. *C. indicum* showed decreased percent frequency of 2.86 in agricultural fields on comparison (Table-1). The percent frequency of each fungus was calculated by the formula given below:-

Percent frequency = No. of encounters of individual fungus* 100/ Total encounter of all the fungi.

Percent increase in the frequency of isolates in wasteland environment when compared to agricultural fields is observed in ; *Absidia glauca, Alternaria tenuis,Aspergillus austus, C. indicum, C. tropicum, Fusarium semitectum, Geomyces sp., Geotrichum candidum, Helminthosporium sp., Malbranchea auriantiaca, Mucor mucedo, Penicillium sp., Rhizopus nigricans, Trichophyton rubrum, Verticillium* sp.This indicated their tolerance towards solid waste pollution which contaminated the wastelands. On the other hand, fungal isolates such as *Alternaria alternata, Aspergillus flavus, Aspergillus fumigatus, Aspergillus nidulans, Aspergillus niger, Candida albicans, C. keratinophilum, Fusarium oxysporum, Fusarium solani, Malbranchea fulva, Rhizopus globosus, Rhizopus nodosus, Trichophyton mentagrophytes, Trichophyton simii, Trichophyton terrestre* showed increased percent frequency in agricultural fields in comparison to the wastelands environment.These fungi seemed to play indicators of pollution affecting agricultural fields (Jain,2003).

However, frequent occurrence of *Chrysosporium* species in solid waste polluted soils has been reported by Srivastava (2002). Ulfig,(1992) found the association of *C. keratinophilum, Malbranchea auriantiaca, Malbranchea fulva*, with sewage sludge without any treatment being used in agricultural fields as manures. The presence of these fungi in agricultural fields pose health risk problems particularly in India, where 75% of people are involved in agricultural farming (Ulfig,1986). Thus, the present study confirmed earlier reports. Ulfig *et al.* (1996), recognized the incidence of pathogenic keratinofers under solid waste polluted conditions which could be used as biological indicators of waste contaminated soils (and it can not be ignored from an epidemiological point of view)(Kornillowicz and Kowalska,1997). Prevalence of *C. indicum* in waste contaminated soil showed its tolerance towards heavy metals, ions concentrations as well as different pollutants has been determined earlier (Srivastava,2002, Jain, 2003) which is in agreement with the present findings.

REFERENCES

Jain A 2003 *Biodeterioration of keratinic wastes by certain keratinophilic fungi and related dermatophytes*. Ph D Thesis Dr B R A University Agra.

Kornillowicz-Kowalska T 1977 Studies on the decomposition of keratin wastes by saprophytic microfungi.I.Criterion for evaluating keratinolytic activity. *Acta Mycologia* **32**(11) 51-79.

Mercantini R Marsella R Prignano G Moretto D Marmo W Leonatto F Fuga G C & Serio G 1989 Isolation of keratinophilic fungi from the dust of ferry boats and Italy. *Mycoses* **32** 590-594.

Srivastava P 2002 *Ecological studies on keratinofers from unusual habitats in and around Agra*. Ph D Thesis Dr B R A University Agra

Ulfig K 1986 Preliminary studies on the growth and survival of selected pathogenic dermatophytes sediments and sewage sludge. (in Polish) *Rozen PZH* **3** 335-340.

Ulfig K 1992 Selected group of geophilic fungi in sediments of Catalonian waters as microorganisms

of ecological and public health significance. Madrid Ministerio de Education Liencia.

Ulfig K Terrakowski G Plaza O & Kasarewoiz T 1996 Keratinophilic fungi in sewage sludge. *Mycopathologia* **136** 41-46.

Ulfig K 2000 The occurrence of keratinophilic fungi in waste and waste contaminated habitats.Revista Iberoamericana De Mycoligia Apdo 699 E48080 Bilbao Spain 1-30.

Vollekova A 1992 Keratinofilne huby v styroch lesnych podach. (Czech) Biologia (Bratislava) 47 477-482.