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Fungal Flora Isolated from Deteriorating Card Board and Corrugated Box Samples in Thane and Bhiwandi, India

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A B S T R A C T

Many cellulose based articles have found their way into human requirements and have become articles of necessity, paper being one such article. Card board and corrugated boxes are used as packing materials universally and alike paper, these are vulnerable to invasion by cellulose degrading fungi. The study in regions of Thane and Bhiwandi cities on the outskirts of Mumbai city in Western coastal part of Maharashtra, India focused on the invasion of cardboard and corrugated paper boxes by fungi. Twenty four species of fungi belonging to 11 genera were found on the deteriorating card board and corrugated paper samples during the study, prominent amongst them being the genus *Aspergillus*, *Chaetomium*, *Curvularia*, *Trichoderma*, *Penicillium*, *Alternaria* and five other cellulolytic fungal genera. Growth of fungi resulted in crumbling of the cardboard at places due to loss of strength. Many of the fungal forms isolated were potential health hazards which was cause for concern.

Introduction

Fungi have received attention in recent years, especially after fungal infections came into limelight during COVID-19 and post COVID-19. Although plenty of studies and surveys have been carried out the true extent of fungal diversity is still not fully known and is expected to be much more than what is known today (Hawksworth, 2001). Apart from taxonomic placement, fungi are also divided into functional groups and one such group with social and environmental relevance is cellulose degrading fungi. Several cellulose based articles having found their way into human civilizations have become articles of necessity. Paper is one such article. Card board and corrugated boxes, derivative

products of the paper industry, have found universal utility as an economical packaging material. Both these cellulosic materials, like paper, are vulnerable to invasion by this consistent group of cellulose degrading fungi, which bring about deterioration of cellulosic materials.

The present study was initiated to conduct a study of cellulose degrading fungi inhibiting cardboard and corrugated paper box samples from cartons used for packaging cutlery, printing papers and other utilities in Mumbai region of Western India. Mumbai is the commercial capital of India and the cities of Thane and Bhiwandi are situated on its outskirts. Both cities, like Mumbai are situated in coastal region of western part of Maharashtra state, India. While Thane is known as the 'lake city', Bhiwandi is famous as the 'Manchester of India' due to its thriving power loom industry. The entire region has a huge demand for paper, cardboard and corrugated packing materials. A survey of literature revealed scattered and scanty literature on fungal organisms invading card board in the areas of study which influenced the study.

Materials and Methods

Samples of cardboard and corrugated boxes showing few signs of deterioration were collected from Thane and Bhiwandi areas in Western parts of Maharashtra state from June-September 2020. The samples were as follows:

Sample No. 1. Cardboard cover of long book register stored in closed dark loft showing signs of dampness during monsoon.

Sample No. 2. Corrugated paper sample from corrugated box used for packaging and storing cutlery. The samples after collection were stored in sterilized polythene bags at room temperature for further processing. Both the samples showed few visible signs of deterioration. Serial dilution method (Pramer and Schmidt, 1966) was used for isolation of pure cultures of fungal organisms from the samples. Suspensions from the samples were diluted up to 10^{-5} and 1 ml each of the respective dilutions was plated on nutrient medium (Czapex Dox Agar, Czapex Dox Agar with cellulose, PDA) in separate petri plates. The plates were incubated at room temperature for expression of fungal growth. Streptomycin (50 mgl^{-1}) was added to suppress bacterial growth and contamination. The isolated fungi were identified using standard literature and the standard system of fungal classification.

Results and Discussion

The different fungal organisms isolated from the cardboard and corrugated paper box samples are depicted in Table 1. Twenty four species of fungi belonging to 11 genera were found on the paper samples during the study. The sample No. 1 viz. cardboard cover of long book register yielded the maximum number of fungal forms followed by the second sample.

The typically dark, humid conditions of storage in the loft, accompanied by dampness during monsoon apparently contributed to the high number of fungi encountered on this sample. Prominent among the isolates was the genus *Aspergillus* with 7 species, followed by *Chaetomium* and *Curvularia* with 3 species each; *Trichoderma*, *Penicillium* and *Alternaria* with 2 species each and the rest of the genera represented by a single species. *Aspergillus*, *Chaetomium*, *Alternaria* and *Memnoniella* were encountered on both the samples.

Table.1 Fungal forms isolated from the samples

No.	Fungal Organism	No. of Isolates	Isolated from sample		Total presence (No. of samples)
			1	2	
1	*Mucor sp.*	1	+		1
2	*Chaetomium globosum* Kunze	3	+	+	2
3	*Chaetomium olivaceum* Cooke & Ellis	2		+	1
4	*Chaetonium crispatum* Fuckel	1	+		1
5	*Trichoderma pseudokoeningii* Rifai	1	+		1
6	*Trichoderma viride* Pers.	1	+		1
7	*Aspergillus fumigatus* Fresenius	4	+	+	2
8	*Aspergillus nidulans* G Winter	1	+		1
9	*Aspergillus sydowii* Thom & Church	1	+		1
10	*Aspergillus flavus* Link	5	+	+	2
11	*Aspergillus terreus* Thom	3	+	+	2
12	*Aspergillus niger* van Tieghem	7	+	+	2
13	*Aspergillus tamari* Kita	4	+	+	2
14	*Penicillium citrinum* Thom, C	2		+	1
15	*Penicillium digitatum* Pers. (Sacc.)	1	+		1
16	*Paecilomyces varioti* Bainier	1	+		1
17	*Memnoliella echinata* (Rivolta) Galloway	2	+	+	2
18	*Cladosporium cladosporioides* (Fresen) G.A. de Vries	1	+		1
19	*Curvulata lunata* (Wakker) Boedjin	1	+		1
20	*Curvularia pallescens* Boedjin	1	+		1
21	*Curvularia brachyspora* (Boedjin)	1	+		1
22	*Alternaria alternata* (Fr.) Keissi	2	+	+	2
23	*Alternaria solani* Sorauer	1	+		1
24	Non sporulating mycelium	1	+		1

The findings are in agreement with those of Dawar *et al.*, 2015. The genus *Aspergillus* dominated the fungal flora which is a fairly common phenomenon (Das and Prasad, 1996). Growth of fungi resulted in symptoms such as crumbling of the cardboard at places due to loss of strength. This is in conformity with symptoms reported by Dhawan and Garg (1993). Many of the fungal forms isolated were potential health hazards by virtue of their capability to cause allergic reactions (Crameri *et al.*, 2014) or as pathogens (English 1980) which was ample cause for concern.

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