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RESEARCH ARTICLE

Grain storage: The role of fungi in biodeterioration

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SUMMARY

The role of fungi associated with cereal grains in storage, in bringing about their deterioration, has been a subject of great interest for long time. The annual estimated losses of the stored food grains are 10-12 million tones. With a view to study the concentration of food grain biodeteriorating fungi and their role in deterioration was studied by operating volumetric Tilak air sampler for a period of one year. All the trapped airborne fungal spores have been categorized under the group 'spore type' and pollen grains, insect parts, plant parts, hyphal fragments and unidentified spore types were categorized under artificially formed group 'other type'. In all 78 types of airborne components were trapped, of which 73 being fungal in origin. Deuteromycotina, the biggest toll (with 40 components) contributed the highest percentage 61.61 per cent to the total airspora, it was followed by Ascomycotina (24 components) 13.81 per cent, Zygomycotina (4 components) 11.09 per cent, other types (5 components) 7.52 per cent, Basidiomycotina (4 components) 5.18 per cent and Myxomycotina (1 component) 0.7 per cent. The data of daily indoor temperature, relative humidity, etc. was recorded regularly and correlated with concentration of biodeteriorating fungi.

Key Words : Food grains, Biodeterioration, Airborne fungal spores, Tilak air sampler

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A major problem of agricultural production is loss of grain during storage in the warehouse. Microorganisms, insects, and rodents contribute greatly to these post harvest losses. A commonly quoted estimate provided by the FAO for worldwide losses for all cereals, leguminous seeds, and oilseeds is 10 per cent (Janicki and Green, 1976). Christensen and Kaufmann (1974) stated that fungi are the major cause of spoilage in stored grains and seeds in the technologically advanced countries, because insects and rodents are effectively controlled. The Agricultural Stabilization and Conservation Service (1976) reported that mold deterioration of the containers occurred in 25 per cent of the shipments of Corn Soya Milk (CSM) under the Food for Peace Programme. This spoilage resulted in losses estimated at 3 per cent of the total amount of CSM shipped. Consequently, it is well-established that fungi destroy food and feed. However, the basic problem remains of implementing effective measures to reduce fungal losses. Christensen and Kaufmann (1969) reported that more than 150 species of fungi have been isolated from within seeds or kernels.

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Fungi are a major cause of deterioration of cereals, legumes and oilseeds. Concern over fungal invasion of these grains has intensified over the last 17 years because mycotoxins (toxic metabolites produced by filamentous fungi) are being detected in some commodities. Apparently, the harvesting of more and more grain at moisture levels too high for safe storage has contributed to the mycotoxin problem.

In addition to moisture, other factors such as temperature, O_2-CO_2 atmosphere, aeration, pH and grain condition interact to affect fungal growth. Fungal deterioration of grain is a dynamic ecological process which often involves a succession of microorganisms, the breakdown of organic matter to yield CO_2 and H_2O and the generation of heat. Nutrients are lost because of changes in carbohydrates, protein, lipids and vitamins. Germinability is lost and aesthetic changes occur which include discoloration, caking and abnormal odors. The present investigations has been carried out on microbial pollution of air inside the food-grains godown, its impact on deterioration of stored grains and effects of atmospheric parameters on their concentration. Numerous reports (Dorworth and Christensen, 1968; Qasem and Christensen, 1960;

Christensen, 1955; Armolik *et al.*, 1956; Papavizas Christensen, 1957; Christensen, 1962; Fields and King, 1962; Lopez and Christensen, 1967) confirm that invasion of seeds by storage fungi can result in loss of germinability.

MATERIAL AND METHODS

Exploration of airborne microbes in indoor atmosphere was conducted for a period of one year *i.e.* from 1st October 1989 to 30th September 1990 in a food grain godown of Food Corporation of India at Ahmednagar (M.S.) India using continuous Tilak airsampler (Tilak and Kulkarni, 1970). The sampler was installed with its orifice kept at a certain height of one meter above the ground level. The slide preparation and scanning was done as described by Tilak and Srinivasulu (1967). The identification was based on reference slide, fungal collection and visual observations. The gunny bags of jowar, wheat and rice were stored by the corporation for about six months to a year.

RESULTS AND DISCUSSION

In all 78 air borne components were trapped, of which 73 being fungal in origin, were identified up to generic level. Other five types were hyphal fragments, insect parts, plant parts, pollen grains and unidentified spore types.

Deuteromycotina, the biggest toll (with 40 components) contributed the highest percentage (61.61%) to the totoal airspora, it was followed by Ascomycotina (24 components) 13.88 per cent, Zygomycotina (4 components) 11.09 per cent, other types (5 components) 7.52 per cent, Basidiomycotina (4 components) 5.18 per cent and Myxomycotina (1 component) 0.7 per cent (Table 1).

Ever since the work of investigation started, airborne components like *Cladosporium, Alternaria, Aspergillus, Sporothrix, Penicillium, Nigrospora, Curvularia* occured significantly throughout the period of investigation. *Cladosporium, Curvularia, Haplosporella, Harknessia, Penicillium* etc. occured maximum in the month of August 1990 during rainy period. The spore types like *Epicoccum, Helminthosporium, Lacellinopsis, Nigrospora, Oidium, Pyricularia* etc. showed their maximum concentration in the month of March 1990 in the atmosphere of food grain godown of FCI. *Alternaria* and *Aspergillus* occured maximum in the months of April and May 1990. High incidence of *Chaetomella* and *Sporothrix* in the air was noticed in the month of December 1989, *Botrytis* occured maximum in the month of October 1989. All the aforesaid spore types, however, occured in the dry period of the year. During the period of investigation. regular incidence of the aforesaid spore types in the air was positively correlated with the occurrence and amount of precipitation, an adequate range of relative humidity, moderately low range of temperature.

The abundance of parasitic and saprophytic forms occured near the trapping site. In fact, the probable source for these spore types could be the availability of dead organic matter, plant parts, vegetation, the dry jowar and bajra fodder in huge quantity for cattles etc. close in vicinity outside the godown. Whereas inside the godown, spore types generally found settled on the storage gunny bags, stored grains' surface. During activities such as handling, cleaning, storing, aerosol spraying, the settled spore types got distributed and disperse inside the godown atmosphere. In addition, open shutters of doors and windows permit wind currents from outside and help in dispersal of the deposited bioparticles inside the godown atmosphere. The grain dust that flies around during harvesting and threshing operations is heavily loaded with fungal spores and is infact an occupational hazard for the workers, likely to cause respiratory ailments (Aher *et al.*, 2004).

Bispora, Botryodiplodia, Ceratophorum, Chlamydomyces, Dictyoarthrinium, Diplodia, Torula etc. also occured with an appreciable number in the air, while *Beltrania, Beltraniella, Corynospora, Deightoniella, Heterosporium, Papularia, Pestalotia, Phaeotrichoconis, Spondylocladiella, Trichothecium* etc. feeble prevalence in the air inside the godown during the period of investigation (Table 2).

The predominant toxic biopollutants of stored grains like *Aspergillus, Penicillium* along with other phytotoxic biopollutents (*Alternaria, Curvularia, Cladoporium, Helminthosporium* etc.) were found associated with cereal grains on the surface or inside the tissues either as saprophytes or parasites.

Some of the characteristic features have been exhibited by airspora during the period of investigation. These are as

Table 1 : Total airspora and percentage contribution of each spore group

Spore group/class	Total no. of spores/m3 of air	Percentage
Myxomycotina	2828	0.71
Phycomycotina	44086	11.09
Ascomycotina	55174	13.88
Basidiomycotina	20608	50.18
Deuteromycotina	244986	61.61
Other types	29904	7.52
Total	397586	99.99

follows:

An ambient air inside the warehouse was found with rich of biopollutents in all months. The maximum concentration (47,838/m^3 of air) of these biopollutents was encountered in the month of August followed by March 1990 (42,756/m^3 of air) and December 1989 (39,424/m^3 of air) etc. In fact, ever since the work started, the fungal spores were recorded more in number from October 1989 to March

Table 2 : Total concentration and percentage contribution of each spore type

Sr. No.	Spore type	Total concentration	% contribution
1.	*Cladosporium*	70168	28.64
2.	*Alternaria*	25802	10.53
3.	*Aspergillus*	25172	10.27
4.	*Sporothrix*	13104	5.34
5.	*Penicillium*	12586	5.13
6.	*Curvularia*	16690	4.77
7.	*Nigrospora*	9912	4.04
8.	*Epicoccum*	8582	3.50
9.	*Periconia*	8218	3.35
10.	*Helminthosporium*	6888	2.81
11.	*Pyricularia*	6664	2.72
12.	*Candida*	5554	2.26
13.	*Lacellinopsis*	4452	1.81
14.	*Cheatomella*	4382	1.78
15.	*Bispora*	4214	1.72
16.	*Oidium*	4088	1.66
17.	*Botrytis*	3990	1.62
18.	*Haplosporella*	3444	1.40
19.	*Memnoniella*	2800	1.14
20.	*Dictyosporium*	2016	0.82
21.	*Chlamydomyces*	1736	0.70
22.	*Ceratophorum*	1316	0.53
23.	*Cordana*	1078	0.44
24.	*Papularia*	1078	0.44
25.	*Botrydiplodia*	1022	0.41
26.	*Dictyoarthrinium*	980	0.40
27.	*Myrothecium*	980	0.40
28.	*Torula*	952	0.38
29.	*Didymosphaeria*	798	0.32
30.	*Harknessia*	770	0.31
31.	*Diplodia*	630	0.25
32.	*Pseudotorula*	602	0.24
33.	*Heterosporium*	350	0.14
34.	*Beltrania*	224	0.09
35.	*Beltraniella*	168	0.06
36.	*Trichothecium*	154	0.06
37.	*Corynospora*	112	0.04
38.	*Deightoniella*	112	0.04
39.	*Pestalotia*	84	0.03
40.	*Spondylocladiella*	70	0.02

1990. Then from April 1990 there was gradual decline in frequency of occurrence of the biopollutents in air up to July also in the month of September 1990. But in the month of August 1990 peak of the biopollutents concentration was observed. Similar results were also reported by Chakravarty (1981) and Tilak and Chakre (1977). High incidence of microflora during rainy period (*i.e.* in the month of August 1990) may be due to adequate quantity of rainfall, markedly high relative humidity, availability of suitable substrates inside and outside the warehouse environment. Apart from that the microbes associated with hay and cereal grains, eventually during manual activities might have become airborne not only in August 1990 but also in the earlier months like December 1989 and March 1990.

The fungal spore types responsible for biodegradation of grains and phytotoxic such as *Penicillium, Aspergillus, Alternaria, Helminthosporium, Cladosporium, Curvularia* etc. were found associated externally and internally with the cereal grains.

The rate of biodeterioration of stored grains is mainly dependent on temperature and humidity prevailing inside the warehouse.

Aspergillus, one of the important grain toxic biopollutents, occured throughout the investigation period and contributed 10.27 per cent to the total airspora of Deuteromycotina (Table 2). The highest peak concentration (3444/m^3 of air) was recorded during morning hours in the month of May 1990. The highest daily catch (1148/m^3 of air) was encountered on 17th May 1990, when there was record of 30^0 C temperature, 57 per cent relative humidity and no precipitation. This observation appeared to be unusual record in the airspora studies of this region. Chakravarty (1981) at Kolkatta, recorded high monthly concentration of *Aspergillus parasiticus* and *Aspergillus niger* in the airspora studies of two different sites.

Penicillium sp. contributed 5.13 per cent to the total airspora of Deuteromycotina. High frequency of occurence (11,214/ m^3 of air) was recorded in the month of August 1990 with highest daily mean concentration (1484/m^3 of air) on 9th August 1990, when there was record of 25.2 ^{0}C temperature, 69.5 per cent relative humidity and 62mm rainfall. However, the weather parameters favoured for the growth, sporulation and dispersion of the spores of *Penicillium* sp. Similar reports have been recorded by Tilak and Chakre (1977) and Chakravarty (1981). Wallace (1973) lists 26 species of *Aspergillus* and 66 species of *Penicillium* which have been isolated from stored grains. Though Penicillia spores occured in the months of August and September but the spores of Aspergilli occured throughout. However, Penicillia were growing on the damp walls of godown where the water percolation through the leaked roof during rainy period hence, high incidence of spores of Penicillia as seen during rainy days.

Cladosporium sp., a common phytotoxic biopollutant occured throughout the year, contributed 28.64 per cent to the total airspora of Deuteromycotina (Table 2). Their maximum incidence (12,768/m^3 of air) was observed in the month of August and highest daily catch (3654/m^3 of air) on 3rd August 1990, when there was prevalence of 25 ^{0}C temperature, 66 per cent relative humidity and 10 mm precipitation around the trappin site.

Alternaria sp. the phytotoxic biopollutant contributed 10.53 per cent to the total airspora of Deuteromycotina (Table 2). It occurred throughout the year. Its maximum monthly and daily incidence (3024/m^3 and 336/m^3 of air) was recorded in the month of November 1989 and on 3rd April 1990, respectively, when there was record of an average 24.9^0 C temperature and 32.5 per cent relative humidity.

Helminthosporium sp., a common phytotoxic microbe, occured throughout the year and contributed 2.81 per cent to the total airspora. Its maximum monthly and daily concentration (1106/m^3 and 126/m^3 of air) was recorded in the month of March 1990 and on 1st July 1990, respectively, when there was prevalence of an average 25.5^0 C temperature and 67 per cent relative humidity.

Curvularia sp. occured throughout the period of investigation. It contributed 4.77 per cent to the total airspora of Deuteromycotina. Its maximum monthly and daily concentration (1330/m3 and 238/m3 of air) was encountered in the month of August 1990 and on 7th August 1990, respectively, when there was a record of 26.2 ^{0}C temperature, 73 per cent relative humidity and 19 mm rainfall. Similar observations were also made by Christensen (1969), Tilak and Chakre (1977), Chakravarty (1981), Aher *et al.* (2004).

From the above observations, it became evident that low temperature associated with high humid conditions helped in increasing not only the percentage of grain toxic biopollutants but also the phytotoxic pollutants.

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