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STUDIES ON STORED GRAIN FUNGI

Part III.—Fungi from Cereals

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Thirty-eight species belonging to 15 genera were isolatde from *Zea mays* Linn. (Pakistan), *Zea mays* Linn. (American), *Hordeum vulgare* Linn., *Sorghum vulgare* Pers and *Pennisetum typhoideum* Rich stored in various godowns in Karachi. About 83% of the organisms belonged to Fungi Imperfecti while Ascomycetes (15%) and Phycomycetes (2%) accounted for the remaining fungi. Species of *Aspergillus* were most predominant among all the fungi recorded and among these species *A. flavus* was the most prevalent organism. In *Zea mays* (American) damage was most severe because of the high moisture contents and increased temperature which resulted in the formation of "hot spots" and extensive damage. Surface sterilization of grains with 1:1000 $HgCl_2$ reduced the number of fungi considerably.

The authors have already reported[1,2] the fungi isolated from wheat and rice and from oilseeds alongwith *Plantago ovata* stored in various godowns in Karachi. Considerable amount of work has been done on fungi inhabiting and infesting cereals alongwith effect of environmental factors, moisture content, temperature in storage etc. on the severity of deterioration of grains. Sinha and Wallace[3] studied fungi causing deterioration of wheat during storage. They observed an ecological succession of species of microorganisms, which often overlapped, in the following order: *Penicillium cyclopium*, *P. funiculosum*, *Aspergillus flavus* Link, *A. versicolor*, *Absidia* sp and *Streptomyces* sp. Lutey and Christensen[4] observed the influence of moisture content, temperature and length of storage upon survival of fungi in barley kernels. Machacek *el. al.*[5] determined the effect of high water content in stored wheat, oat, and barley seeds on susceptibility to invasion by moulds. Qasem and Christensen[6] carried out investigations on the influence of various factors on the deterioration of stored corn by fungi. Barron and Lichwardt[7] worked on estimation of fungi associated with deterioration of stored maize. Junejo and Malik[8] carried out investigations on the microflora associated with sorghum and have reported a number of fungi causing deterioration of the grains.

The present investigation is an effort to determine the fungi infesting and causing deterioration of *Zea mays* Linn., *Hordeum vulgare* Linn., *Sorghum vulgare* Pers. and *Pennisetum typhoideum* Rich in storage.

Materials and Method

The stored grains were collected by random sampling method from the godowns in Karachi at different times of the year during 1969. The grains were stored in jute bags mostly placed on wooden platforms. The grain which were sampled for presence of fungi were *Zea mays* Linn. (Pakistani), *Zea mays* Linn. (American), *Hordeum vulgare* *Sorghum vulgare* Pers. and *Pennisetum typhoideum* Rich.

The temperature, outside and inside the bags of grain, was also determined. Air oven method[9] was applied for determining the moisture contents of the grains. For details of the methods reference may be made to our earlier communications.[1,2]

Results

A total number of 38 species belonging to 15 genera were isolated from the four cereals under investigation. Some organisms were common among all the cereals while some of them were isolated from only one or two cereals. Species of *Aspergillus* formed the nucleus of fungi infesting the cereals in godowns (Fig. 1).

There were two kinds of maize stored in godowns, one grown in Pakistan, the other imported from U.S.A. The one imported from U.S.A. was stored in godowns and some of them had hot spots (a condition where the temperature inside the heap of grains becomes high due to activities of microorganisms and high moisture contents which result in much more deterioration of the grain).

Table 1 lists the fungi isolated from the four cereals under investigation.

Investigations were also carried out to observe the effect of surface sterilization on the grains with respect to emergence of different organisms from them. The grains were surface sterilized with 1:1000 $HgCl_2$ for different lengths of time. After 10-sec exposure the number of organisms considerably reduced and only three species of fungi from each cereal, with the exception of *Sorghum vulgare* where only two organisms appeared, were isolated. At 20 sec, with the exception of *Hordeum vulgare* where three organisms emerged, the rest of the cereals yielded only 2 species each. When the grains were sterilized for 40 sec, no organism

appeared from *Sorghum vulgare*, two organisms each were isolated from *Hordeum vulgare*, *Zea mays* (American) and *Zea mays* (American burned) while only one fungus each emerged from *Pennisetum typhoideum* and *Zea mays* (Pakistani). At 60 sec no organism appeared from *Pennisetum typhoideum* while one species each was isolated from the rest of the cereals. *Aspergillus flavus* was isolated from *Hordeum vulgare* and *Zea mays* (Pakistani) while *A. niger* appeared in the American maize and no organism was isolated from other cereals when the seeds were surface sterilized for 80 sec. At 100 sec, *A. niger* and *A. flavus* were isolated from the American variety while the rest of the grains failed to yield any fungi. Table 2 shows the fungi isolated from various cereals after exposure to 1:1000 $HgCl_2$ for different lengths of time. Figure 2 shows the relative prevalence of fungi after surface disinfection.

Discussion

Fungi Imperfecti was the dominant group with a prevalence of 83%. Phycomycetes and Ascomycetes have a prevalence of 15 and 2% respectively.

The genus *Aspergillus* was the most prevalent organism with 11 species and among the species *A. flavus* topped the list with a prevalence of 22%. *A. tamarii*, however, was the least prevalent or-

TABLE 1.—FUNGI ISOLATED FROM CEREALS BEFORE SURFACE DISINFECTION.

Hordeum vulgare Linn.

1. *Aspergillus niger* van Tiegh
2. *A. flavus* Link.
3. *A. sydowii* (Bain and Sart) Thom & Church
4. *A. fumigatuts* Fres.
5. *A. candidus* Linn.
6. *A. violaceus* Raper & Fennel
7. *A. nidulans* (Eidam) Wint.
8. *A. terrcus* Thom.
9. *Alternaria alternata* (Fr.) Keissler.
10. *A. triticina* Prasada and Prabhu.
11. *Curvularia lunata* (Wakker) Boedijn.
12. *Drechslera hawaiiensis* (Bugn.) Subram & Jain
13. *Cochliobolus spicifer* Nelson.
14. *Cladosporium sphaerospermum* Penz.
15. *Rhizopus arrhizus* Fischer
16. *Penicillium decumbens* Thom
17. *P. chrysogenum* Thom.
18. *Fusarium equseti* (Corda) Sacc.
19. *Monilia* sp.

Pennisetum typhoideum Rich

1. *Aspergillus niger* van Tiegh.
2. *A. flavus* Link.
3. *A. oryzae* (Ahlb.) Cohn.
4. *A. violaceus* Raper and Fennel
5. *A. nidulans* (Eidam) Wint.
6. *A. ochraceus* Wilhelm.
7. *Penicillium notatum* Westling.
8. *P. brevicompactum* Dierckx.
9. *Rhizopus arrhizus* Fischer
10. *Alternaria alternata* (Fr.) Keissler.
11. *Curvularia lunata* (Wakker) Boedijn.
12. *C. penniseti* (Mitra) Boedijn.
13. *Drechslera hawaiiensis* (Bugn.) Subram & Jain.

Zea mays Linn. (American)

1. *Aspergillus niger* van. Tiegh.
2. *A. flavus* Link.
3. *A. violaceus* Raper & Fennel
4. *Rhizopus arrhizus* Fischer.
5. *Penicillium decumbens* Thom.

Sorghum vulgare Pers.

1. *Aspergillus niger* van Tiegh
2. *A. flavus* Link.
3. *A. flumigatus* Fres.
4. *A. nidulans* (Eidam) Wint.
5. *Fusarium semitectum* Berk & Rav
6. *F. chlamydosporum* Wr. & Rg
7. *Grechslera hawaiiensis* (Bugn.) Subram & Jain
8. *Trichoderma harzianum* Rifai agg.
9. *Absidia ramosa* (Lindt) Lender.
10. *Alternaria alternata* (Fr.) Keissler.
11. *Pleospora infectoria* Fuckel
12. *Cladosporium sphacrospermium* Penz.

Zea mays Linn. (Pakistani)

1. *Aspergillus niger* van Tiegh.
2. *A. flavus* Linnk.
3. *A. ochraceus* Wilhelm
4. *A. nidulans* (Eidam) Wint.
5. *A. tamarii* Kita.
6. *A. fumigatus* Fres.
7. *Penicillium notatum* Westling.
8. *Rhizopus arrhizus* Fischer
9. *R. oryzae* (Went & Geerl)
10. *Trichoderma polysporum* (Link ex Pers) Rifai
11. *Fusarium moniliforme* Sheld.
12. *Neurospora sitophila* Shear & Dodge
13. *Alternaria alternata* (Fr.) Keissler.
14. *Monilia* sp.

Zea mays Linn. (with hot spots)

1. *Aspergillus niger* van Tiegh.
2. *A. flavus* Link
3. *A. amstelodami* (Mangin). Thom & Church.
4. *A. violaceus* Raper and Fennel.
5. *A. nidulans* (Eidam) Wint.
6. *A. terrcus* Thom.
7. *Penicillium islandicum* Sopp.
8. *P. notatum* Westlng
9. *P. decumbens* Thom.
10. *P. citrinum* Thom.
11. *Curvularia lunata* (Wakker). Boedijn.
12. *Cladosporium sphaerospermum* Penz.
13. *Sepedonium* sp.

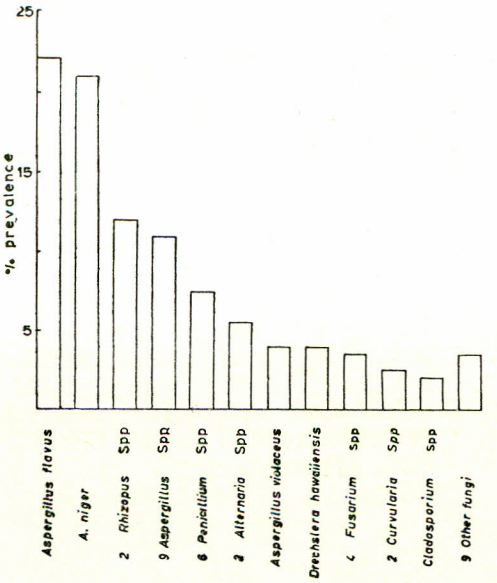

Fig. 1.—Percent prevalence of fungi before surface disinfection.

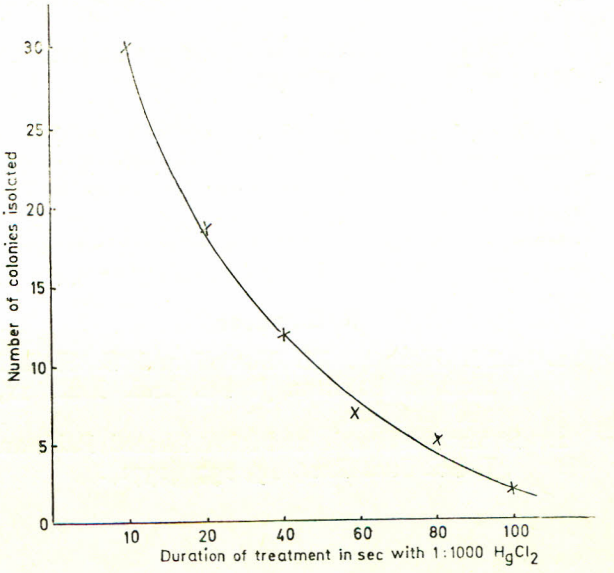

Fig. 2.—The number of colonies isolated after surface disinfection with 1:1000 $HgCl_2$.

ganism among the *Aspergillus* spp. with a frequency of 0.3%. Species of *Penicillium* were also fairly dominant as the genus was represented by six species. *P. decumbens* and *P. citrinum* with 2% and 0.30% respectively were the most and the least prevalent fungi. The species of *Penicillium* and *Aspergillus* accounted for almost half the isolated organisms. The rest of the organisms were mostly field fungi and were represented by a few species. As has been observed, temperature plays an extremely important role in the deterioration of stored food grains. Maximum temperature of 37°C was recorded inside the heap of grains of the American variety of maize which had hot spots, while the outside temperature was 32°C. The grains were very badly damaged. The moisture content of the grain in this variety was highest (27%). The temperature of all the other grains was about 34.5°C.

Moisture content, of course, varied greatly: *Pennisetum typhoideum*, 10%; *Hordeum vulgare*, 23%; *Sorghum vulgare* 11%; *Zea mays* (American), 16%; *Zea mays* (with hot spots), 27%; *Zea mays* (Pakistani), 18%.

Although less moisture content is generally associated with decrease in microorganism activity, however in case of *Pennisetum typhoideum* considerable number of fungi were isolated and the damage to the grains was also fairly widespread.

As expected, surface sterilization of grain with 1:1000 $HgCl_2$ reduced the number of fungi considerably and as may be noted from Table 2 very

TABLE 2.—FUNGI ISOLATED AFTER SURFACE DISINFECTION WITH 1:1000 $HgCl_2$.

	Pennisetum typhoideum	*Hordeum vulgare*	*Sorghum vulgare*	*Zea mays* (American)	*Zea mays* (with hot spots)	*Zea mays* (Pakistani)
10 sec	*Aspergillus flavus* *A, niger* *Penicillium notatum*	*Aspergillus flavus* *A. niger* *Alternaria altenata*	*Aspergillus flavus* *A. niger*	*Aspergillus flavus* *Rhizopus arrhizus*	*Aspergillus flavus* *A. amstelodami* *Penicillium islandicum*	*Aspergillus flavus* *Rhizopus arrhizus*
20 sec	*Aspergillus flavus* *A. niger*	*Aspergillus flavus* *A. niger* *A. candidus*	*Aspergillus niger* *Alternaria alternata*	*Aspergillus niger* *A. flavus* *A. niger*	*Aspergillus niger* *A. amstelodami* *A. niger*	*Aspergillus flavus* *A. niger*
40 sec	*Alternaria alternata*	*A. fumigatus* *Rhizopus arrhizus*	—	*Rhizopus arrhizus*	*A. flavus*	*A. nidulans*
60 sec	—	*Aspergillus flavus*	*Aspergillus niger*	*Aspergillus niger*	*A. flavus*	*A. flavus*
80 sec	—	*Aspergillus flavus*	—	*A. niger*	*A. niger*	*A. flavus*
100 sec	—	—	—	*A. niger*	*A. flavus*	—

few organisms appeared when the grains were exposed to $HgCl_2$ for 100 sec.

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