Fungal Spores: Hazardous to Health?

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Fungi have long been known to affect human well being in various ways, including disease of essential crop plants, decay of stored foods with possible concomitant production of mycotoxins, superficial and systemic infection of human tissues, and disease associated with immune stimulation such as hypersensitivity pneumonitis and toxic pneumonitis. The spores of a large number of important fungi are less than 5 μm aerodynamic diameter, and therefore are able to enter the lungs. They also may contain significant amounts of mycotoxins. Diseases associated with inhalation of fungal spores include toxic pneumonitis, hypersensitivity pneumonitis, tremors, chronic fatigue syndrome, kidney failure, and cancer. Key words: mold, fungi, mycotoxin, lung disease, toxic pneumonitis. — Environ Health Perspect 107(suppl 3):469-472 (1999). http://ehpnet1.niehs.nih.gov/docs/1999/suppl-3/469-472sorenson/abstract.html

Background

Fungi are heterotrophic, filamentous organisms that by virtue of their dependence on external sources of organic carbon and their rigid cell walls are confined to a saprobic and/or parasitic lifestyle in which they absorb soluble nutrients through the cell membrane. The fungi, together with the bacteria, are responsible for decay of organic matter and the fungi have been estimated to comprise approximately 25% of the biomass of the earth (1). As such, they are among the principal microorganisms involved in biodeterioration and are found universally in human habitats and occupational settings. The possible role of airborne microorganisms in indoor air health problems as a result of energy-saving measures was accentuated by Tobin et al. (2) and a number of studies have noted a strong relationship between mold growth in homes and respiratory symptoms (3–5). When one considers that urban residents typically spend more than 90% of their time indoors (6), it is readily recognized that very large numbers of people, both adults and children, are potentially exposed to indoor air contaminants. Although spore concentrations are usually much lower in homes than in agricultural workplaces, concentrations as high as 450,000 colony-forming units/m³ have been reported (7). In some of these homes, the toxigenic Stachybotrys chartarum (= Stachybotrys atra) was prominent on the walls and in the air. This article focuses on health effects due to mycotoxins, but fungi in general also cause other diseases such as infections, allergy, and inflammation.

Mycotoxins and Mycotoxigenic Fungi

Excluding mushroom toxins, approximately 350 to 400 fungal metabolites are considered to be toxic. Most of these are relatively small molecules of greater than 200 and the majority are less than 500 mass units (8). Perhaps the most important mycotoxins in agriculture are the aflatoxins, the 1,2,3-trisubstituted pyrroles, the fumonisins, and ochratoxin. Species belonging to the genera Acremonium, Penicillium, and Fusarium are common contaminants of agricultural commodities, and some of the mycotoxins produced by these species are produced by fungi common in house dust (2). In addition, some toxigenic fungi produce many different mycotoxins. For example, the Penicillium verrucosum complex (P. verrucosum, P. aurantiogriseum, P. viridicatum, P. crustosum, and P. solitum) produce nearly 20 different mycotoxins (9) and S. chartarum produces several trichothecenes including the highly potent macrocyclic trichothecenes as well as a variety of other mycotoxins (10). Other toxigenic fungi include species of Alternaria, Paecilomyces, Rhizopus, Trichoderma, and Trichothecium. All of these fungi occur commonly in soil, agricultural products, grain dust, and house dust (2).

Health Effects Linked with Inhalation of Mycotoxins

Although extensive literature has been developed since the discovery of the aflatoxins, few studies have been conducted to document the occurrence of these substances in airborne grain or other organic dust or to estimate the inhalation hazard to workers and others exposed to contaminated airborne dust. Several studies have provided evidence for the association of cancer in humans with inhalation of aflatoxin-contaminated dust, e.g., lung cancer (11–13) or colon cancer (14). Olsen et al. (13) noted elevated risks for liver cancer and cancers of biliary tract among animal feed workers that increased by 2- to 3-fold significance after a 10-year latency. Their daily pulmonary exposure was estimated to be approximately 170 ng. Autrup et al. (15) used measurements of aflatoxin bound to serum albumin as an index of exposure and showed that 7 of 45 workers exposed to feed contaminated with low levels of aflatoxin B₁ (AFB₁; 0–26 μg/kg) had detectable levels of AFB₁ bound to serum albumin, confirming systemic exposure. Zarba et al. (16) demonstrated that aerosol inhalation is an effective route of exposure to AFB₁ in rats. In their experiments, approximately 2% of the administered dose became bound to liver DNA and the amounts of DNA adducts were statistically different among the treated groups. These workers (17) also demonstrated that nose-only aerosol in exposure of rats at an estimated dose of 16.8 μg/kg body weight suppressed alveolar macrophage (AM) phagocytosis, with the effect persisting for approximately 2 weeks. These findings indicate that inhalation exposure to AFB₁ is an occupational hazard where exposure to AFB₁-laden dust is common.

Inhalation exposure to spores of S. chartarum has also been associated with episodes of human illness. Andrassy et al. (18) reported an outbreak of illness in which workers exposed to hay heavily contaminated with S. chartarum unanymously complained of dyspnea, airway obstruction, sore throat, bloody nose or nasal secretions, conjunctivitis, and inflammation of the...
Two other recent reports of human disease thought to be due to inhalation of mycotoxins are noteworthy. Di Paolo et al. (30) reported acute renal failure in a female agricultural worker exposed to grain dust in an enclosed granary, believed to be due to inhalation of ochratoxin in the spores of Aspergillus ochraceous. Ochratoxin was not demonstrated in airborne dust in the granary, but the authors were able to isolate A. ochraceous from a sample of wheat from the granary. Ochratoxin A was observed in extracts of ground moldy grains, and Di Paolo and colleagues were able to demonstrate acute kidney failure in experimental animals (rabbits and guinea pigs) exposed for 8 hr to aerosols generated by their natural movement on moldy wheat in their cages.

Gordon et al. (31) reported tremorgenic encephalopathy in a young man exposed to high concentrations of grain dust contaminated with several species of fungi known to be capable of producing tremorgenic mycotoxins. Because of the circumstances of exposure, the similarity of his syndrome to that of an animal model, and the lack of an alternative explanation, the authors proposed that his illness may have resulted from inhalation of tremorgenic mycotoxin(a).

Mycotoxins in Spores
Species of fungi in which mycotoxins have been reported in the spores include Alternaria alternata (32), Aspergillus fumigatus (33–35), Aspergillus flavus and Aspergillus parasiticus (36), Fusarium graminearum (1), Fusarium sporotrichioides (1), and S. chartarum (37). Several different mycotoxins were demonstrated in these investigations including deoxynivalenol (1), fumitremorgen and verruculogen (35), fumigaclavine C (34), T-2 toxin (1), trypacidin (33), alternariol and alternariol monomethyl ether (32), and the macrocyclic trichotheccenes satratoxins G and H (37). Gliotoxin has been demonstrated in tissues infected by A. fumigatus (38) and Candida albicans (39) but was not detected in spores of A. fumigatus (40). It is likely that mycotoxins occur in spores of toxicogenic species much more commonly than is currently appreciated, as they have been found in spores in a high proportion of species in which attempts were made to find them. In a study in Scotland, extracts from spores of 47% of a group of 83 isolates collected from damp public sector housing in Scotland were cytotoxic to the human embryonic hybrid fibroblast lung cell line MRC-5 (41).

These findings seem to support earlier findings of health hazards in epidemiologic studies of the inhabitants of damp, moldy houses (42). Spores of F. aurantiogriseus containing the benzodiazepine metabolite aurantidine cause nephrotoxicity and pathology typical of Balkan endemic nephropathy when mixed with feed and fed to rats (43). These findings confirm the presence of aurantidine in the spores and suggest that workers and others who handle infected grain may be at risk of exposure by inhalation. The vast majority of mycotoxins are nonvolatile and therefore mycotoxin exposure by inhalation is most likely to occur via inhalation of spores.

Effects of Mycotoxins on Alveolar Macrophages and Immune Function
T-2 toxin, patulin, and penicillic acid were shown to be acutely toxic to rat AM in vitro, causing membrane damage, inhibition of protein and RNA synthesis, inhibition of phagocytosis, and inhibition of the ability of AM to respond to lymphokines (44–47). Ayral et al. (48) showed that the trichotheccenes diacetoxyscirpenol and deoxynivalenol reduce phagocytosis, suppress microbicidal activity, and inhibit superoxide anion production and phagosome–lysosome fusion of peritoneal macrophages at concentrations that did not affect cell viability. Similarly, Vidal and Mavet (49) demonstrated inhibition of phagocytosis of Pseudomonas aeruginosa by murine peritoneal macrophages in the presence of 0.001 μM T-2 toxin.

The trichotheccene mycotoxins are immunotoxic in rats and mice, causing acute inhibition of antibody and delay of skin graft rejection (50,51). Gliotoxin has antiphagocytic and immunomodulating activity, it is produced by A. fumigatus (52), and it has been demonstrated in tissues infected by A. fumigatus (38). Gliotoxin also contributes to the pathogenesis of vaginal candidiasis (39). Jakab et al. (17) confirmed earlier reports that dietary exposure to AFB1 impairs innate and acquired host defenses. Subsequent work by these authors also showed that AM phagocytosis was suppressed for approximately 2 weeks following nose-only inhalation exposure to an estimated dose of 16.8 μg/kg. Exposure using intracheal instillation (IT) of AFB1 also suppressed AM phagocytosis in a dose-dependent manner, but approximately 10-fold higher doses were required for IT than for inhalation. Animals exposed by IT administration also had impaired release of tumor

Skin on the face and body. Croft et al. (19) reported long-term illness, apparently caused by heavy infestation of a water-damaged home in Chicago, Illinois. Between 1993 and 1998, there were 34 cases, including 10 deaths, of idiopathic pulmonary hemosiderosis among infants in Cleveland, Ohio. Incidence of this disease is generally quite low (20,21). The vast majority of cases occurred within a very small geographic area of the city. Case homes tended to have a higher prevalence of water damage (22) and higher quantities of toxigenic S. chartarum (23). Strains of S. chartarum isolated from both case and control homes were shown to be toxigenic, but there was no correlation between toxigenicity of the isolate and source, i.e., case versus control. The disease seemed to correlate with frequency of occurrence and quantity of the organism rather than with strain (24). Memnoniella echinata, a close relative of S. chartarum with similar biochemical and physiologic characteristics and habitat preferences, was also found in samples from a case home. M. echinata produces some of the same toxins as S. chartarum (24,25) but has smaller spores, although the conidia of both species are small enough to reach the alveoli (25). Aqueous rinses of the spores of both species were nearly as toxic as methanolic extracts of the spores themselves (25), indicating that these highly potent toxins would be readily released into the microenvironment of the developing lung cells in vivo.

Further support for the proposed mycotoxin etiology of infant pulmonary hemosiderosis comes from animal studies in which mice were treated intranasally with both nontoxic and toxic spores of S. chartarum (26). Severe inflammatory changes, including hemorrhage, were observed in mice receiving toxic spores of this species. Two recent studies report occupational illness associated with chronic, low-level exposure to S. chartarum in water-damaged office buildings (27,28). Dill et al. (29) noted massive development of S. chartarum on decomposable flower pots made of recycled paper, which led to the development of very painful inflamed efflorescences at the fingertips followed by scaling of the skin. The symptoms disappeared when gloves were worn to handle the pots and were ascribed to trichothecene mycotoxins. Although respiratory symptoms were not noted in the report, viable spore counts of S. chartarum up to 7,500 spores/m³ were determined when the pots were handled (30–100 spores/m³ when the pots were not handled).
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