Maize Mycotoxins in Latin America

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Fungi are capable of producing a number of secondary metabolites, including pigments, compounds with antibiotic properties, and the group of chemicals called mycotoxins. Mycotoxins are fungal metabolites that are toxic to man and animals. Although we have known for some time that the consumption of moldy grains or foods can cause health disorders in animals, it was not until the outbreak of the Turkey X disease of poultry in the United Kingdom in the 1960s (16) that we became fully aware of the potential hazard of mycotoxins in our food chain. A flood of research papers on mycotoxicology has been published since Turkey X disease, covering all aspects of mycotoxicology.

The Food, Drug and Cosmetic Act was made law in the United States in order to protect consumers from harmful substances that might become incorporated into foods. In 1969, because of the earlier discovery of aflatoxins in feedstuffs, the Food and Drug Administration declared an "action threshold" of 20 ppb or more of aflatoxins. This meant that any commodity containing this quantity of aflatoxin destined for either human or animal consumption may not be shipped interstate. Except for an occasional exemption, this policy remains in effect.

Pier et al (14) have categorized the effects of mycotoxins on animals into acute primary and chronic primary mycotoxicoses and secondary mycotoxin diseases. Acute primary mycotoxicosis occurs when moderate to high concentrations of mycotoxins have been consumed, causing various reactions such as hepatitis, hemorrhage, nephritis, and death. The effects of various mycotoxins on poultry range from impaired immunogenesis to acute death (Table 1), depending on the level of aflatoxin consumed. Diagnosis of chronic primary mycotoxicosis is more difficult because the level of toxins being injested is lower and macroscopic changes are not visible. Some examples of chronic mycotoxicosis include reduced milk yields in cows, reduced egg production and increased egg crackage in poultry, and slowed growth rates in affected animals. Secondary mycotoxin disease results when animals are exposed to low levels of mycotoxins resulting in increased susceptibility to various diseases. For example, exposure of poultry to sublethal levels of ochratoxin A causes a depression in the number of immunoglobulin-containing cells.

There is also evidence that the consumption of mycotoxins by humans can cause health problems. Stoloff (18) compared the occurrence of liver cancer in various parts of the world with the incidence and concentration of aflatoxins in peanut products being consumed. It has been hypothesized that since aflatoxins can cause liver carcinomas in animals, they may also be responsible for liver cancer in humans. Stoloff (18) reported that in countries with a low incidence of liver cancer, such as the United States and Canada, the mean concentration of aflatoxin per kilogram of peanut products sampled was 1 μg, whereas in Thailand, where the incidence of liver cancer is much higher, the mean concentration was 470 $\mu g\ kg^{-1}$. The outbreak of alimentary toxic aleukia in the Soviet Union during 1942–1947 has been attributed to the consumption of molded cereals contaminated with the *Fusarium* mycotoxins called trichothecenes. The high incidence of esophageal cancer in the Transkei section of South Africa and parts of China is believed to have resulted from the consumption of maize contaminated with *Fusarium moniliforme* and its mycotoxin. Other mycotoxin-induced diseases in man include Kashin-Bek disease caused by an additional *Fusarium* toxin and the *Penicillium* mycotoxin disease called cardiac beriberi.

Mycotoxicologists now recognize that mycotoxin occurrences are not "rare" biological phenomena but that mycotoxins can occur wherever the organisms producing these chemicals are able to grow. Fungi that produce mycotoxins represent virtually all members of the fungal kingdom, although the most

Table 1. Effects of mycotoxins on poultry[a]

Mycotoxin	Effects	Quantity of toxin (ppm)
Aflatoxin	Acute death, hepatic necrosis, hemorrhage	1–10
	Impaired immunogenesis	0.25
	Reduced resistance	0.6–1.0
	Reduced gain	1.5–2.5
	Decreased egg production	2–8
Ochratoxin	Acute disease, diarrhea, death	4–16
	Toxic nephropathy	4
	Reduced gain	2–4
	Decreased egg production	2
T-2	Oral necrosis	4
	Reduced gain	4
	Decreased egg production	20

[a]From Pier et al (14).

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frequently encountered toxigenic fungi are species of *Aspergillus, Penicillium*, and *Fusarium* (17). Chemical structures of mycotoxins are diverse, and so many have been described that they are frequently grouped according to the biosynthetic pathway that produces them rather than as separate compounds.

Mycotoxin occurrence is not limited to undeveloped countries; deoxynivalenol has been reported in wheat in Canada (20) and aflatoxins, in maize in the southeastern United States (22). We do know that mycotoxins are more likely to occur at higher concentrations in the tropical or subtropical developing countries of the world, for a number of reasons. A developing country is less likely to have adequate drying or storage facilities for food and feed crops and often lacks adequate funding to educate its people about the steps that should be taken to prevent mycotoxins and about the hazards of consuming contaminated foods and feeds. A tropical or subtropical country also experiences hot, humid conditions that tend to stimulate growth and toxin production by mold fungi.

Studies on mycotoxins in developing tropical countries have focused on India, Southeast Asia, and parts of Africa that yearly experience severe losses of groundnuts and other food commodities because of mycotoxin contamination (17). The geographic area of the world largely overlooked by mycotoxicologists is Central and South America, where the climate is ideal for mycotoxin development. Two meetings in 1986 were significant in that they were the first times that U.S. scientists met with colleagues from Latin America to discuss the health and economic implications of maize mycotoxins. Symposia on aflatoxin contamination of maize were held at the University of Puerto Rico, Mayaguez, in January 1986 and at the International Maize and Wheat Improvement Center (CIMMYT) in Mexico in April 1986 (23).

In the following review of the findings of these meetings and of previously published literature, we have attempted to provide a synopsis of: 1) the magnitude of the maize mycotoxin problem in Latin America, 2) the reasons why these countries, in particular, have been so severely affected by mycotoxins, and 3) what we as plant scientists can do to help alleviate this worldwide problem.

The Magnitude of the Problem

Research efforts to establish the magnitude of the mycotoxin problem in Latin America were initiated in the late 1960s after the outbreak of Turkey X disease (16). Grain lots moving within commercial channels were often found to be contaminated with mycotoxins, primarily AFB_1, in concentrations harmful if used as food or animal feed (10). The bulk of mycotoxin research in Latin America has been conducted on maize and specifically on aflatoxin, although other toxins such as zearalenone, T-2, deoxynivalenol, penicillic acid, kojic acid, and ochratoxin have been detected in maize (3). Multiple toxins can also occur in moldy products, so it is possible that animals, including humans, may consume more than one toxin at a time. Although we have no evidence, it would seem more likely that moldy grains are used as feed for domestic animals as well as food for humans in a developing country, thus greatly increasing the likelihood of exposure to mycotoxins. The most reliable means of estimating the severity of a mycotoxin problem in a country is by surveys of food and feed products for the presence of mycotoxins. In addition, the incidence of mycotoxicoses in farm animals provides evidence for high levels of mycotoxin production.

In Africa and the Far East, an association between long-term exposure to AFB_1 and human liver cancer has been reported (12). To date, no such finding has been reported for Latin America. However, there are surveys reporting *A. flavus* and AFB_1 in maize (30–40 ppb) and also cases of aflatoxicosis in livestock and poultry (4,15). In Colombia, for example, 14 cases of aflatoxicosis in poultry were reported in 1975 and 41 cases in 1976 (1). The feeds were formulated from maize, sorghum, soybean, and cottonseed meal—good substrates for growth of aflatoxigenic *A. flavus* strains. At present, however, there is no epidemiological evidence of an increase in primary liver cancer in these countries. Without citing any evidence, Calderon (4) stated that the incidence of hepatic and gastrointestinal disease in El Salvador had been on the increase.

There is more evidence for the occurrence of mycotoxins in maize, both before and after storage, and in commodities manufactured from maize. Banchero (3) reviewed the data on the incidence of AFB_1 and zearalenone in maize collected in Argentina from 1976 to 1984 that was destined for exportation and human consumption. In 1976, 50% of 50 samples of maize contained AFB_1, and in 1977, 10% of 267 samples were contaminated (the levels of AFB_1 were not given). In 1980, 33% of 85 samples contained zearalenone in levels from 200 to 1,600 ppb. Banchero (3) also investigated the level of AFB_1 and zearalenone in maize in 1981 and 1982 (Table 2). Concentrations of AFB_1 were slightly higher (3–64 ppb) in maize consumed in Argentina than in maize to be exported, which contained 13 positive samples within the FDA guidelines of 20 ppb of aflatoxin. Although zearalenone was detected in only one sample of maize for local consumption, the levels were in excess of 900 ppb. In 10% of the samples of exported maize, concentrations of zearalenone ranged from 30 to 305 ppb.

Calderon (4) reported in 1979 on the natural occurrence of AFB_1 in government-owned silos containing maize, beans, sorghum, or rice in El Salvador. Of 98 silos sampled, 72 contained detectable levels of AFB_1: 33 of 36 black bean samples (trace to 79 ppb), 16 of 23 red bean samples (trace to 63 ppb), 16 of 32 white corn samples and both of two yellow corn samples (trace to 22 ppb), four sorghum samples (trace to 28 ppb), and one sample of rice (trace).

Recently, Cuero et al (6) compared the population of toxigenic fungi in maize at harvest collected from two different latitudes in Columbia (Table 3). All 54 samples were contaminated by toxigenic strains of *A. flavus, A. ochraceus, P. citrinum*, or *F. graminearum*. Ninety fungal isolates were grown in culture, and analysis of the extracts by thin-layer

Table 2. Levels of AFB_1 and zearalenone in maize produced in Argentina for local human consumption and for exportation in 1981 and 1982[a]

	AFB_1		Zearalenone	
Destination	Positive/total samples	Toxin level (ppb)	Positive/total samples	Toxin level (ppb)
Local consumption	14/41	3–64	1/41	912
Exportation	13/53	2–15	5/53	30–305

[a]From Banchero (3).

Table 3. Isolation of toxigenic fungi at harvest collected from two locations in Colombia[a]

Species	Isolates from highlands	Isolates from lowlands	Toxigenic isolates
Aspergillus flavus	6(1)[b]	98(18)	15(50)
A. ochraceus	61(11)	354(64)	9(45)
Penicillium citrinum	237(28)	278(33)	13(65)
Fusarium graminearum	678(45)	350(23)	12(60)

[a]From Cuero et al (6).
[b]Number of isolates (percentage of total).

chromatography showed that 50% of the *A. flavus* isolates produced AFB_1, 45% of the *A. ochraceus* isolates produced ochratoxin A, 65% of the *P. citrinum* isolates produced citrinin, and 60% of the *F. graminearum* isolates produced zearalenone. Mora (13) analyzed maize, beans, and rice samples collected from commercial markets in Costa Rica for AFB_1 (Table 4). Rice did not contain detectable AFB_1, but both bean and maize were contaminated with the toxin. In the first year of the survey, 39% of the maize samples contained more than 20 ppb, whereas in the second year, 30% had 20 ppb or higher. Bean samples had much lower concentrations of AFB_1; less than 5% of the samples had concentrations higher than 20 ppb.

The most convincing evidence that a serious maize mycotoxin problem exists in Latin America comes from the results of a survey done by Torreblanca et al (19) in 1980. They examined 65 samples of tortillas purchased from 50 stores in Mexico City for AFB_1 and found that 54% were contaminated with levels of AFB_1 in excess of 20 ppb; four samples contained 160 ppb and one, 500 ppb. The average Mexican eats approximately 1 lb (over 450 g) of tortillas daily, which means consuming as much as 0.2 mg/kg of AFB_1. Van Rensburg (21) stated that humans eating food containing 1.7 mg/kg of AFB_1 "may within a short time develop serious liver damage."

In summary, the presence of toxigenic fungi and the mycotoxins they produce in foods and feed before and during storage, the reports of mycotoxicoses in animals, and the poor storage facilities available for maize all point to a potentially serious maize mycotoxin problem in Latin America.

Prevention and Control of Maize Mycotoxins

Although AFB_1 in maize begins in the field (11), few preventive measures are available to a small grower in an economically stressed country. Adequate fertilization, irrigation, and control of insect and weed competitors are commonplace in a highly technological country. We are, however, unaware of any Latin American field studies done to reduce the level of AFB_1 development in the field other than a report by Echandi (8) on the influence of bending of maize stalks so that the ears hang down. The lack of storage facilities often means that maize must remain in the field for several months after maturity. Maize stalks are bent to reduce the moisture levels in the ears. Echandi (8) found that this practice did not influence the occurrence of AFB_1 but did prevent kernel germination in ears still attached to the plant. He also found that storing corn unshelled rather than shelled, which may reduce kernel or insect damage during storage, also reduced the risks of aflatoxin formation. He suggested that some simple cultural techniques compatible with even the most primitive production systems could be developed to eliminate or reduce the likelihood of AFB_1 production in maize. Because adequate storage facilities for maize in most instances do not exist in Latin America, proper farm storage is very important. The development of maize genotypes resistant to *Aspergillus* spp. has been suggested in some Latin American studies as a means of controlling AFB_1 production (6). However, research for many years in the United States on the development of maize genotypes resistant to aflatoxin has been essentially unsuccessful (7). Therefore, the development of other means of preventing AFB_1 may be more productive in countries with limited research capabilities.

Gonzalez (9) has been investigating the use of urea as a means of preventing growth and AFB_1 production by *A. flavus*. Maize was treated with urea, stored in containers that allowed growth and AFB_1 production by *A. flavus*, and, after varying periods of time, analyzed for growth and AFB_1 concentration. After 6 wk of incubation, AFB_1 could be detected in maize without urea. In maize treated with urea, however, AFB_1 production was not detected until after 10 wk of incubation, when it reached the level of untreated maize (Table 5). Gonzalez (9) suggested that although the results were preliminary, they appeared to be encouraging enough to warrant further investigation. In Mexico, however, the cost of chemicals to treat maize is of great importance and should not exceed 10 per kilogram (8) to be economically feasible. This severely restricts which chemicals could be used to control maize mycotoxins.

The only other studies we are aware of concerning the control or decontamination of AFB_1-contaminated maize deal with the influence of cooking and lime water on AFB_1 levels in maize during the manufacture of tortillas. In the preparation of tortillas from white corn, the kernels are soaked in lime water, cooked, then stored overnight before being formed into tortillas. Carvajal et al (5) found that AFB_1 and AFB_2 levels were 20% lower in tortillas than in the originally contaminated maize. Arriola et al (2) found similar results and concluded that the level of lime and the temperature normally used in the manufacture of tortillas did not reduce aflatoxin levels in contaminated maize enough to make the maize safe for consumption. They found, however, that increasing the lime level above 1.87% w/v, which is normally used in tortilla production, reduced the aflatoxin level as much as 97.1%; the tortillas had objectionable color and flavor, however. Further studies on modifications of methods used in the manufacture of tortillas to reduce AFB_1 levels appears justifiable.

Outlook

The scientists who attended the CIMMYT workshop on maize mycotoxins made a series of recommendations to justify the development of international and interdisciplinary research projects dealing with the problem of maize mycotoxins (23). Our outlook can best be presented by determining where we must go from here, considering what knowledge has accumulated in the last 20 yr concerning mycotoxins. Knowledge of mycotoxins in tropical and subtropical countries appears to be in its infancy compared with that in other countries of the world. It is the respon-

Table 4. Occurrence of AFB_1 in grains collected at commercial markets in Costa Rica[a]

Grain	Year of collection	AFB_1 (ppb) 0–20	21–50	51–100	101–500	500+	Maximum level
Maize	First	32(61)[b]	9(17)	4(8)	4(8)	3(6)	1,000
	Second	40(70)	10(16)	1(2)	4(7)	2(5)	3,500
Beans	First	69(96)	2(3)	0	1(1)	0	150
	Second	64(98)	1(1)	1(1)	0	0	90
Rice	First	64(100)	0	0	0	0	20
	Second	53(100)	0	0	0	0	20
	Total	322(88)	22(6)	6(2)	9(2)	5(2)	

[a]From Mora (13).
[b]Number of samples (percentage of total).

Table 5. Influence of urea on level of AFB_1 in stored maize[a]

Treatment	AFB_1 (ppb) in maize stored at 25 C, 95% RH for: 2 wk	4 wk	6 wk	8 wk	10 wk	12 wk
No urea	0(+)[b]	0(+)	10(+)	10(+)	15(+)	25(+)
Urea (0.5% w/w)	0(−)	0(−)	0(+)	0(+)	15(+)	25(+)

[a]From Gonzalez (9).
[b]Average of three replications; + = growth, − = no growth of *Aspergillus flavus*.

sibility of scientists to share their knowledge and expertise with others so that eventually all peoples of the world are able to consume agricultural products without fear of mycotoxin contamination. The establishment of communication among scientists—which in the case of mycotoxicology involves toxicologists, mycologists, physicians, veterinarians, microbiologists, chemists, biochemists, and, of course, plant pathologists—is the first step in the development of interdisciplinary cooperative research programs. We saw this happen in 1987, and we can only hope that it will continue and expand.

We offer the following general recommendations: 1) Alert regulatory and administrative personnel in tropical and subtropical countries to the potential hazards of mycotoxin-contaminated maize and 2) develop means of reducing contamination of maize by aflatoxins and other mycotoxins through cooperative research, information dissemination, and education of the producers, processors, and consumers of maize.

Specific areas of proposed research, which should take into account the economic and cultural practices of an area, include surveys of maize to establish the severity and distribution of mycotoxins, studies on the use of preservatives to reduce mycotoxin contamination, and biological control of mycotoxins by use of competing microorganisms. Other areas of research include: 1) the influence of preharvest environment on mycotoxin development, such as studies on inoculum sources and the infection process; 2) the development of standardized methods for sampling and mycotoxin quantification, including use of immunoassays; 3) the establishment of guidelines to study the toxicology of maize mycotoxins in human and animal populations; 4) the detoxification of maize mycotoxins by the addition of preservatives or modifications of methods used in the processing of maize; and 5) the development of management strategies to strengthen international cooperative research projects.

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