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## MYCOTOXINS AND MYCOTOXICOSES

Mycotoxins are fungal metabolites that are toxic when consumed by animals or humans. They can accumulate in corn, cereals, soybeans, sorghum, peanuts, and other food and feed crops in the field, in grain during transportation and during improper storage favorable for the growth of the toxin-producing fungus or fungi.

Diseases resulting from the consumption of mycotoxins are called mycotoxicoses. The effects range from unthriftiness, loss of appetite, feed refusal and decreased feed efficiency to cancer (liver cirrhosis) and mortality in domestic animals. In many years in the Midwest, vomitoxin, produced by the *Fusarium* fungus is found in wheat or other grain crops. This causes a "feed refusal" by animals and can also produce vomiting and poor weight gain if it goes undetected in feed supplies. In human populations, mycotoxins are often found in developing nations where storage facilities and conditions do not keep grains at a low moisture content. Consumption of grains containing these mycotoxins can produce various cancers and other serious medical problems.

Three mycotoxins account for 99 percent of the diagnosed animal mycotoxicoses in the Midwest— aflatoxin, zearalenone and deoxynivalenol. Deoxynivalenol (also called DON, vomitoxin or feed refusal toxin) and zearalenone (estrogenic mycotoxin) often coexist in corn with *Fusarium graminearum*.

DON has historically been detected far more in Illinois corn samples than any other mycotoxin. Another group of toxins called aflatoxins can develop in corn, cereals, sorghum, peanuts, and other oil-seed crops and is the most potent of naturally occurring animal carcinogens. If aflatoxin is consumed regularly by sensitive young animals at 50 to 100 parts per billion (ppb), the result can be fatal liver cancer; in older or mature animals, the effect may only be minor. Most species of animals appear to be susceptible, although susceptibility may vary. Animals on a protein-deficient diet are more sensitive to aflatoxin injury than are those on a well-balanced ration.

Aflatoxin has been associated with liver cancer in humans. Although the FDA sets limits for aflatoxins (20 ppb) in commercial grains, these toxins may not be equally distributed throughout a load of grain. Thus, although the tests may indicate a low level in one part of the load, other areas may be well-above the 200 ppb limit. Thus, careful sampling is essential to detect true levels of aflatoxins. Grains with high levels are subject to confiscation. Mixing high and low aflatoxin-content corn to achieve a blend, which meets FDA standards, constitutes adulteration, and is subject to severe FDA penalties. However, under some circumstances such as in 1988, FDA and state department of agriculture regulations have permitted the blending of aflatoxin-contaminated and sound grain to obtain mixes that can be fed to some tolerant nonlactating animals. Such feeds can be used on the farm where it is produced, but **cannot** be sold.

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*For further information on field crop disease, contact your nearest Extension office or an Extension Specialist in the Department of Crop Sciences, University of Illinois, Urbana-Champaign.*

The “action levels” as stated by the FDA are levels which “may be injurious” due to sampling or other error and may not always be 100 percent accurate. Corn with 100 ppb of aflatoxin can be fed to mature nonlactating beef cattle and swine or mature poultry—such as laying hens—and 300 ppb for finishing (i.e. feedlot) beef cattle and 200 ppb for finishing swine, e.g., 100 pounds or greater without damage to the animals themselves or to humans eating edible portions of the animals. Lactating cows consuming feed containing 20 ppb or less of aflatoxin will have less than 0.1 ppb of aflatoxin in the milk, the smallest amount detectable.

If a grower produces home-grown feed, it is essential to be aware of the aflatoxin hazard locally and, if necessary, have the grain checked for aflatoxin before it is made into feed. This also applies to the toxins produced by other fungi discussed in later sections.

Three genera of fungi—*Aspergillus*, *Penicillium*, and *Fusarium* (sexual state *Gibberella*)—are involved most frequently in cases of mycotoxin contamination in corn, small grains, and soybeans. *Aspergillus flavus* produces aflatoxins in starchy cereal grains (e.g., corn, wheat, sorghum, oats, barley, millet, rice) at a moisture content of about 18 percent and above, i.e., in equilibrium with 85 percent relative humidity (0.85 available water), and at temperatures of 55° to 100°F (13° to 37°C) with an optimum at 73° to 95°F (25° to 35°C). The critical moisture content for soybeans is 17 to 17.5 percent below which the toxin is not produced. *A. flavus* will grow slowly below 55°F (24°C), and rapidly up to 131°F (55°C) but will not produce aflatoxin at temperatures below 55°F (24°C) or above 104°F (42°C). Under optimum conditions for growth some aflatoxin can be produced by *A. flavus* within 24 to 36 hours and a biologically significant amount in a few days.

These moisture numbers reflect the moisture content of the **wettest** kernels in a grain mass, **not** the average. Even if the average moisture content is below 15 percent, some grain in that mass may be above the critical moisture content.

Fungi may grow well under a given set of conditions but not necessarily produce mycotoxins. The quality of the grain and its suitability for storage are adversely affected by (1) a high moisture content, (2) physical damage to the kernels, and (3) the extent to which storage fungi have invaded the seed. Although *A. flavus* flourishes on many crop plants, it does not produce equal amounts of aflatoxin on all of them. Aflatoxins are also much more likely to be formed in warm-to-hot, humid regions such as the southeastern states than in Illinois and the rest of the Corn Belt.

## AFLATOXINS AND AFLATOXICOSES

*Aspergillus flavus* and the closely related *A. parasiticus*, present everywhere in soils and in all kinds of decaying plant materials, cause stored grains to heat and decay and, under certain conditions, commonly invade corn and the cereal grains in the field when the weather is dry and hot (more than 86°F or 30°C) during the silking to late stage of kernel development such as occurred in 1988.

Aflatoxin produced by *A. flavus* is most likely to be produced in the field and storage after the kernels have been damaged by insects, birds, mites, hail, early frost, heat and drought stress, windstorms and other unfavorable weather conditions. The presence of *A. flavus* in a given feed sample does **not** imply that the feed is unwholesome or that it will contain high levels of aflatoxin. Aflatoxin persists under extreme environmental conditions and is even relatively heat stable at temperatures above boiling water (212°F or 100°C).

Regular or occasional consumption by farm animals of feed containing aflatoxin in the range of less than 100 ppb to a few hundred parts per million (ppm) results in decreased feed consumption, poor feed conversion, stunting, and decreased production—flesh or eggs in poultry, milk in dairy cows, and meat in pigs and beef cattle. The reduced growth and productivity may be accompanied by damage to the liver, hemorrhaging into the muscles or body cavities, and suppression of natural immunity to parasites and pathogens always present in the environment. Once the damage has been done, the animals will not fully recover even if returned to a toxin-free ration.

## ZEARALENONE AND THE ESTROGENIC SYNDROME

Zearalenone is produced by *Fusarium* species that contribute to the ear and stalk rot complex growing in the ears of corn and on the heads of cereal grains standing in the field or in stored ear corn in the Corn Belt. Zearalenone also forms in ears of corn left unharvested in the field. When consumed by swine at more than 0.1 to 5 parts per million (ppm), these compounds cause the estrogenic syndrome, which is characterized externally by a swollen and edematous vulva in females with enlarged mammary glands and a shrinking of the testes in young males. The financial loss to farmers comes out primarily through poor reproductive performance.

Estrogenism in swine and dairy cows usually is more prevalent in the winter and early spring when a new bin or silo is opened because once the fungus is established in the grain, it requires at least 22 percent moisture to grow regardless of the temperature. A cool, wet growing season is required to produce biologically significant amounts of zearalenone. When growing in corn, some strains of *Fusarium graminearum* produce a mixture of toxins along with zearalenone. One or more of these when consumed by swine can cause infertility, severe stunting, and other deleterious effects.

*F. graminearum* (*Gibberella zeae*) requires a minimum of 22 to 25 percent moisture to grow, and if shelled corn is stored at that moisture content, it is likely to be invaded by a mixture of other fungi, yeasts, and bacteria with which *F. graminearum* may not compete well, but there may be pockets of the fungus. There might be situations, as in low temperature drying, in which *F. graminearum* could continue to grow for a short time, but there are no reported instances of this happening. Also, there is no record of zearalenone being formed anew in high moisture shelled corn stored in silos. *F. graminearum* ear rot is primarily a problem in the field and in stored corn in cribs exposed to low temperatures.

## DEOXYNIVALENOL (VOMITOXIN) AND FEED REFUSAL IN SWINE

*Fusarium graminearum* growing in the ears of corn and on the heads of cereal grains before harvest is more likely to produce other toxins besides zearalenone, including deoxynivalenol or DON that make the grain unpalatable to swine. Apparently different strains of *F. graminearum* produce DON than those that produce zearalenone. Field-infected corn with visibly damaged kernels of more than about 4 percent may be refused by pigs, depending on the DON concentration. Feed refusal may occasionally or rarely be accompanied by swollen vulvas and reproductive problems from zearalenone and DON in the same ration, and sometimes a complex of effects can occur.

Wet, rainy, warm and humid weather from flowering time on promotes infection of corn and cereals by *Fusarium* species, resulting in ear rot in corn and scab or head blight in wheat, barley, oats, and rye. Low temperatures following infection may increase the production of DON. The toxin already present in corn at harvest may increase in ear corn stored in cribs, as does zearalenone. DON is **not** known to increase in stored shelled corn or in small grains that become contaminated in the field; growth of *Fusarium*

requires a minimum moisture of 22 to 25 percent in corn. Grains free of the toxin at harvest will not develop it in storage.

Feeds that contain 1 ppm of DON may result in significant reductions in swine feed consumption and weight gain. Vomiting is rather uncommon in field cases because pigs will ordinarily not eat enough of the contaminated feed.

#### FUSARIUM MONILIFORME, FUMONISINS, AND BLIND STAGGERS IN HORSES

Blind staggers (technically known as equine leucoencephalomalacia) occurs sporadically in many countries in horses, donkeys and mules foraging corn left standing in the field after harvest or consuming corn screenings or sweepings. The fumonisin B1 toxin is generally considered to be produced only by certain strains of *Fusarium moniliforme*. It is common even in food-grade corn and often abundant in ground feeds and in silage. Growing pigs fed a ration containing 78 to 82 percent corn heavily colonized by *F. moniliforme* grew as well as the control pigs fed a ration of sound corn.

#### ERGOT AND ERGOTISM

Ergot toxicity, caused by the fungus *Claviceps purpurea*, differs from other mycotoxicoses since it results from the consumption of considerable amounts of fungal tissue. In other mycotoxicoses the toxins are secreted into plant tissues in which the fungus is growing, and very little fungus material is consumed. The ergot fungus infects the flowers of cereals and many grasses forming characteristic black, spur-like sclerotia that replace the ovaries. The sclerotia or ergot bodies contain toxic alkaloids that when consumed regularly in small amounts result in a complex of signs collectively called ergotism.

We do not yet know all we need to know about mycotoxins and mycotoxicoses, but we do know enough to say that they constitute real and present problems for growers, marketers, processors, and users of agricultural products.

#### SAMPLING FOR MYCOTOXINS AND SAMPLE PREPARATION

An adequate sample of suspect grain must be obtained to use any assay method. Proper sampling is essential because one aflatoxin-contaminated kernel in 1,000 kernels of grain may be a source of significant contamination. Occasionally, a biased sample may be more revealing than a truly representative one. A 10-pound (5-kilogram) sample is usually collected by pooling 5 or more probes collected from an auger discharge of one or more combine hopper loads. Continue sampling as weather and harvesting conditions change. The sample(s) is then ground finely as coffee so that it will pass through a screen of 15 to 20 mesh and be thoroughly blended to obtain a subsample appropriate for analysis. Samples stored for subsequent analysis should be kept under cool, dry conditions that will not permit fungal growth and a possible production of mycotoxin or mycotoxins.

#### DETECTING AFLATOXIN

The methods of aflatoxin analysis fall into three categories (Table 5): (1) visual inspection of the grain, which may locate lots presumed to be contaminated with aflatoxin (blacklight test); (2) rapid screening procedures to determine the presence or absence of aflatoxin (antibody or ELISA immunoassay kits, and minicolumn tests); and (3) laboratory procedures quantifying the **actual** amounts of toxin present (thin-layer chromatography, gas-liquid chromatography, high-pressure liquid chromatography tests or approved

ELISA immunoassay kits). Various commercial, state, and federal laboratories perform aflatoxin analyses on a fee basis. Several confirmation tests are available for identifying the presence of aflatoxins.

#### MINIMIZING MYCOTIXIN PRODUCTION IN CORN, SMALL GRAINS, AND SOYBEANS AFTER HARVEST.

1. **Harvest drought-stressed and insect-infested grain at early maturity as soon as the moisture content allows minimum grain damage:** for shelled corn (23 to 25 percent moisture), ear corn (25 to 30 percent), small grains, including sorghum (12 to 17 percent), and soybeans (11 to 15 percent). Unfortunately, exact timing is not always possible because of unfavorable harvesting conditions.
2. **Adjust the combine header speed to minimize cracking and reduce the content of trash, fines, and small broken or mold-infected kernels, especially those kernels near the tips where mold infestation is most likely to be present.** Upward of 50 percent reduction in existing aflatoxin levels (or below 20 ppb) can be achieved in some fields by careful monitoring of combine cylinder, screen and air flow levels.
3. **Dry all grain to at least 13- to 14-percent moisture as rapidly as possible, not to exceed a 24- to 48-hour period after harvest to prevent production of aflatoxins.** Safe, long-term storage (9 months or longer) can be achieved at a **uniform moisture level of 13 percent** or somewhat below. Moisture may be 14 percent if grain is to be moved or sold in a shorter period of time. Slow drying (accomplished by low heat or high-volume ambient dry air drying) is being used increasingly, but the grain should contain no more than 18 to 20 percent moisture in full-bin drying. Another possibility is high-temperature drying until the grain reaches 18 to 20 percent moisture, followed by low-heat drying to 13 percent moisture. Avoid air drying of mold-damaged corn without heat. Research has shown aflatoxin increases of 100 to more than 2000 ppb in three days when recently harvested field corn was stored at high moisture levels. Delays in transit to the storage bin or buying point should be minimal. Aflatoxins have been shown to increase in truckloads of contaminated corn by as much as 6 percent per hour of delay.

#### Maximum storage time in months for shelled corn\*

	Corn moisture content					
Corn	13%	14%	15%	16%	17%	18%
Temperature						
40°F	150	61	29.0	15.0	9.4	6.1
50	84	34	16.0	8.9	5.3	3.4
60	47	19	9.2	5.0	3.0	1.9
70	26	11	5.2	2.8	1.7	1.1
80	15	6	2.9	1.6	0.9	.9

\*Based on 0.5% maximum dry matter loss-calculated on the basis of USDA research at Iowa State University.

4. **Cool the grain after drying and maintain dry storage conditions.** When possible, continue cooling until the grain temperature reaches 35° to 40°F (2° to 4°C).
5. **Thoroughly screen and clean the grain and all bins before storage** to remove dirt, dust, and other foreign matter, crop debris, chaff, and cracked or broken seeds and kernels. Most of the contaminated corn is in the small and broken kernels, which will drop through a screen.

6. **Store in water-, insect-, and rodent-tight structures.**
7. **Continue periodic aeration and probing for “hot spots”** at intervals of 1 to 4 weeks throughout the storage period.
8. **Producers can ensile high-moisture corn or treat it with propionic acid as registered on high-moisture grain in storage.** This is an option to increase the safety of holding high-moisture grain which will eventually have to be dried to a safe level for long-term storage. Propionic acid is sold under various trade names. Formulations containing 80 to 100 percent propionic acid are very effective against grain spoilage for a period up to one year. Follow label rates and use an application method that will uniformly distribute the acid over all the grain. Although this acid will not remove any aflatoxins already present in the grain, it will prevent the growth of fungi when properly applied. The corn, however, should be regularly checked for signs of heating, crusting, rise in the moisture level and development of mold growth. **Grains treated with propionic acid can be used only for livestock and poultry feeds.**
9. **Choose regionally adapted varieties resistant to insects, diseases and mechanical damage.** Choosing resistant varieties will help decrease the potential entry of *A. flavus* and other toxin-forming fungi and to toxin formation once the fungus or fungi has invaded the grain.
10. **Where feasible, irrigate thoroughly** (soil moist 6 inches or more deep) **during hot dry periods to avoid moisture stress during the critical grain filling period.**