

Aflatoxins

Hazards in Grain / Aflatoxicosis and Livestock

Figure 1: *Aspergillus flavus* mold growing on an unfilled ear tip.

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Figure 2: Moldy corn grain. The yellow mold is *Aspergillus flavus*, a leading source of aflatoxin. Many molds may grow in corn and different mycotoxins are associated with each mold.

Many different fungi may grow as molds on stored grains. *Fusarium* and *Aspergillus* fungi (Figs. 1 and 2) are among the most common grain molds. Not all fungi produce toxins, but *Aspergillus*, which produces aflatoxin, is among the most common grain mold fungi. Aflatoxins are poisonous, carcinogenic byproducts produced during the growth of several species of the mold fungus *Aspergillus*. These byproducts are produced as the fungi grow in feed grains, processed feed, and food products.

In South Dakota, aflatoxins are primarily a problem in corn but can also occur in other grain crops. Aflatoxins are highly toxic to livestock, poultry, and people. Consumption of low concentrations by animals sensitive to aflatoxins can lead to death in 72 hours. In general, at nonfatal levels, the health and productivity of animals fed contaminated feed are seriously impaired. As a result, the Food and Drug Administration (FDA) has set an action level for aflatoxins in corn at 20 parts per billion (ppb). Corn containing aflatoxin levels of 20 ppb or more cannot be sold in interstate commerce, and, in general, should not be fed to young poultry, swine, and livestock, or to lactating animals, and must not be milled for human consumption.

Understanding *Aspergillus* and Aflatoxin Contamination

Development in the Field

The development of aflatoxins depends on the infestation and growth of the *Aspergillus* mold in grain. High temperatures and high humidity favor the infection of corn kernels through the silks by the *Aspergillus* fungi. In the southeastern United States and the eastern Corn Belt this environmental condition occurs more frequently and is the main reason *Aspergillus* infections and aflatoxin contamination occur more frequently.

Below-normal soil moisture (drought stress) has also been found to increase the number of *Aspergillus* spores in the air. Therefore, when drought stress occurs during pollination, the increased inoculum load (spores in the air) greatly increases the chances of infection. Furthermore, drought stress, nitrogen stress, and other stresses that affect plant growth during pollination can increase the level of aflatoxins produced by *Aspergillus* fungi. Often, *Aspergillus* will grow on the unfilled portions of the ear (Fig. 1).

In the past, insect injury to the maturing ear of corn was considered a requirement for infection to occur. This is now known to be false. However, insect damage to ears provides wounds that allow *Aspergillus* to more readily infect the kernels. Insects also transport *Aspergillus* spores to the silks and the kernels. Therefore, insect damage, especially during pollination in drought-stressed corn, can increase the occurrence of *Aspergillus* and the levels of aflatoxins.

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Several other factors play a role in the development of *Aspergillus* mold and aflatoxin production. Because drought stress plays such an important role, practices that reduce drought stress in plants should reduce the levels of infection and aflatoxin production.

Irrigation has been shown to be very effective in reducing *Aspergillus* infection and aflatoxin development, even if done only during pollination. Tillage practices have not been as effective and have only been demonstrated to reduce aflatoxin by subsoiling in areas with hardpans. Occasionally during droughty periods, hybrids of differing maturities or those planted early will pollinate during periods when drought stress is less often observed in South Dakota. Escaping drought with planting dates and hybrid maturity may differ from one year to the next.

Time of harvest has also been shown to be important in influencing the occurrence and levels of aflatoxin because *Aspergillus* does not compete well with other molds when corn is above the 20 percent moisture content. Harvesting corn when moisture content is above 20 percent followed by rapid drying to at least a moisture content of 14 percent within 24 to 48 hours of harvest keeps further *Aspergillus* growth and toxin production at a minimum.

Aflatoxin Development in Storage

Mature corn that remains in the field or corn that is stored without adequate drying can be subject to *Aspergillus* growth and aflatoxin production. Temperatures between 80F and 100F and relative humidity of 85 percent (corresponding to 18 percent grain moisture) are optimum for growth of *Aspergillus*. Growth of the fungus is poor below 55F, but if the grain is moist enough, toxins can still be produced. However, simply reducing the moisture content to as low as 12 percent does not kill the fungus and does not reduce the levels of toxins that have already been produced. If moisture levels rise again above 12 percent anytime during storage, and temperatures are high enough, then mold growth and toxin production will resume.

It is important to note that conditions favoring the growth of *Aspergillus* also favor the growth of other fungi that can have harmful effects on humans if they are inhaled or ingested while working in grain handling facilities. Always wear a dust respirator when working in grain or feed storage and handling areas.

Detecting Aflatoxin Contamination

Once aflatoxin is produced, it is stable. Heat, cold and light do not affect it. It is also colorless, odorless and tasteless, and because of the low concentrations involved and the uneven distribution in grain bins, aflatoxins are difficult to detect.

In the past, elevator operators and buyers used the blacklight test (Fig. 3), but this test simply detects compounds that fluoresce (aflatoxins and others) and should only be used to select samples that require further testing. Similarly, minicolumn tests are no longer recommended, as they were prone to give false positive results if used improperly.

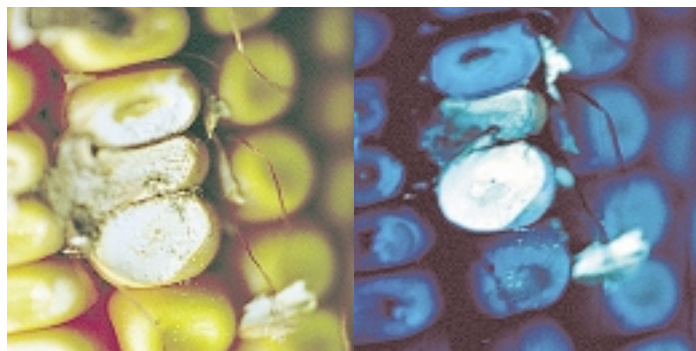


Figure 3: Fluorescence of grain under ultraviolet light is an indicator of possible aflatoxin contamination, but does not confirm the presence of aflatoxin. Photo reproduced, with permission, from APS Digital Image Collection: Diseases of Cereal Grains, 2000, The American Phytopathological Society, St. Paul, MN.

Serological tests are now considered to be more reliable and their accuracy has been validated by comparison to more costly and time-consuming analytical procedures. Serological test kits using such methods as ELISA (Fig. 4) do not require specialized labs, equipment, or training and when conducted according to manufacturer's instructions can give accurate results for the presence (qualification) and amount (quantification) of aflatoxin in grain samples.

Figure 4: An ELISA test can detect very small quantities of specific contaminants in grain. The development of color (blue in this test) indicate the presence of the toxin. Intensity of the color can indicate the amount of toxin present. Photo: M. A. Draper.



Sampling for Aflatoxins

Regardless of the test procedure used, the single most important factor for reliable and accurate testing of grain for aflatoxins is obtaining a representative sample. The ideal sample size should be at least 10 pounds of corn. The sample should consist of several smaller subsamples (10 or more 1-pound samples) that have been taken from different spots and then mixed together. Handle each bin or truck separately, and take a 10-pound sample from each source.

Place samples in a cloth or paper container that allows air exchange. Air-tight containers or plastic bags allow condensation, which raises the moisture content, resulting in the possibility of continued growth and toxin production of the fungus.

Send or take samples to a testing lab as quickly as possible. Monday through Wednesday is the preferred time of week to ship samples for testing. Samples mailed later in the week are more likely to be delayed in transit. This gives the fungus a chance to continue to grow and produce toxins. Samples should be properly identified and include: the source of the sample (truck or bin), the sender of the sample, full address, and telephone number.

Preventing Aflatoxin Contamination

Resistance to aflatoxin accumulation in corn kernels has been recently identified. Hybrids resistant to aflatoxin and other fungal toxins should become available in the near future.

To minimize aflatoxin contamination of corn products:

- **Reduce plant stress.**

Use recommended production practices to minimize plant stress and maximize yields. These include insect, weed, and disease control practices, and the use of recommended plant populations and fertility practices. When possible, irrigation during pollination can decrease predisposition of the crop to aflatoxin. Plant corn as early as possible, and plant several different hybrids of different pollination periods to reduce the chance of environmental stress at pollination in at least part of the crop. Care should be taken to store uncontaminated hybrids away from those that are contaminated.

- **Harvest corn early and dry it immediately.**

Harvesting corn when it is above the 20 percent moisture content and drying it within 24 to 48 hours to a moisture content no greater than 14 percent greatly reduces the infection, growth, and toxin production by *Aspergillus*.

- **Avoid damaged kernels.**

Damaged kernels are more likely to become infected with molds both in the field and in storage. Corn hybrid selection and insect control can play a role in reducing kernel damage. Corn hybrids with good husk coverage of the ear have been shown to have less infection and aflatoxin development. Also, *Bt* corn hybrids derived from transformational events that express the *Bt* trait in the ears and the silks are less likely to become infected with toxin-producing fungi as a result of reduced kernel damage. Adjustment of the combine to reduce mechanical damage of the kernels at harvest is a very important means of reducing contamination in storage.

- **Store corn at 12 percent moisture content.**

Maximum moisture content for corn storage should be 14 percent. Moisture content at or below 12 percent is ideal for storage of corn, because growth and toxin production by *Aspergillus* cannot occur.

- **Keep storage and feeding facilities clean.**

Aspergillus fungi can survive on residues left in storage areas. When environmental conditions become favorable, infection and toxin production can reoccur year after year in storage systems that are not properly cleaned and disinfected.

Using Contaminated Corn

Recommended levels are 0 ppb aflatoxins in feed. However, aflatoxin-contaminated feed can be tolerated by some livestock, particularly older animals. Obviously, the higher the level of contamination, the greater the risk in feeding contaminated corn

to animals. Furthermore, continued proper storage is essential so that aflatoxin levels do not continue to increase in the corn or feed before use.

Detoxification of feed continues to be an elusive goal. However, certain feed additives have been successfully used to inhibit mold growth and to reduce the incidence of aflatoxicosis in animals. Organic acids such as propionic, sorbic, and benzoic acids as well as their salts such as calcium propionate and potassium sorbate, and copper sulfate can be used to inhibit mold growth in feed. Mineral clays such as zeolite and bentonite as well as hydrated sodium calcium aluminosilicate (HSCAS) can bind to aflatoxin, protecting animals from absorbing the toxin that may be in the feed. These products, according to FDA rules, cannot as yet be labeled as mycotoxin binders, and are sold as anti-caking and free-flow feed additives.

There are no clear-cut safe levels for different animal species regarding their resistance or tolerance to aflatoxins. The following section on aflatoxicosis and ruminants and the general guidelines for dealing with aflatoxin-contaminated feed may assist you in deciding whether to assume the risk,

Aflatoxicosis and Livestock

Aflatoxicosis is a disease caused by the consumption of aflatoxins. Aflatoxins are secondary mold metabolites produced by some strains of *Aspergillus flavus* and other related species of *Aspergillus* fungi. The four most common aflatoxins are B1, B2, G1, and G2. Contaminated grains and grain byproducts are the most common sources of aflatoxin. Corn silage may also be a source of aflatoxins, because the ensiling process does not destroy toxins already present in silage.

Aflatoxins are metabolized in ruminants by the liver and are excreted in the bile. Aflatoxin B1 is the most potent mycotoxin (toxic substance produced by a mold) to affect cattle. B1 increases the apparent protein requirement of cattle and is a potent carcinogen (cancer causing agent). When significant quantities of B1 are consumed, the metabolite M1 appears in milk within 12 hours. Research suggests M1 is not as carcinogenic or mutagenic as B1, but it does appear to be as toxic as its parent compound.

Symptoms

Beef and dairy cattle are more susceptible to aflatoxicosis than sheep or horses, although other mycotoxicoses occur in these species, such as facial eczema in sheep and leukoencephalomalacia in horses. Young animals of all species are more susceptible than mature animals to the effects of aflatoxin. Pregnant and growing animals are less susceptible than young animals, but more susceptible than mature animals.

Feed refusal, reduced growth rate and decreased feed efficiency are the predominant signs of chronic aflatoxin poisoning. In addition, listlessness, weight loss, rough hair coat and mild diarrhea may occur. Anemia along with bruises and subcutaneous

hemorrhage are also symptoms of aflatoxicosis. The disease may also impair reproductive efficiency, including abnormal estrous cycles (too short and too long) and abortions. Other symptoms include impaired immune system response, increased susceptibility to disease, and rectal prolapse.

Pathology

Clinical laboratory findings vary with the animal species, level of aflatoxin in the ration, and the duration of feeding. There are no consistent diagnostic changes in hematocrit, hemoglobin, and differential cell counts in animals fed aflatoxin.

Leukocytosis may occur in animals with secondary bacterial infections. Serum bilirubin levels may be elevated and typically serum protein levels are decreased.

Lesions observed at necropsy related to either acute or chronic liver disease are dependent upon the level of aflatoxin and the duration of feeding. A majority of acute liver damage observed has been the result of experimentally high doses, while chronic liver damage is a more common field observation. The liver is usually pale tan, yellow or orange. Hepatic fibrosis and edema of the gallbladder may also be observed.

Diagnosis

The diagnosis of aflatoxicosis is often difficult because of the variation in clinical signs, gross pathological conditions and the presence of infectious diseases due to the suppression of the immune system. On the farm, more than one mold or toxin may be present in the contaminated feed, which often makes definitive diagnosis of aflatoxicosis difficult.

The prognosis of aflatoxicosis depends upon the severity of liver damage. Once overt symptoms are noticed the prognosis is poor. Treatment should be directed at the severely affected animals in the herd and further poisoning prevented.

Treatment

Aflatoxicosis is typically a herd rather than an individual cow problem. If aflatoxicosis is suspected, the ration should be analyzed immediately. If aflatoxins are present, the source should be eliminated immediately. Levels of protein in the ration and vitamins A,D,E,K and B should be increased as the toxin binds vitamins and affects protein synthesis. Good management practices to alleviate stress are essential to reduce the risk of secondary infections. Secondary infections must receive immediate attention and treatment.

Prevention

Aflatoxicosis can only be prevented by feeding rations free of aflatoxin. Preventing aflatoxin contamination is outlined on the preceding page, but since preventing contamination is not always possible, here are a few **keys facts to remember when dealing with contaminated feeds in animal rations:**

- The recommended feeding level is 0 parts per billion (ppb).
- The level of aflatoxin an animal can tolerate will depend upon the age and sex of the animal, its health status, and overall management level of the farm.
- To avoid contamination of milk, lactating dairy cattle should not receive more than 20 ppb in the total ration.
- Calves should not receive milk from cows fed in excess of 20 ppb, because they can ingest aflatoxin from the milk.
- Beef cattle can tolerate slightly higher levels of aflatoxin, but yearlings and mature cows should not receive more than 400 ppb in the total ration. Weanlings should not receive more than 100 ppb in their total daily ration.
- Poultry and swine are more sensitive to aflatoxin contamination. **Under no circumstances should these livestock species be fed more than 20 ppb aflatoxin in their daily rations.**

The above are only guidelines. This does not suggest that feeding at these levels or below will reduce or eliminate the potential for aflatoxicosis. There are no clear cut safe feeding levels. Safe levels vary with each individual animal. Remember that ingestion of aflatoxins at levels even lower than those listed in the guidelines may cause some undesirable side effects and depends on such factors as age, sex, and general health of the animals. To feed at a level other than 0 ppb is a risk assumed by the person making the decision to do so. In all cases, monitor animal health closely and discontinue the use of contaminated feed immediately if undesirable effects are noticed.

Conclusion

Aflatoxins are highly toxic to livestock, poultry, and people. Even when fed at nonfatal levels, aflatoxin can seriously impair animal health and productivity. For lactating dairy cattle, do not exceed 20 ppb aflatoxin in rations to avoid exceeding the Food and Drug Administration level of 0.5 ppb in milk. Aflatoxin is just one of many mycotoxins that can adversely affect animal health and productivity.

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